University of Agronomic Sciences
and Veterinary Medicine of Bucharest
Faculty of Agriculture

SCIENTIFIC PAPERS
SERIES A. AGRONOMY

Volume LXII, No. 1

2019
BUCHAREST

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PUBLISHER: University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Agriculture, Romania
Address: 59 Mărăști Blvd, District 1, 011464, Bucharest, Romania
Phone/Fax: + 40 213 318-0466; E-mail: journal@agro-bucuresti.ro
Webpage: http://agronomyjournal.usamv.ro

To be cited: Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019

ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

International Database Indexing: Web of Science Core Collection (Emerging Sources Citation Index), CABI, Index Copernicus, Google Scholar, CNCSIS B+, Ulrich's Periodicals Directory, Research Bible, Scipio, Scientific Indexing Service, PBN (Polish Scholarly Bibliography), OCLC (WorldCat)

SUMMARY

SOIL SCIENCES

CROP SCIENCES

MISCELLANEOUS

SOIL
SCIENCES

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EVALUATION OF QUALITY AMENDMENTS IN ORDINRY CHERNOZEM AFTER INCORPORATION IN THE SOIL A HARVEST OF NTERMEDIATE CULTURE THE VETCH AS A GREEN MASS

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Abstract

Agriculture in the Republic of Moldova accounts for 10-15% of gross domestic product (GDP) and represents the most important branch of production. In agriculture, soil is the main means of production. Agriculture currently practiced in the country faces a number of major problems that seriously affect rural development. Creating a balanced or positive balance of the organic substance in the soil is the main condition for preserving its long-term fertility and avoiding degradation of the arable layer by dehumidification, destructuring and excessive secondary compacting. This can only be achieved by the regular introduction into the soil of organic fertilizers - manure or green fertilizers (Cerbari, 2010; Cerbari et al., 2013). At present the livestock sector is practically destroyed, the number of cattle has decreased 6 times, while the volume of manure produced has decreased by 6 times. According to the statistical directories 3 million tons of manure is produced, of which 1.0-1.5 million tons are used as an energy source, and 1.5-2.0 million tons are dumped in the rubbish, a more serious source of pollution of rural areas. In the soil, according to statistical data, only 10-20 kg/ha/year of manure is introduced, which means nothing. As a result, the balance of humus in the soil became deeply negative -1.0 t/ha/year. The need for this article is dictated by the fact that the recommended procedure has the only real possibility to increase the flow of organic matter to the soil and to form a balanced or weakly positive balance for this organo-mineral natural system that plays the most important role in ensuring the food and environmental security of the country. The situation can only be changed by undertaking a series of legislative, organizational and financial measures. So, the remediation of the quality condition and the increased production capacity of the studied soil is possible only by increasing the flow of organic matter into the arable layer. The use of vetch as a green fertilizer is an effective process to achieve this objective (Cerbari et al., 2013).

Key words*: conservative agriculture, ordinary chernozem, green fertilizer, quality status.*

INTRODUCTION

Soil is an organo-mineral system that can provide a high agricultural production capacity only if it has a permanent flow of fresh organic matter. Creating a balanced or positive balance of the organic substance in the soil is the main condition for preserving its long-term fertility and avoiding degradation of the arable layer by dehumidification, destructuring and excessive secondary compacting. This can only be achieved by the regular introduction into the soil of organic fertilizers - manure or fertilizers (Capcelea, 1996; Cerbari 2011).

In Moldova, the situation regarding the use of organic fertilizers is the following. In order to ensure a balanced or positive balance of the organic substance it is necessary to introduce at least 10 t/ha/year of manure into the soil. In the 90^s of the previous century, due to the existing zootechnical sector, about 7-8 t/ha/year of manure was introduced into the arable soil. Combined with respect for crop rotation in which there is a field of leguminous perennial herbs and a field of annual grasses with the participation of leguminous crops, a balanced balance of the organic substance in the soil is ensured.

At present the livestock sector is practically destroyed, the number of cattle has decreased 6 times, while the volume of manure produced has decreased by 6 times.

According to the statistical directories 3 million tons of manure are produced, of which 1.0-1.5 million tons are used as an energy source, and 1.5-2.0 million tons are dumped in the rubbish, a more serious source of pollution of rural areas. In the soil, according to statistical data, only 10-20 kg/ha/year of manure is introduced, which means nothing. As a result, the balance of humus in the soil became deeply negative: -1.0 t/ha/year. Agriculture currently practiced in the country faces a number of major problems that seriously affect rural development. The classical system of soil works has led to a gradual increase in production, but it has also led to the appearance of phenomena of degradation of features, a decrease in the production capacity of agricultural land.

Excessive work has favoured processes to reduce organic soil content, damage to structure, increased erosion hazard; heavy traffic and too often led to increased compaction and, as a result, to trigger other negative phenomena (Cerbari et al., 2013).

MATERIALS AND METHODS

The polygon for the study of the influence of green vines on the properties of common chernozems in Southern Moldova was located on the agricultural territory of Lebedenco in the district of Cahul (Figure 1).

The field works were carried out according to the methodology of pedological field research. Laboratory analyzes were performed according to classical methods and STAS [STAS 17.4.4.02, STAS 28298-89] and Standards [SR SR 7184-3: 2003, SM SR ISO 11272: 2003, SM STAS 7184/16: 2003, SM STAS 8619/3: 2002] existing. The polygon (3 ha) is a land area located on the quasi-horizontal surface of a long elongated ridge. The mixture of vines (80%) and wheat (20%) was sowed on the surface of 3 hectares of heights (Figure 1) in the first decade of September. On April 25, the field was divided into 3 strips with a surface of about 1 ha each (Figure 2): Strip no. 1 - vines cultivated for the production of seed material; Strip no. 2 - vines, busy field, two crops grown and incorporated into the soil as green fertilizer; Strip no. 3 - intermediate culture for use as green fertilizer; after the incorporation into the soil of the meadows in the field sown corn.

Parallel to the varieties of varieties of different destination, in the eastern part of the field a 10-meter-wide blank was sowed with maize to compare the corn harvest on this strip with corn harvest on strip no. 3, sowed with corn after incorporation into the soil of a harvest of vines crop (Figure 2).

RESULTS AND DISCUSSIONS

The necessity of this article is dictated by the fact that the recommended method presents the only real possibility to increase the flow of organic matter to the soil and to form a balanced or poorly positive balance for this organo-mineral natural system that plays the most important role in ensuring the food and environmental security of the country.

The high cost of chemical fertilizers and their negative effects on the environment have convinced the human community to seek an alternative to the existing farming system (Cerbari, 2010; 2011; Cojocaru, 2018; Toncea, 1999).

Figure 1. The location of the research polygon

The physical, chemical and biological degradation of chernozems, soils occupying about 80% of the land surface of the agricultural land, leads to the expansion of the desertification processes of the land and to stop the increase of the agricultural production volume in the country. The existing agricultural system does not ensure that soil quality is maintained in the future and leads to worsening economic and environmental conditions in the country (Cerbari et al., 2013).

	Strip No. 1, length 200 m, width 50 m	Strip No. 2, length 200 m, width 40 m	Strip No. 3, length 200 m, width 60 m	Strip No. 4, length 200 m, width 10 m
 Tractor brigade	 Vetch sown for the production of seed material	 Vetch, busy field, 2 crops grown and incorporated into the soil as green fertilizer	 Vetch, intermediate culture incorporated into the soil as fertilizer green, basic culture after vetch - maize	 Control, sown with maize
The central road				

Figure 2. Scheme of experience located on the territory of Lebedenco commune, Cahul district

The overcoming of this situation is possible through the gradual implementation of a sustainable agriculture system, based primarily on the use of natural processes, on the biological and renewable resources of the household and only secondly on the resources procured. The situation can only be changed by undertaking a series of legislative, organizational and financial measures. This serves as a motivation for the fact that in order to reduce the dehumification processes, damage to the structure, strong compaction of the arable soil layer and increase of the soil erosion resistance, it is recommended 5 field crop, a field occupied with a leguminous sidney crop, autumn and spring varieties (2 varieties of vetches incorporated into the soil as a green fertilizer on each field once every 5 years). The structure of the crop can be as follows: Field covered with vines → Maize → Autumn wheat → Autumn wheat or barley → Sunflower. This process, used in the framework of any agricultural system, will lead to the remediation of soil quality and the increase of its production capacity (Capcelea, 1996; Cerbari, 2010; 2011; Cojocaru et al., 2018).

The research envisaged the creation of a single polygon to study the processes of remediation of the quality of common chernozems in Southern Moldova under the influence of green fertilizers, but in 2013 the possibility was created for organizing the second unpredicted research polygon. This polygon was organized in Central Moldova on the territory of Ivancea commune, Orhei rayon for cambic chernozems - soils with physical characteristics more difficult than those of ordinary chernozems in Southern Moldova, formed under other conditions.

The state of the crop on the sown field with vetches on 25.04.2013 is presented in Figure 3 and Figure 4. The introduction of ground vermicelli in the soil and the preparation of the ground for sowing was done by talking with a disk with a disk (Figures 5 and 6). Sowing maize (basic crop) was done on April 29, 2013.

Figure 3. Field sown in autumn with vetch on April 25, 2013

On both polygons, some preliminary results on the influence of green vetches on the quality of soils and on the size of the basic crop crops were obtained.

The study of the initial soil quality of the soil of the research polygon was carried out between september 4-5 of 2012, until the basic soil work was carried out. After performing the basic soil work by discussing at a depth of about 12 cm and subsoiling at a depth of 35 cm with the combined aggregate (Figure 5) and sowing the meadows (10.09.2012), over 10 days, the apparent density of the 0-35 cm, loosened through the underlay. Of all semi-graphs located on the territory of the polygon, soil samples were collected for laboratory analysis.

Figure 4. The state of the vetch at 25.05. 2013 and the distribution of the crop roots on the soil profile as a result of using the Mini-Till system with its underlayment

Figure 5. Incorporating the green mass of vetch by disking

In the spring of 2013, in the centre of the polygon, where the semi-profile 102 was 50 cm deep, repeated until the incorporation of the vetch vegetable mass into the soil, a soil profile with a depth of 120 cm was placed.

Figure 6. Field prepared by disking for sowing the basic crop after incorporation into the soil of the green vetch mass

In the soil samples from this profile, more detailed researches of the properties of the studied soil were carried out. At the same time in harvested soil micromonolites the content of the roots of vetches was appreciated, and in the laboratory - the chemical composition of roots and green vetch.

Field research to assess the size of the crop of basic crops (maize) and changes in soil quality on line no. 3 (broth culture) were performed on 26.09. 2013.

The basic soil work was carried out with a combined aggregate (Maximulch aggregate, Agroisem International), which passes the soil at a depth of 35-40 cm and prepares the top of the loose sowing layer (Figure 7).

Figure 7. Technique for Implementing the Mini-Till Soil System in Combination with subsoiling to work the land polygon basic of research

The initial state of the attributes of the common chernozem on the South Moldovan polygon was assessed by field research of the morphological characters of the 4 semiprofiles and the soil profile 102 (located on the 2nd line), harvesting the soil samples and analyzing them in laboratory. Profile picture 102 is shown in Figure 4.

Next, we describe the profile of the studied soil.

Ordinary arable chernozem is characterized by a profile of the type: **Ahp1 - Ahp2 - Ah - Bhk1 - Bhk2 - BCk1 - BCk2 - Ck**. Efervescence - from a depth of 47 cm. Carbonates in the form of efflorescences - from 48 cm, pseudomics - from 63 cm. The following genetic horizons were highlighted and studied on the soil profile.

Ahp1 (0-20 cm) - arable layer of dark gray to brown color weak, wet, clay-loam, glomerular-bulgarian structure, layer 0-12 cm tilted by discussion, layer 12-20 cm inhomogeneous loosened by subsoiling, medium and large pores very frequent, many roots of vines and organic scraps, gradual passage.

Ahp2 (20-35 cm) - the newly built compact partially sloping layer (60-70%) by underlay, dark gray with a dull brown, wet, clay-loam, glomerular-bulgarian structure, porosity, different pores, many roots of vetch in the loose spaces of this layer, clear passage.

Ah (35-47 cm) - substrate to the arable layer, continuation of humus accumulation horizon, wet, dark gray with light brown, luteum-clay, glomerular natural structure, compact, medium and small frequent pores, frequent thin and medium roots, larvae, coprolite, passage gradually on the next horizon.

Bhk1 (47-70 cm) - continuation of the humerus profile, beginning of the passage to the parental rocks, wet, dark brown, clay-clay, glomerular structure, aggregates of 1-3 cm, compact, small and medium-sized pores, thin roots, larvae, coprolite, of carbonates in the form of efflorescences, gradual passage.

Bhk2 (63-81 cm) - the humerus profile, the lower part of the horizon to the rocks, the brown, the clay-clay, the weakly structure, the small, compact, small and fine pores, the rare thin roots, the larva beds, the moderate effervescence, in the form of efflorescences, the transition to the next gradual horizon.

BCk1 (81-93 cm) - the upper part of the parental soil poorly modified by the pedogenesis process, yellow with brown hue, loam-clayey, very weak structure, compact, frequent fine pores, pseudomics carbonate, very thin roots, larva beds, switching to the next gradual horizon.

BCk2 (93-110 cm) - the lower part of the parental soil poorly modified by the pedogenesis process, yellow with a very low brown hue, loam-clayey, the massive compact structure, frequent fine pores, the accumulation horizon of the pseudomics and bieloglass carbonate neoformations, the concrete and crotovine the transition to the next gradual horizon.

Ck (>110 cm) - yellow parental rocks virtually unchanged by the pedogenesis process, rare pseudomics, concretes of $CaCO_3$, compact, easily crushed, frequent fine pores.

The soil of the research polygon is characterized by very favorable lute-clayey texture. The dust behaves in the soil as well as the microstructural elements, ensuring a more favorable physical condition of the soil (lower resistance to plowing, lower cohesion between the soil particles, maturity humidity ensures better shredding at the work of the ground).

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The values of the chemical properties of the studied soil are typical for the common chernozems in South Moldova. As a negative factor, it is necessary to highlight the obvious dehumification of the arable layer of the common research chernozems, which has negative action on all the features and processes in the soil.

So, the remediation of the quality condition and the increased production capacity of the studied soil is possible only by increasing the flow of organic matter into the arable layer. The use of the meadow as a green fertilizer is an effective process for achieving this genre.

CONCLUSIONS

The estimation of the size of the green vetch mass harvest was carried out on 25.04.2013 on 1 m^2 surface micro-polygons in five repetitions.

The results of the individual measurements of the green vetch harvest were as follows: 1 - 30 t/ha; 2-29 t/ha; 3-28 t/ha; 4-29 t/ha; 5-32 t/ha. Average harvest of green vetch mass on 25.04. 2013 turned out to be 81.5% from the wet mass of the meadow. So, 5.6 t/ha of absolutely dry vetch meal with a nitrogen content of 4.1% was incorporated into the soil, which is equal to about 230 kg/ha of biological nitrogen, of which 60% is a symbiotic nitrogen (from the atmosphere).

The use of the meadow as a green fertilizer is an effective process for achieving this genre.

REFERENCES

Capcelea, A. (1996). *Republic of Moldova on the path to sustainable development*. Science Edition, Chisinau, 236 pp.

Cerbari, V. (2010). No-Till - an agricultural system that protects the soil. *Agricultural Journal of Moldova, 8-9*, 9–14.

Cerbari, V. (2011). The Program for Development and Implementation of Conservative Technologies in Agriculture. *Agricultural Journal of Moldova, 4-5*, 7–9.

Cerbari, V., Scorpan, V., Taranu, M., Bacean, I. (2013). Phyto-technology for Remediation of Chernozem in the South of Moldova. *Soil as World Heritage* (381–389). Ed. D. Dent, Springer Dordrecht Heidelberg New York London.

Cojocaru, O., Cerbari, V. (2018). Agriculture in the republic of Moldova in terms of soil remediation and protection. *AgroLife Scientific Journal, 7*(1), 42–47.

Cojocaru, O. (2018). Researches on the efficacy of soil moisture release by range of aerospace areas and conservation of humidity area. *International Conference on Theoretical and Applied Economic Practices „Economic growth in Conditions of Globalization: Competitiveness, Innovation, Sustainability" the 13th Edition, October 11-12,* Chisinau, 293–300.

Toncea, I. (1999). *Organic farming in the context of sustainable agriculture//Sustainable agriculture.* Agris Publishing House, Bucharest, 101–128.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE INFLUENCE OF THE TILLAGE SYSTEM AND THE CROP ROTATION ON THE SOIL AVAILABILITY IN THE MAIN NUTRIENTS AND THE YIELDS OBTAINED IN TURDA AREA

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Abstract

The research on the application of different soil tillage systems enjoys considerable worldwide popularity, and this can be explained by the effect of soil tillage on the conservation of water resources of the soil, as well as on the production costs. Yet, the main objective of this kind of research is the identification of an appropriate balance between the effects of soil tillage systems on soil quality and on yield. After 12 years of experimentation at ARDS Turda, changes in soil pH, from low acid and neutral to low alkaline in all experimental variants. In the case of the classical system (CS) there is a slight reduction of the humus content in the surface layer of the soil on the first cm at both levels of fertilization but instead there are increases on the deeper layers in the variant with additional fertilization. In the MT system there is a slight increase of total N in the first cm in both fertilization variants. Increases content in phosphorus in the MT system, from a weak content to low content. There is an increase in the K content, especially in the arable layer in the MT system compared to CS. The average yields recorded at crops had lower values especially in the conservative system on the first years.

Key words*: tillage system, crop rotation, nutrients, clime condition, yield.*

INTRODUCTION

Conservative tillage restores the soil structure and improves global soil drainage, resulting in a more productive soil, protected against water and wind erosion and requiring less fuel for the preparation of the germinating bed (Cociu, 2011; Cheţan et al., 2011; 2016). Poor rotation on crops can favor soil compaction by limiting the plants root system and the major role it plays in restoring physical features of the soil and even breaking the deep compacted layers. Ciontu et al. (2012) found that the three - four years rotation play an important role in achieving safe and stable yields and Šeremeši et al. (2013) found that the effect of crop rotation on maize yield was inversely proportional to the ratio of the maize in the crop rotation.

When mineral nitrogen fertilizers are applied, it is necessary to consider not only the production of superior quantities in terms of quantitative and quality, but also the evolution of the physical, chemical and biological characteristics of the soil. Comparing the "minimum tillage" and "no-tillage" systems with the conventional tillage, which involves the ploughing and/or the disc harrowing for the preparation of the seedbed, it can be concluded that there is a reduction in the content of organic material, mainly under tropical and subtropical conditions (Balota et al., 2003).

MATERIALS AND METHODS

The experiments were conducted 2007-2018 at ARDS Turda, located at 23°47′longitude and 46°35′ latitude on 345-493 m altitude, in West of the Transylvanian Plain, Turda town.

The relief is represented by a hilly orthographic framework, with a dominant proportion of 71% and specific with low altitude of 345-493 m, with different exhibitions and inclinations and an advanced erosion stage. The valleys between these hills, representing 11%, are relatively narrow, oriented mainly in the East-West direction and have a poor natural drainage. The groundwater is at different depths, depending on the relief, reaching 1.5-2 m on the valleys, 15-20 m on the plateaus and 0-18 m on the slopes. The experiment was designed as a randomized complete block. The experience was based on a Phaeozem (SRTS, 2012), with clay-clay texture with good hydrophobic properties, 59% porosity on the surface and 47% in depth, high water retention capacity of 32% and Co 18%.

The experimental factors are:

- *factor (A)* the soil tillage system with three graduations: a_1- conventional system (CS); a_2 - minimum tillage system (MT); a_3- no-tillage system (NT);
- *factor (B)* the fertilization level with two graduations: b_1- basic fertilization with $N_{40}P_{40}$ (at same with sowing); b_2- basic fertilization with $N_{40}P_{40}$ + N_{40} on vegetation (at wheat on resumption of vegetation in spring; soybean at 3-5 trifoliate leaves, maize at 4-6 leaves);
- *factor (C)* the agricultural year with 12 graduations: c_1 - 2007, c_2 - 2008, c_3 - 2009, c_4 - 2010, c_5 - 2011, c_6 - 2012, c_7 - 2013, c_8 - 2014, c_9 - 2015, c_{10} - 2016, c_{11} - 2017, c_{12} - 2018.

The soil system were included three variants of the processing of the land: classic with plow; minimum tillage and no-tillage. The wheat was cultivated in a classic system in parallel with the no tillage system; soy and maize in a classic system in parallel to the minimum tillage system.

In this experiment (three years rotation with winter wheat - maize - soy) in the first year it was sown directly in the stubble of the previously cultivated plant, in this case winter wheat after soy, two years after the soil was processed with the chisel.

At experimental factor B - fertilization, there are two graduation of application to all three crops (winter wheat - maize - soybean), only the application moment differs.

The sowing was done with the Directa-400 machine (at the same time as sowing was applied the fertilization) at 18 cm distance between the rows, the seed introduced at 5 cm depth, winter wheat density 550 b.g./m^2 and soybean 55 b.g./m^2. In the maize culture, sowing was performed with MT-6 machine at distance between rows 70 cm, density 65,000 plants/ha, of 22.5 cm distance between seed/row, the seed incorporation 5 cm depth.

The soil samples for chemical analyses were collected along a depth of 0-20 and 20-40 cm before instalated the experiments (2007) and after harvesting the soybean culture (2018). The method used in 2007 for determining the pH was the Potentiometric method; the nitrogen content was measured through the Kjeldhal method; the phosphorous content was measured through the Colorimetric method, whereas the Flame Photometric method determined the content of potassium in the soil (Pedological and Agrochemical Studies Office, Cluj). After soybeans have been harvested in 2018 the availability of soil in nutrients was determined with the laboratory equipment compact photometer PF12-Plus (with Visocolor Eco test kit and Nanocolor teste tube) from the Agricultural Research and Development Station Turda.

The results obtained were statistically processed according to the method of analyzing the variant and establishing the lowest significant differences, LSD (5%, 1% and 0.1%) (ANOVA, 2015).

The weed control was based on the use of herbicides: in soybean and maize culture infested with weeds was applied treatments out in two stages: pre-emergence and post-emergence in both system using of the present products exist on the market that can be applied individually or in combination, depending on the weed spectrum present: pre-emergence for soybean: Sencor (*metribuzin* 600 g/l) 0.35 l/ha + Tender (960 g/l *S-metolachlor*) 1.5 l/ha and post-emergence: Pulsar 40 (40 g/l *imazamox*) in dose 1.0 l/ha (for dicotyledonous and some monocotyledonous weeds) + Agil 100 EC (100 g/*l propaquizafop*) in dose 1.5 l/ha for the *Agropyron repens* control (izolated present); pre-emergence for maize: Merlin Flexx (240 g/ha *isoxaflutol* 240 g/l and *ciprosulfamida* (safener) 0.4 l/ha + Tender (960 g/l *S-metolachlor*) 1.5 l/ha and post-emergence: Cerlit (*fluroxipir* 250 g/l) 1.0 l/ha for control of dicotyledonous weeds (especially *Rubus caesius*) + Astral 40SC (*nicosulfuron* 40 g/l) 1.5 l/ha for the monocotyledonous weeds; on vegetation at winter wheat culture: 0.15 l/ha Sekator Progres OD (*amidosulfuron* 100 g/l + *iodosulfuron-metyl-Na* 25 g/l + *mefenpyr dietyl* 250 g/l (safener) + 0.6 l/ha DMA 6 (660 g/l *acide* from 2.4% D *dimethyl amine salt*: 825 g/l 2.4 *D dimethyl amine salt*).

The weather conditions during the 12 years of trials (measured at Turda Meteorological Station, 23°47′ longitude; 46°35′ latitude; 427 m altitude) are shown in Figures 1 and 2.

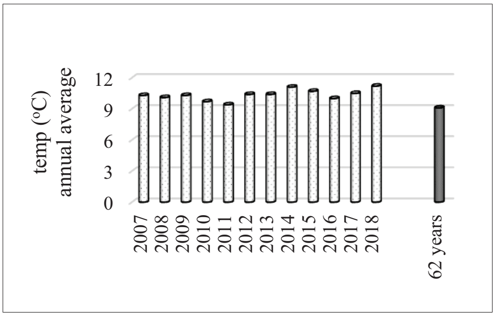

Figure 1. The thermic regime at Turda (°C), 2007-2018

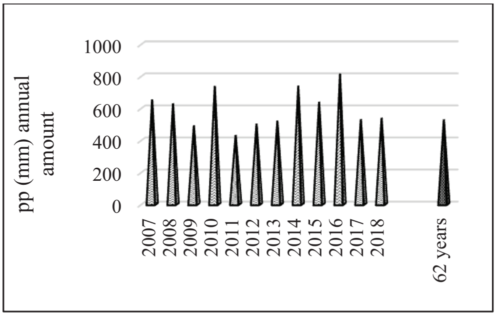

Figure 2. The rainfall regime at Turda (mm), 2007-2018

Of the 12 years considered in terms of the thermal regime, 6 were warm, 4 warm and only 2 normal and the precipitation was in 4 years excessive rain, 2 years very rainy, 5 years normal and just only 1 dry year 2011 with an annual rainfall of 433 mm). Annual mean values refer to multiannual averages (62 years) with average temperature 9.1°C and precipitation 531 mm. Also during this period the most rainy year was recorded in 2016 with 816.8 mm, the deviation is + 303.2 mm but with a non-uniform distribution of precipitation. The rainfall regime has increased in the Turda area in the last years, during the experimentation period, the most rain was 2010, 2014 and 2016. The average value of 609.8 mm, for the last 12 years, is maintained in the area with medium aggressiveness.

Specifically for the 12 years was the uneven distribution of precipitation (2009, 2011, 2012, 2017, 2018), drought prolonged was followed by torrential rains, which although they had large quantities of water, did not have often managed to restore the water reserve in the soil and the drought dominating this whole period. During the last years the climate in the Transylvanian Plain has changed, with the increase of the annual average temperature as well as the non-uniformity of rainfall, that is why the agrotechnique applied must be adapted to more oscillating ecological conditions.

REZULTS AND DISCUSSIONS

The soil reaction in the ARDS Turda area is neutral, on the ground where the experience was located, the soil reaction is weakly acidic in the first 20 cm and neutral on the depth of 20-40 cm. After 12 years, can be seen from Table 1, changes in soil pH, from weakly acid (0-20 cm) and neutral (20-40 cm) to low alkaline in all experimental variants and on both of them (7.7-8.4).

Table 1. The influence of the soil tillage system, crop-rotation and fertilization upon the soil pH (ARDS Turda 2007, 2018)

Year	Tillage system/ crop rotation/fertilization	Deepth sample	pH_{H2O}
2007	Classic $N_{40}P_{40}$	0-20 cm	6.30
		20-40 cm	7.00
2018	Classic $N_{40}P_{40}$	0-20 cm	7.7
		20-40 cm	8.4
	Minimum tillage $N_{40}P_{40}$	0-20 cm	7.6
		20-40 cm	8.3
	Classic $N_{40}P_{40+}N_{30}$	0-20 cm	7.8
		20-40 cm	7.8
	Minimum tillage $N_{40}P_{40+}N_{30}$	0-20 cm	7.5
		20-40 cm	8.3

The soil on the experiment have an average supply of N (0.162) in the 0-20 cm layer and low in the 20-40 cm (0.124) layer. Comparing the N content from 0-20 cm to 0.162% in 2007, 0.147 % came in 2018 by talking about SC + a fertilization and 0.150% on the same level with the supplementary fertilization. In the deeper layers of soil the nitrogen content decreases in the CS, while in the MT there is a slight increase on 0-20 cm in both fertilization variants (0.178 with basic fertilization and 0.183 with additional fertilization) as shown in Table 2. Similar reports on the increase of total N content by application of systems for soil preservation and direct sowing were also presented in other studies. The results obtained by Neugsghwandtner et al. (2014) in Austria under similar condition to Turda area, presented values of the N content total (%): classic system with a 25-30 cm, the total N content was 0.194 (0-10 cm) and 0.195 (20-30 cm); conservative system 0.231 (0-10 cm), 0.198 (20-30 cm) in the NT system and 0.220-0.231 (0-10 cm), 0.206-0.192 (20-30 cm).

Table 2. The influence of the soil tillage system, of the crop-rotation and fertilization on the availability of soil in N (ARDS Turda 2007, 2018)

Year	Tillage system/ crop rotation/fertilization	Deepth sample	Total N (%)
2007	Classic $N_{40}P_{40}$	0-20 cm	0.162
		20-40 cm	0.124
2018	Classic $N_{40}P_{40}$	0-20 cm	0.147
		20-40 cm	0.126
	Minimum tillage $N_{40}P_{40}$	0-20 cm	0.178
		20-40 cm	0.153
	Classic $N_{40}P_{40}+N_{30}$	0-20 cm	0.150
		20-40 cm	0.122
	Minimum tillage $N_{40}P_{40}+N_{30}$	0-20 cm	0.183
		20-40 cm	0.134

The changes recorded in the phosphorus content, from the low values of 5 ppm and 9 ppm recorded in 2007 the highest increase was registered in the minimum tillage in both variants of fertilization in the 20-40 cm (45 ppm, 49 ppm) in 2018 (good supply). Also, in the classic system, the highest increase in the same depth of 20-40 cm was achieved (42 ppm, 27 ppm), the content being medium to good (Table 3).

Table 3. The influence of the soil tillage system, of the crop-rotation and fertilization on the availability of soil in P (ARDS Turda 2007, 2018)

Year	Tillage system/ crop rotation/fertilization	Deepth sample	P (ppm)
2007	Classic $N_{40}P_{40}$	0-20 cm	5
		20-40 cm	9
2018	Classic $N_{40}P_{40}$	0-20 cm	8
		20-40 cm	42
	Minimum tillage $N_{40}P_{40}$	0-20 cm	10
		20-40 cm	45
	Classic $N_{40}P_{40}+N_{30}$	0-20 cm	10
		20-40 cm	27
	Minimum tillage $N_{40}P_{40}+N_{30}$	0-20 cm	12
		20-40 cm	49

The soil on which the experiment was placed have a good supply of K (140 ppm) for the arable layer 0-20 cm and middle (126 ppm) for the layer 20-40 cm, the values determined in 2007. An increase of the K content can be ascertained, especially in the layer 0-20 cm in the MT (207 ppm-219 ppm) compared to CS (159 ppm -167 ppm) as show in Table 4.

The yield obtained at winter wheat cultivated in MT system, was slightly higher than those from conventional soil cultivation and at maize the yield was 5-11% lower, results obtained by Marin et al. (2015) from the research carried out at the Moara Domnească.

Average yields recorded in winter wheat, maize and soybean crops (included in the three-year rotation) had lower values especially in the NT and MT on the first year (Table 5).

Table 4. The influence of the soil tillage system, of the crop-rotation and fertilization on the availability of soil in K (ARDS Turda 2007, 2018)

Year	Tillage system/ crop rotation/fertilization	Deepth sample	K (ppm)
2007	Classic $N_{40}P_{40}$	0-20 cm	140
		20-40 cm	126
2018	Classic $N_{40}P_{40}$	0-20 cm	159
		20-40 cm	242
	Minimum tillage $N_{40}P_{40}$	0-20 cm	207
		20-40 cm	195
	Classic $N_{40}P_{40}+N_{30}$	0-20 cm	167
		20-40 cm	201
	Minimum tillage $N_{40}P_{40}+N_{30}$	0-20 cm	219
		20-40 cm	253

Table 5. The influence of the soil tillage system and the crop-rotation on the yield during 2007-2018, ARDS Turda

No.	Cultivar	Preceding crop	Year	Tillage system	Yield ($kgha^{-1}$)
1.	winter wheat	soybean	2007	Classic	4988^{Ct}
				No tillage	4830^{Ct}
			2010	Classic	5373^{**}
				No tillage	5448^{***}
			2013	Classic	5000^{ns}
				No tillage	5076^{*}
			2016	Classic	7198^{***}
				No tillage	7245^{***}
LSD (p 5%) = 188; LSD (p 1%) = 285; LSD (p 0.1%) = 458					
2.	maize	winter wheat	2008	Classic	5668^{Ct}
				Minimum tillage	5459^{Ct}
			2011	Classic	4851^{000}
				Minimum tillage	4783^{000}
			2014	Classic	6718^{***}
				Minimum tillage	6600^{***}
			2017	Classic	7919^{***}
				Minimum tillage	7726^{***}
LSD (p 5%) = 123; LSD (p 1%) = 186; LSD (p 0.1%) = 299.					
3.	soybean	maize	2009	Classic	1967^{Ct}
				Minimum tillage	1850^{Ct}
			2012	Classic	2096^{ns}
				Minimum tillage	2158^{**}
			2015	Classic	3163^{***}
				Minimum tillage	3295^{***}
			2018	Classic	2217^{**}
				Minimum tillage	2388^{**}
LSD (p 5%) = 135; LSD (p 1%) = 289; LSD (p 0.1%) = 432.					

^{Ct}-control; ^{ns}-not significant.

We believe that the largest production recorded in conservative systems is due to better accumulation and storage of water in the soil, as is shown by the research conducted by Şimon et al. (2018), the highest quantity of water from the soil was determined the case of applying NT, with a very significant positive difference of 52.7 m^3/ha compared to CS. The tillage system influences a lot the moisture reserve of the soil, the NT and MT systems are an important factor for the accumulation and preserve of the soil moisture.

CONCLUSIONS

After 12 years of experimentation, changes in soil pH, from low acid (0-20 cm) and neutral (20-40 cm) to low alkaline in all experimental variants and on both depth (7.6-8.4).

In the MT there is a slight increase of total N in the 0-20 cm layer in both fertilization variants (0.178% with $N_{40}P_{40}$ and 0.183% with $N_{40}P_{40+}N_{30}$) and decrease in the 20-40 cm (0.153% - 0,154%).

Increase content in phosphorus in the MT, from a weak content of 5 ppm and 9 ppm in 2007 to good content (0-20 cm) and very good on the 20-40 cm depth, in 2018.

The K content increase in arable layer of 0-20 cm in the MT (207 ppm -219 ppm) compared to CS (159 ppm-167 ppm).

The winter wheat in the four years where it occupied the place in the rotation set only in 2007 realized a lower yield (4830 kg/ha) in NT than CS (4988 kg/ha).

The yield obtained at soybean in MT has values close to those obtained in the CS, on the four years it occupied the rotation place the yield was higher in MT (2012, 2015, 2018) compared with CS.

Maize it is pretentious by cultivation in MT in the Turda area, during the whole experimental period, the yields were lower than the yields obtained in CS.

ACKNOWLEDGEMENTS

This work was supported by a grant of the Romanian Ministry of Research and Innovation, CCCDI-UEFISCDI, project PN-III-P1-1.2-PCCDI-2017-0056, within PNCDI III.

REFERENCES

Balota, E.L., Colozzi-Filho, A., Andrade, D.S., Dick, R.P. (2003). Microbial biomass in soils under different tillage and crop rotation systems. *Biol Fertil Soils, 38*, 15–20.

Cheţan, F., Haş, I., Malschi, D., Deac, V., Ignea, M., Şimon, A., Ivaş, A. (2011). Tehnological Features of the Winter Crop in the Conservation Agricultural System, The Agricultural Research-Development Station Turda. *Buletin USAMV Agriculture, 68*(1), 401. Book Abstract. Ed. Academic Pres Cluj.

Chetan, F., Rusu, T., Chetan, C., Moraru, P.I. (2016). Influence of soil tillage upon weeds, production and economic efficiency of corn crop. *AgroLife Scientific Journal, 5*(1), 36–43. ISSN 2285-5718.

Ciontu, C., Săndoiu, D.I., Penescu, A., Gâdea, M., Şchiopu, T., Obrişcă M., Dincă, L. (2012). The essential role of crop rotation and nitrogen fertilization in wheat and maize in the sustainable agriculture system of reddish preluvosoil. *AgroLife Scientific Journal, 1*(1), 68–77.

Marin, D.I., Rusu, T., Mihalache, M., Ilie, L., Nistor, E., Bolohan, C. (2015). Influence of soil tillage system upon the yield and energy balance of corn and wheat crops. *Agrolife Scientific Journal, 4*(2), 43–47.

Neugschwandtner, R.W., Liebhard, P., Kaul, H.P., Wagentrist, H. (2014). Soil chemical properties as affected by tillage and crop rotation in a long-term field experiment. *Plant Soil Environ, 60*(2), 57–622.

Šeremešic, S., Dalovic, I., Milošev, D., Jockovic, D., Pejic, B. (2013). Maize (*Zea mays* L.) yield stability dependence on crop rotation, fertilization and climatic conditions in a long-term experiment on Haplic Chernozem. *Zemdirbyste-Agriculture, 100*(2), 137–142.

Şimon, A., Rusu, T., Cheţan, F., Moraru, P.I. (2018). Impact of minimum tillage systems in conservation of water in the soil in the case of pea crops. *AgroLife Scientific Journal, 7*(1), 115–122.

***ANOVA, 2015. PoliFact and Duncan's test pc program for variant analyses made for completely randomized polifactorial experiences. USAMV Cluj-Napoca, Romania.

***SRTS, 2012. Romanian System of Soil Taxonomy. Ed. Estfalia, Bucharest.

***Turda Meteorological Station, 23°47′ longitude; 46°35′ latitude; 427 m altitude.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EVOLUTION OF LAND USE CATEGORIES AND THE EROSION DEGRADATION STATE OF THE AGRICULTURAL LANDS IN THE SUCEAVA COUNTY

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Abstract

The paper aims to bring in the foreground the long-term negative effects of soil erosion on the slope agricultural land in Suceava County. Located in the northeastern part of Romania, Suceava County occupies an area of 8553.5 km^2 (about 3.6% of the country`s surface), being the second largest county in the country, after Timis County. Related to the country`s largest geographical units, the territory of the county overlaps partially with the Eastern Carpathians and the Suceava Plateau. The relief characterized by a wide variety of forms (mountains, intramontane depressions, hills, plateaus, terraced valleys and river meadows) and the characteristics of the climatic, hydrographic, geological and pedological elements, correlated with the inappropriate exploitation of the land slopes, have determined over time the emergence of the erosion process on agricultural land. In 1990, Suceava County had an agricultural area of 349,502 ha, out of which 182,486 ha of arable land, 91,465 ha of pastureland, 69,960 ha of meadows and 5,491 ha of orchards. Compared to 1990, the total agricultural area increased to 354,820 ha, where 180,451 ha represent arable land, 2,913 ha orchards, 93,052 ha pastures and 78,404 ha of meadows. Between 1990 and 2017, the total agricultural area has increased by 5,318 ha, but the arable land and orchards were reduced by approximately 2,500 ha each, pastures and meadows increased by about 1,500 and, respectively 8,000 ha. More than half of the agricultural area is located on land with a slope of more than 5%. The lack of measures to prevent and combat erosion has led to a decrease in soil fertility, and in some cases to irreversible removing of the large area of land from the agricultural circuit.

Key words*: soil erosion, slope land, land use categories, eroded land.*

INTRODUCTION

The geographic position of Suceava County in the NE of Romania, with specific climate conditions, more severe than in the rest of the territory (low temperatures, abundant precipitation, high frequency and intensity of the wind) and higher forms of relief that are dominant in the area, represent factors that determine a relatively high degree of soil erosion (Savu P., 1999).

Suceava County occupies an area of 855,350 ha, out of which 354,820 ha (41.48%) represent agricultural land, and more than half of them are situated on slopes with a slope of more than 5%.

In these conditions, an accelerated soil erosion process is noticed, determined also by the intervention of the human factor on the environment, by massive deforestation, excessive grazing and the cultivation of land with large slopes without taking soil protection measures (Ungureanu Irina, 1998).

MATERIALS AND METHODS

The study was conducted using existing data from specialized state institutions such as the Suceava Agriculture and Rural Development Department, the Suceava Agricultural Payments and Intervention Agency and the National Land Improvement Agency Suceava regarding the evolution of the land use categories and surfaces degraded by erosion during the period 1998-2017.

Also from the national database (BDUST-Databased of the Agricultural soil-land units) conducted through the County soil-land Monitoring System for agriculture, data on land degradation by sheet erosion and gully erosion processes were extracted together with the data regarding the lands affected by landslides for the 65 territorial-administrative pedologically mapped units of Suceava County. These studies were carried out at the scale of 1: 10000 or 1: 5000 by the County Office of Pedology and Agrochemistry Suceava.

RESULTS AND DISCUSSIONS

In 1990, Suceava County had an agricultural area of 349,502 ha, out of which 182,486 ha of arable land, 91,465 ha of pastureland, 69,960 ha of meadows and 5,491 ha of orchards. Compared to 1990, the total agricultural area increased to 354,820 ha, where 180,451 ha represent arable land, 2,913 ha orchards, 93,052 ha pastures and 78,404 ha of meadows (Savu P, Bucur D., 2002).

Agricultural land has a significant distribution in the plateau unit, the arable surface is found in the plateau side and the pastures and meadow in the mountain unit (Figure 1).

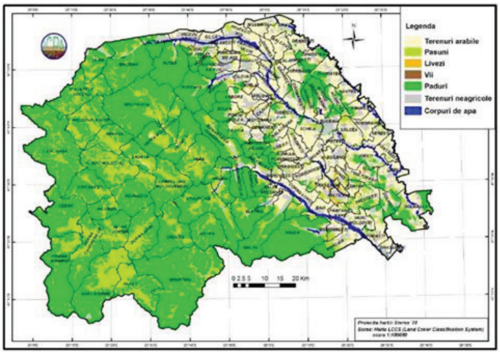

Figure 1. The map of land use categories distribution in Suceava County

As can be seen in Table 1, between 1990 and 2017 there were no significant changes in the total land area, but in 2015, with the changes of the surface of the forest fund, important land areas were introduced in the agricultural circuit. The reduction of the forest fund in the period 2015-2016 by about 11,000 ha was caused by massive and uncontrolled deforestation, most of it illegal.

Out of 11,000 ha deforested land, 5,318 ha were converted into agricultural surface, the land use categories recording the most significant increase are pastures and meadows.

More than half of the agricultural area is located on land with a slope of more than 5% (Figure 2).

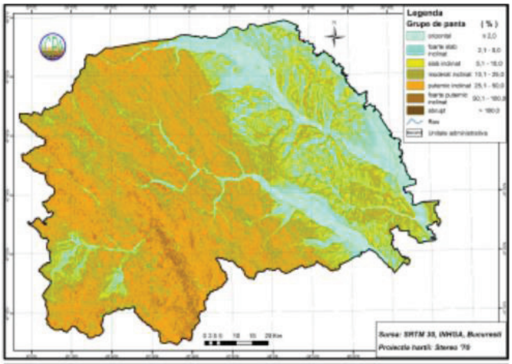

Figure 2. Sloping map in Suceava County

Between 1998 and 2017, the dynamic of land use categories on sloping agricultural lands shows a decrease of about 300 ha/year, by year 2017 arable land decreased by 11%, from 59,600 ha to 53,600 ha. At the same time, the orchards decreased by 37%, at the end of year 2017 remained only 1,400 ha of 2,200 ha as they existed in 1998, and the meadows were reduced by 2% (800 ha), the only increase was recorded by pastures, from 72,600 ha to 77,000 ha (Table 2).

The reduction of the areas cultivated on sloping land, especially arable land and orchards, was caused by the expansion of urban and rural areas, and also by the abandonment of degraded land where no profitable agriculture was possible.

Regarding the areas heavily affected by erosion, as seen in Figure 3, predominant in the Suceava Plateau, especially at the contact between different relief forms, is preponderant an agricultural area.

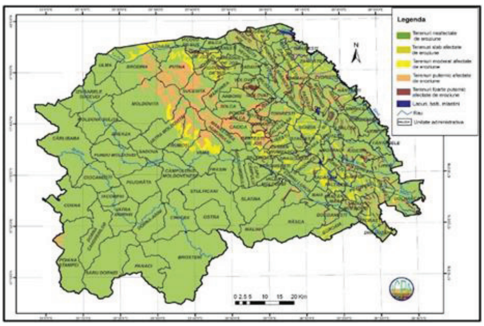

Figure 3. Soil erosion map in Suceava County

Table 1. The distribution evolution of land use categories in Suceava County, 1990-2017
(Suceava Agriculture and Rural Development Department)

Year	Total agricultural land		Land use categories							
	ha	%*	Arable		Pastures		Meadows		Orchards	
			ha	%	ha	%	ha	%	ha	%
1990	349,502	40.86	182,486	52.21	91,465	26.20	69,960	20.02	5,491	1.57
2000	349,502	40.86	178,473	51.06	93,749	26.82	74,009	21.17	3,271	0.93
2005	349,762	40.89	180,771	51.68	90,250	25.80	75,711	21.64	3,030	0.86
2010	347,920	40.68	180,678	51,93	90,274	25.95	73,960	21.26	3,008	0.86
2011	347,900	40,67	180,650	51.92	90,450	25.99	73,600	21.15	2,900	0.83
2012	347,805	40.66	180,372	51.86	90,570	26.03	74,053	21.29	2,810	0.80
2013	346,762	40.54	179,495	51.76	90,367	26.06	73,897	21.29	3,003	0.86
2014	347,835	40.66	179,646	51.64	90,563	26.03	74,175	21.32	3,001	0.86
2015	354,821	41.48	180,455	50.85	93,107	26.24	78,407	22.09	2,852	0.80
2016	354,820	41.48	180,451	50.85	93,052	26.22	78,404	22.09	2,913	0.82
2017	354,820	41.48	180,451	50.85	93,052	26.22	78,404	22.09	2,913	0.82

* of the county's surface

Table 2. The evolution in the period 1998-2017 of the land use categories
on the sloping agricultural lands in Suceava county (ANIF Suceava)

Land use categories	Cultivated surface				Situation of the year 2017 compared to year 1998
	1998		2017		
	ha	%	ha	%	
Arable	59,600	100	53,600	89	Surface decreased by 11%
Pastures	72,600	100	77,000	106	Surface increased by 6%
Meadow	63,800	100	63,000	98	Surface decreased by 2%
Orchard	2,200	100	1,400	63	Surface decreased by 37%

Approximately 31,000 ha of agricultural land is affected by sheet erosion, from low to excessive, and on about half of them the erosion process is manifested with high, very high and excessive intensity (Figure 4).

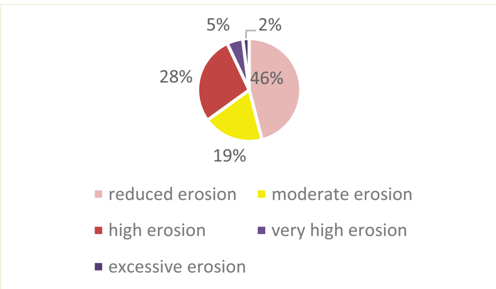

Figure 4. Percentage distribution
of agricultural land degraded by sheet erosion

From the analysis of the areas affected by gully erosion, it can be observed a large area occupied gully erosion (Figure 5) due to lack of works and measures to stop them from evolving.

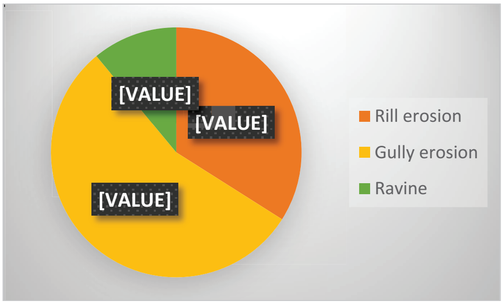

Figure 5. Percentage distribution of agricultural
surface degraded by gully erosion

If in 1998 only 4% of the slope agricultural land fund was in an advanced state of degradation by slope processes, in 2017, 18% (35,547.92 ha) of slope agricultural land fund (195,000 ha) are in different stages of degradation through sheet erosion and gully erosion. The increase of surface degraded by erosion was determined by the replacement of anti-erosion cultivation systems with hill to valley agricultural works, the deterioration or decommissioning of special hydro-technical

facilities built up to 1990 for the control of gully erosion, and also by the predominance of the maize continuous cropping in the crop structure.
Also, the most affected land use categories are arable lands (Figures 6 and 7), followed by pastures (Figure 8), orchards (Figure 9) and meadows.

Figure 7. A stabilized gully erosion on arable land in Foraşti

Figure 8. Pasture affected by sheet erosion and gully erosion, Preuţeşti

Figure 9. Terraced surface of a degraded orchard affected by sheet erosion, Fălticeni

Recommended measures and practices for preventing and combating soil erosion on agricultural land in Suceava County:

After the implementation of Land Fund Law 18/1991 and land repossession, an excessive fragmentation of the land areas occurred, most of them being arranged with the long side in the direction of the hill-valley, therefore obliging the owners of the land to execute the soil works in the direction from hill to valley (Savu P., Bucur D., Dascălu C., 1999; John R., 2018).
Thus, by merging the cultivated surfaces, the anti-erosion cultivation systems previously used can be implemented: contour plowing, contour buffer strips, strip cropping and terracing (Savu P., Bucur D, Ilişescu C, 1999; Dan S. et al., 2018).
Also, another urgent measure is aimed to (re) afforesting highly degraded land through ravines, where both soil fertility and level of soil mechanization have low values (Rafael Blanco-Sepúlveda, 2018; Hao C. et al., 2018; Auerswald K. et al., 2019).
The deforestation of large forest areas and their conversion into agricultural land where unsustainable agriculture is practiced has led to a decrease of soil fertility, and implicitly of agricultural production (Zhujun C. et al., 2019). It also determined the degradation not only of cultivated land, but also of roads, access ways and properties (Muqi Xiong et al., 2018) Therefore, in order to restore the ecological balance in the deforested areas, it is necessary to reforest them, but also to extend the silvic protection plantations on all the degraded agricultural lands and silvic areas (M. E. Lucas-Borja et al., 2019; Mahbubul A., 2018; Camera C. et al., 2018).
Regarding the special hydro-technical constructions for controlling gully erosion forms, the absence of maintenance work, inappropriate agricultural practices, as well as certain natural factors (especially the relief and torrential precipitations) led to their degradation (Léa Kervroëdan et al. 2018; Jianlin Z. et al., 2018). Consequently, in order to ensure hydrological balance on gully erosion forms, it is advisable to rehabilitate degraded works, wherever possible, through unclogging activities, rehabilitation of the drainage network, rehabilitation of the road platform etc.

Last but not least, the application of relatively new soil conservation measures such as the minimum tillage and no tillage system, which suppose abandoning ploughing or even all the soil works such as direct drilling (Valentin G. et al., 2018; Jesús Rodrigo-Comino et al., 2018; H. Vijith et al., 2018). It is also recommended to use the so-called green crops, sown immediately after harvesting the previous crop, which are designed to protect the soil during winter and to improve the physical, chemical and biological properties, implicitly to increase the fertility and resistance to erosion.

CONCLUSIONS

In 2017, Suceava County had an area of 354,820 ha of agricultural land, arable land of 180,451 ha, orchards and nurseries of 2,913 ha. The pastures and meadows totals 93,052 ha and respectively 78,404 ha.

Between 1990 and 2017, the total agricultural area has increased by 5,318 ha, but the arable land and orchards were reduced by approximately 2,500 ha each, pastures and meadows increased by about 1,500 and respectively 8,000 ha.

The reduction of the forest fund in the period 2015-2016 by about 11,000 ha was caused by massive and uncontrolled deforestation, most of it illegal.

Out of 11,000 ha deforested land, 5,318 ha were converted into agricultural surface, the land use categories recording the most significant increase are pastures and meadows.

The dynamic of land use categories between 1998 and 2017, on sloping agricultural lands shows a decrease of about 300 ha/year, by year 2017 arable land decreased by 11%, from 59,600 ha to 53,600 ha. At the same time, the orchards decreased by 37%, at the end of year 2017 remained only 1,400 ha of 2,200 ha as they existed in 1998, and the meadows were reduced by 2% (800 ha), the only increase was recorded by pastures, from 72,600 ha to 77,000 ha.

In 2017, 18% (35,547 ha) of the slope agricultural land fund (195,000 ha) are in various degree of sheet erosion and gully erosion.

The ecological and economic consequences of soil erosion will continue to manifest with increasing intensity as long as prevention and control measures are not taken, simple measures such as the cultivation of good protective plants, mulching, contour plowing, and other more complex works, such as special hydro-technical amelioration works for combating ravines (gabions, brushwood dams, concrete weir, dams, etc.).

ACKNOWLEDGEMENTS

This work was co-financed from Competitiveness Operational Program (COP) 2014 - 2020, under the project number 4/AXA1/1.2.3. G/05.06.2018, SMIS2014+ code 119611, with the title “Establishing and implementing knowledge transfer partnerships between the Institute of Research for Agriculture and Environment - Iasi and agricultural economic environment”.

REFERENCES

Camera, C., Djuma, H., Bruggeman, A., Zoumides, C., Faka, M.. (2018). Quantifying the effectiveness of mountain terraces on soil erosion protection with sediment traps and dry-stone wall laser scans. *CATENA*, *171*. 251–264.

Chen, H., Zhang, X., Abla, M., Du, Lü, Xihua, Yang (2018) Effects of vegetation and rainfall types on surface runoff and soil erosion on steep slopes on the Loess Plateau. *CATENA*, 141–149.

Chen, Z., Wang, L., Wei, A., Gao, J., Zhou, J. (2019). Land-use change from arable lands to orchards reduced soil erosion and increased nutrient loss in a small catchment. *Science of The Total Environment*, *648*. 1097–1104.

Dan, S., Weixin, Z., Yongbiao, L., Zhanfeng, L., Shenglei, F. (2018). Soil erosion and water retention varies with plantation type and age. *Forest Ecology and Management*, *422*. 1–10.

Golosov, V., Belyaev, V. (2013). The history and assessment of effectiveness of soil erosion control measures deployed in Russia. *International Soil and Water Conservation Research*, *1*(2), 26–35.

Hurmain, V., Hurmain, A., Dodge-Wan, D. (2018). Impacts of land use changes and land cover alteration on soil erosion rates and vulnerability of tropical mountain ranges in Borneo. *Remote Sensing Applications: Society and Environment*, *12*. 57–69.

Jesús, R., Manuel, S., Thomas, I., José María, S.G., Johannes, B.R. (2019). Rainfall-simulated quantification of initial soil erosion processes in sloping and poorly maintained terraced vineyards - Key issues for sustainable management systems. *Science of The Total Environment*, *660*. 1047–1057.

Jianlin, Z., Zhiqiang, Y., Gerard, G. (2019). Soil and water conservation measures reduce soil and water

losses in China but not down to background levels: Evidence from erosion plot data. *Geoderma*, *337*. 729–741.
John, R., Dymond, S., Vale., S. (2018). An event-based model of soil erosion and sediment transport at the catchment scale. *Geomorphology*, *318*. 240–249.
Karl, A., Peter, F. (2019). Soil organic carbon storage following conversion from cropland to grassland on sites differing in soil drainage and erosion history. *Science of The Total Environment*, *661*. 481–491.
Léa, K., Romain, A., Mathieu, S., Jean-François, O., Michel-Pierre, F. (2018). Plant functional trait effects on runoff to design herbaceous hedges for soil erosion control. *Ecological Engineering*, *118*. 143–151.
Lucas-Borja, K., González-Romero, J., Plaza-Álvarez, P.A., Sagra, J., de las Heras, J. (2019). The impact of straw mulching and salvage logging on post-fire runoff and soil erosion generation under Mediterranean climate conditions. *Science of The Total Environment*, *654*. 441–451.
Mahbubul, A. (2018). Ecological and economic indicators for measuring erosion control services provided by ecosystems. *Ecological Indicators*, *95*(1), 695–701.
Muqi, X., Ranhao, S., Liding, C. (2018). Effects of soil conservation techniques on water erosion control: A global analysis. *Science of The Total Environment*, *645*. 753–760.
Rafael, Blanco-Sepúlveda. (2018). An erosion control and soil conservation method for agrarian uses based on determining the erosion threshold. *MethodsX*, *5*. 761–772.
Savu, P., and Bucur, D. (2002). Organizarea și amenajarea teritoriului agricol cu lucrări de îmbunătățiri funciare. *Revista Lucrări Științifice. Seria Agronomie*, *Iași,* 89–96.
Savu, P. (1999). Proiect de cercetare, Lucrările de combatere a eroziunii solului, componentă majoră a menținerii echilibrului ecologic în Podișul Moldovei. *Revista Lucrări științifice. Seria Agronomie*, 36–41.
Savu, P., Bucur, D, Ilișescu, C. (2004). Cu privire la evoluția modului de folosință și a lucrărilor antierozionale pe terenurile agricole în pantă din județul Botoșani în perioada 1990-1995. *Revista Lucrări științifice. Seria Agronomie*, 42–49.
Savu, P., Bucur, D., Dascălu, C. (2009). Starea actuală și perspectivele combaterii eroziunii solului pe teritoriul județului Iași. *Revista Lucrări Științifice. Seria Agronomie*, 58–65.
Ungureanu, I. (1988). Dinamica peisajului geografic din Podișul Sucevei. *Universitatea „Al. I. Cuza”*, Iași.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SITE SELECTION FOR MANURE STORAGE FACILITIES USING GIS TECHNOLOGIES

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Abstract

Selection of potential suitable sites for the construction of manure storage facilities was made by use of Multi-Criteria Data Analysis in the GIS environment. From the wide variety of prescriptive models of multi-criteria data analyses, we selected and used in the research the decision model based on Boolean logic. In order to choose suitable locations for the construction of the manure storage facilities the decision model based on the Boolean logic was built taking into account 6 factors: slope; proximity to access roads; proximity to wells and springs; proximity to villages and sheepfolds; proximity to water bodies; land use. As a result of studying Moldovan legislation and bibliographic data, selection criteria were established for each factor that ensures the correct ecological, sanitary and aesthetic location of the manure storage facilities. As a result of the combination of the mentioned factors, the potential sites for the construction of the manure storage facilities with an area of 337400 m^2 or 33.74 ha were identified. The potential interest present 7 areas with an area greater than 1.8 ha.

Key words: *Boolean logic model, GIS, manure, site selection.*

INTRODUCTION

The problem of the use of manure in the Republic of Moldova is a priority both from the environmental and human health point of view. Actually most animals and birds are maintained in smallholdings of rural population. According to the statistical data for the period of 2013-2017 about 425-743 thousand tons of manure is formed in the Republic of Moldova yearly (Statistical Yearbook of the Republic of Moldova, 2018). At the same time only 29-55% of the manure was used as soil fertilizer. The rest of manure is usually discarded into unauthorized places. This illegal action leads to the nitrate pollution of the main drinking water sources and creates great health problems of rural population. The best way to minimize the impact of manure on the environment is to use it as soil fertilizer. Manure is an excellent source of nutrients for crops and it provides organic matter that plays enormous role in restoration of soil fertility. Collecting the manure in a locality requires building manure storage facilities where it will be stored and fermented accordingly to the ecological requirements.

The purpose of the research is to adapt and implement modern methods at selection of suitable locations for manure storage facilities using Geographic Information Systems (GIS) technologies. The location should be selected basing on the ecological and sanitary requirements, as well as the aesthetic point of view.

To achieve the purpose of the research, a decision model was built on the basis of the Multicriteria Data Analysis in the GIS environment. Generation of decision models was made by integrating relevant study factors. The result of the integration is a complex index, which represents the gradual suitability of the possible multitudes of alternatives. Such analyses are known as Multicriterial Data Analysis (Eastman J.R., 1999).

The research was carried out with the financial support of Academy of Sciences of Moldova under the project for young researchers Nr. 35/IND “Selection of suitable sites for manure application using Geographical Information Systems (GIS) technologies. Case study of Grinauti village, Riscani district”, project leader PhD Ciolacu Tatiana.

MATERIALS AND METHODS

The research was carried out in the northern part of the Republic of Moldova, Rascani district, Grinauti commune (Figure 1).

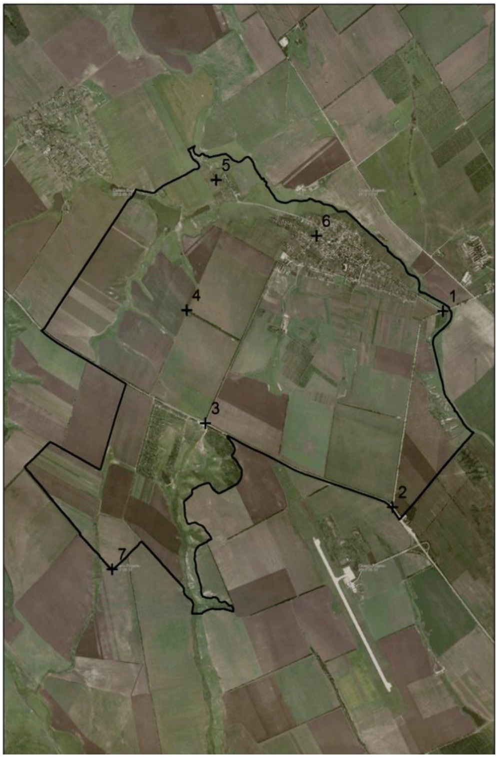

Figure 1. Research area of Grinauti commune, Rascani district

Initial data were collected from the next available sources:

Slopes layer was obtained through the built-in Slope tool in ArcGIS based on the Digital Elevation Model. The Digital Elevation Model (Figure 3) was generated using the Topo to Raster tool of ArcGIS based on the topographic map 1: 25000 available on the Land and Cadaster Agency website (Figure 2).

Proximity to access roads. Roads have been digitized from orthophoto images.

Proximity to wells and springs. The data regarding spatial location of wells and springs were taken with GPS.

Proximity to villages and sheepfolds. The data about location of villages and sheepfolds were extracted from the land use map. Land use, hydrographic network, road network were digitized from the ortofoto image available on the web portal of the Land and Cadaster Agency.

Land use. Data regarding land use were digitized from the orthophoto image available on the Land and Cadaster Agency web portal.

There is a wide variety of prescriptive models for multi-criteria analyses. In this paper a decision model based on Boolean logic was built. The Boolean model is performed by multiplying the criteria represented in binary form (O'Sullivan D., Unwin D.J., 2010):

$$S_i = \prod_{j=1}^{n} Cr_i^j$$

where:

Si represents index of suitability for spatial unit i;

Cr_i^j - value of criterion j for space unit i;

n - number of criteria.

Each pixel of a criterion in a Boolean model is considered "zero" if there cannot be installed manure storage facility, and "one" if installation is possible. By multiplying raster data composed of pixels with a value of "one" or "zero" as the result we obtain a raster of suitability where each pixel can have only "zero" or one "values".

Construction of decision models was performed using raster data because data of this type does not require topology validation after data combining. A raster data layer is a set of quadrants of the same size called pixels or cells, each pixel having a numeric value representing a real world feature. An object represented in a raster layer is made up of at least one pixel.

The following factors were used to construct decision models: slope; land use; proximity to villages and sheepfolds; proximity to the access roads (asphalt road, country road); proximity to springs and wells; proximity to water bodies (lakes, rivers); The factors were represented in the form of raster thematic layers.

RESULTS AND DISCUSSIONS

As a result of studying Moldovan legislation and bibliographic data, the selection criteria were established for each factor that ensures the correct ecological, sanitary and aesthetic location of the manure storage facilities (Ciolacu T. et al., 2018).

Slopes. Lands with a slope greater than 5° were considered unsuitable (Figure 4).

Proximity to access roads. Areas located at a distance less than 100 m from roads were considered unsuitable. By use of built-in instruments Buffers, Multiple Ring Buffer, and Clip we obtained buffer zones of 100 m from all the roads from aesthetic reasons (Figure 5).

Proximity to wells and springs. Areas at a distance more than 50 m from fountains and springs were considered acceptable. By use of built-in tools Buffers, Multiple Ring Buffers, and Clip we obtained buffer zones of 50 m from unsuitable areas for the construction of manure storage facilities (Figure 6).

Proximity to villages and sheepfolds. Based on the experience of other states, it was decided to use a 250 m buffer zone from all the villages and sheepfolds from the studied area (Basnet B.B. et al., 2001). Another argument for using a buffer zone for localities was the reduction of the polluting impact on aquifers near the locality. During the field researches, it was noticed that the lands near the sheepfolds were in a deplorable state because of manure accumulations, which made us apply here the same restriction as for the localities. Thus buffer zones of 250 m from unsuitable areas were built (Figure 7).

Proximity to water bodies. According to the RM Law Regarding Protection Areas for Rivers and Lakes no. 440-XIII 1995, art. 7, the width of riparian water protection strips should be not less than 100 m for large rivers, not less than 50 m for medium rivers and at least 20 m for small rivers (Ungureanu et. al., 2006). Thus, the land at a distance of less than 100 m from Raut river and 25 m from the rest of the small rivers from the territory was considered unsuitable for our purposes.

In the case of lakes was used 50 m buffer zone (Figure 8). Due to the fact that there was a wet area on the territory of the commune, it was decided to apply here the same restrictions as for the small rivers.

Land use. Pastures and the territory of the former farm were considered suitable for our purpose (Figure 9).

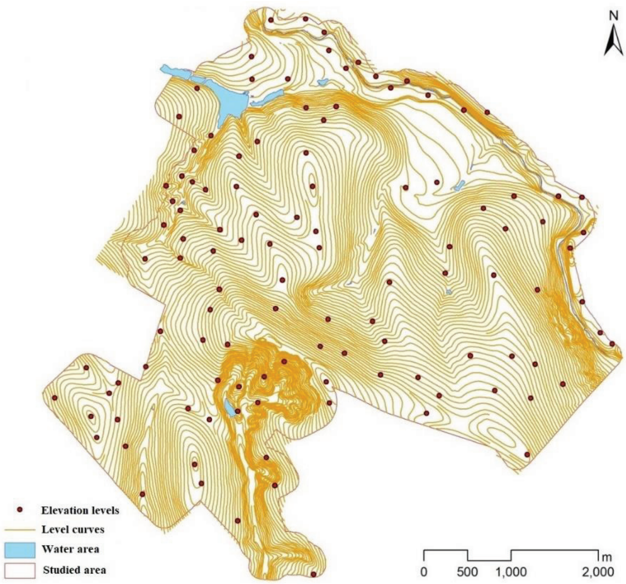

Figure 2. Topographic map of Grinauti commune, Rascani district

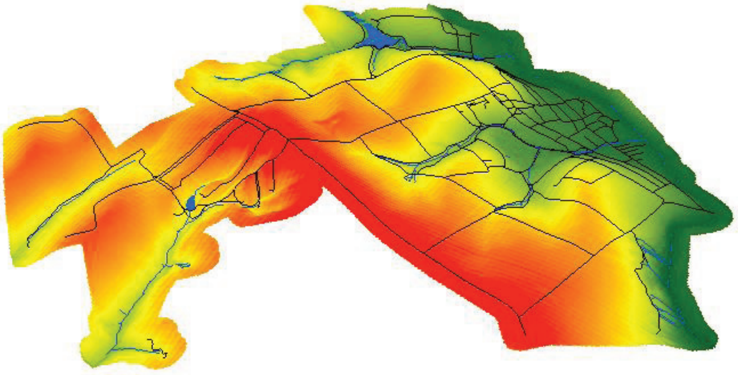

Figure 3. Digital Elevation Model of Grinauti commune, Rascani district

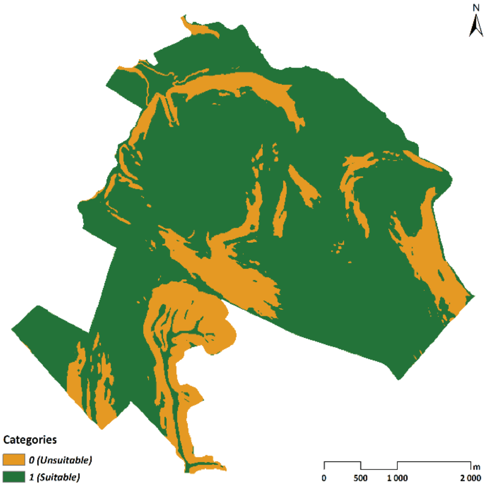

Figure 4. Slopes

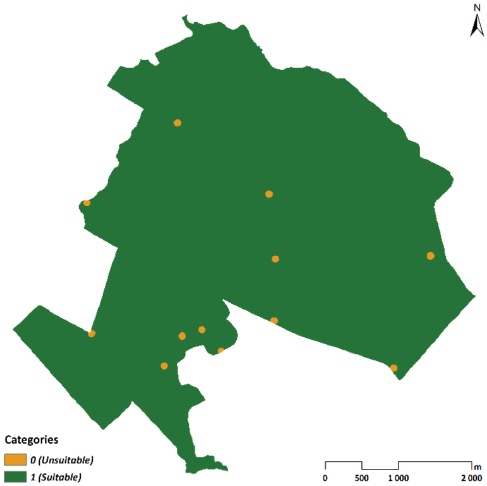

Figure 6. Proximity to wells and springs

Figure 5. Proximity to access roads

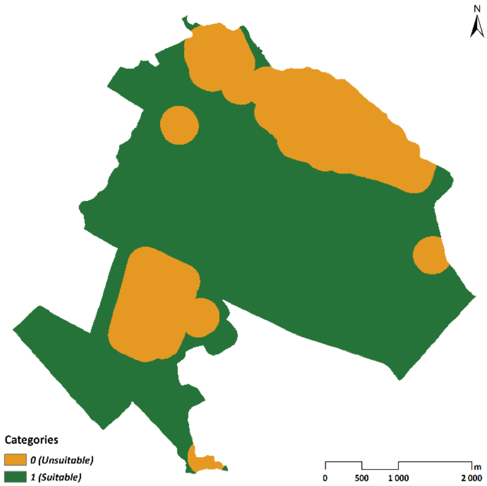

Figure 7. Proximity to villages and sheepfolds

Figure 8. Proximity to water bodies

Figure 9. Land use in Grinauti commune, Rascani district

Each of the above mentioned criteria was prepared to be combined in the Boolean model. Thus, the input data was reclassified in binary form according to the parameters described above.

As a result of the combination of the factors we obtained the results presented in Figure 10.

The potential sites suitable for the construction of manure storage facilities were identified with the total area of 337400 m^2 or 33.74 ha. The potential interest present 7 plots with an area greater than 1.8 ha.

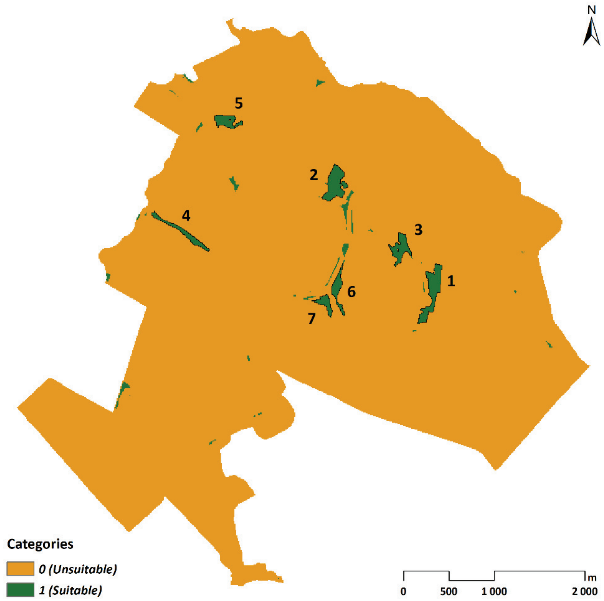

Figure 10. Boolean decision model determining suitable locations for construction of manure storage facilities

CONCLUSIONS

Application of Multicriteria Data Analysis in site selection of manure storage facilities was performed through the construction of Boolean decision model. We took into account six factors, that will ensure correct from ecological point of view, location of the facilities: slope; proximity to access roads; proximity to wells and springs; proximity to villages and sheepfolds; proximity to water bodies; land use. On the base of the constructed Boolean model, we identified seven sites, greater than 1.8 ha, suitable for the construction of the manure storage facilities.

Storing manure in authorized places such as manure storage facilities will minimize its environmental and human health impact in the studied community. Introduction of the manure into the soil as a fertilizer will increase soil organic matter content, will contribute to soil structure restoration and will diminish negative humus balance in soils.

ACKNOWLEDGEMENTS

This research work was carried out with the financial support of Academy of Sciences of Moldova under the project for young researchers Nr. 35/IND “Selection of suitable sites for manure application using Geographical Information Systems (GIS) technologies. Case study of Grinauti village, Riscani district”.

REFERENCES

Basnet, B.B., Apan, A.A., Raine, S.R. (2001). Selecting suitable sites for animal waste application using a raster GIS. *Environmental Management*, *28*(4), 519–531.

Ciolacu, T., Ciolacu, E., Lungu, M. (2018). Research regarding the possibility of fertilization of arable land from local manure sources using GIS technologies. *Scientific Papers. Series A. Agronomy, LXI*(2), 15–21. ISSN 2285-5785.

Eastman, J.R. (1999). *Multi-Criteria Evaluation and GIS. Geographical Information Systems.* Second Edition, Vol. 1, *Principles and Technical Issues*, Edited by Longley P.A., Goodchild M.F., Maguire D.J., Rhind D.W, 493–502. John Wiley & Sons Inc.

O’Sullivan, D., Unwin, D.J. (2010). *Geographic Information Analysis*. Second Edition. John Wiley & Sons, Inc., New Jersey.

Ungureanu, V., et al. (2006). *Practici agricole prietenoase mediului* (Environmental friendly farming practices). Guide. Chisinau: S.E. Central typography.

Statistical Yearbook of the Republic of Moldova (2018). National Bureau of Statistics of the Republic of Moldova. Chisinau: Central Typography.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

DESTRUCTION OF INTER-AGGREGATE RELATIONS BETWEEN PARTICLES OF SOIL IN THE PROCESS OF WATER EROSION

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Abstract

The resistance to the breaking of soil samples is three orders of magnitude greater than the shear stresses at the bottom of the slope flows, which produce work for the separation and transport of soil particles. Mirtskhulava believed that the separation of soil particles by a stream occurs as a result of fatigue breaking of bonds between soil particles (Mirtskhulava, 1966). With this in mind, the resistance of the soil to the gap is reduced by two orders of magnitude. Nearing M.A. believed that the separation of soil particles occurs at the points of vortex breakdown from the bottom of the stream, where the shear stress is two orders of magnitude greater than its average value (Nearing, 1991). These approaches do not explain the overcoming of the adhesion force between soil particles by slope flows. The results of the study of the effect of water temperature on the erosion rate of model soil samples showed a strong dependence of erosion on water temperature, which is close to the Vant-Hoff`s rule. It was experimentally shown that the destruction of bonds between particles soil in a sample of black earth mono-grained soil occurs under a layer of still water (Rose, 1985).

Key words*: erosion, inter-aggregate relations, soil resistance to tearing, Vant-Hoff rule.*

INTRODUCTION

Nearing et al. (1991) paid much attention to this issue. Studies of resistance to the breakthrough of 33 different soils of the United States showed that, according to this characteristic, all soils fall within the range of 0.9 to 3.2 kPa. An analysis of the results of his studies on monoaggregate soil samples showed that their resistance to rupture fits within the given range and is three orders of magnitude more than the shear stresses at the bottom of the slope shallow-water flows. However, soil samples were successfully eroded.

To explain this paradox, Nearing (1991) used the results of Grasse's research (Grass, 1970), according to which the tangential stresses at the points of the near-bottom area, where the vortex disturbances (burst event) are separated, exceed by two orders of magnitude the average tangent stresses at the bottom of the stream that it is at these points that the particles of the soil break away. Since the separation of the vortices from the bottom of the flow is stochastic, the equation for the separation of soil particles includes a probability block.

However, this solution does not seem to be entirely correct, since the tangential stress at the point of separation is the same, although it is two orders of magnitude higher than the average value of the tangent stress, and yet it is an order of magnitude lower than the resistance of the soil sample to the gap.

At the same time, the solution to this problem proposed by Mirtskhulava, also cannot be considered perfect. If we assume that the adhesion of the soil, determined by the method of pressing a spherical punch into the soil with full water saturation (Tsitovich, 1963), is equivalent to the resistance of the rock to break, as Mirtskhulava (1966), then the fatigue strength of the gap, of the same order as the shear stress in the places of the separation of the vortices.

Thus, the solution of the problem of the separation of particles of a cohesive soil by sloping water flows from the position of fatigue failure of the connection between soil particles is not quite correct.

MATERIALS AND METHODS

The resistance of soil particles to separation by the flow of water, as a rule, is much greater than those of hydraulic origin that the flow bed undergoes as a whole, as well as the soil particles composing it. That is why Mirtskhulava, in the erosion equation proposed by him back in 1970, used the notion of fatigue breaking of bonds between soil particles and soil particles under the influence of dynamic loads experienced by the flow bed due to local pulsations of water velocity.

Studies on the fatigue strength of soils and the rate of erosion showed the similarity of these processes, therefore, by analogy with the equation describing the fatigue strength curve, the number of stress cycles (N) before the moment of separation of a particle can be represented as:

$$N = \frac{188000}{\frac{v_{\Delta_x}^2}{v_{\Delta_{don}}^2} - 1} \quad (1)$$

Where: v_{Δ_x} and $\nu_{\Delta_{don}}$, respectively, the bottom velocity at the height of the roughness protrusions at a distance x from the beginning of the stream and the admissible (not blurring) speed.

The ratio of the squares of these velocities is equivalent, respectively, to the tension of the gap at the initial moment of the load on the soil particle and at the moment of its separation by the flow of water.

From equation (1), knowing the frequency of the velocity pulsations in the stream, it is easy to determine the number of particles disrupted by the stream per unit time and, spreading this number per unit area, get the flush equation in the form:

$$q = 0.0000064 * \gamma\omega d^2 (\frac{v_{\Delta_x}^2}{v_{\Delta_{don}}^2} - 1) \quad (2)$$

Where: q is the flushing of soil per unit area d - diameter of detachable particles; γ is the particle density; ω - the frequency of the velocity pulsations in the stream. Later Mirtskhulava (1970) accepted the frequency of pulsations in sloping streams as a constant. The permissible speed, according to his ideas, is a function of the square root of the weight of a soil particle in water and the fatigue strength of a soil to tear (S_y). The latter is determined by the dependency:

$$S_\gamma = 0.035 * S \quad (3)$$

Where: S is the adhesion determined by indentation of a spherical stamp into the soil.

RESULTS AND DISCUSSIONS

There are other facts that contradict the idea of the hydraulic nature of the forces responsible for the destruction of bonds between soil particles. According to many erosion models (Foster, 1982; Rose, 1985), the intensity of the flush is linearly dependent on the magnitude of the active erosion factor.

In the hydrophysical model of erosion (Larionov & Krasnov, 2000), the intensity of the washout is proportional to the cube of the flow velocity in the velocity range 1.5-2 times higher than the threshold values. To confirm this position, experiments were conducted on erosion of model soil samples with a density of 1.2 g/cm^3 in a wide range of velocities. The experiments yielded positive results (Figure 1), except for the region of low velocities, a linear relationship was obtained between the flush and the cube of the flow velocity.

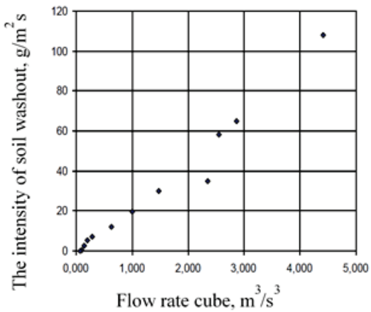

Figure 1. The dependence of the intensity of erosion model soil sample with a density of 1.2 g/cm^3 from the cube flow rate

However, the study of samples of higher density found that the relationship between the rate of erosion of the samples and the cube of the flow velocity has a distinct fracture (Figure 2).

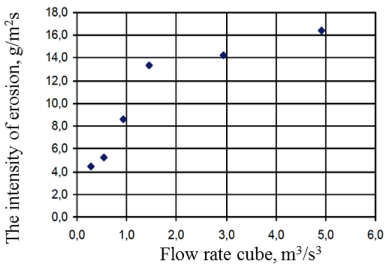

Figure 2. The dependence of the intensity of erosion model soil sample with a density of 1.5 g/cm^3 from the cube flow velocity

It turned out that the intensity of the separation of soil particles is not proportional to the cube of the average flow velocity in the entire studied range of velocities. Starting from a certain speed, there is a sharp flattening of the trend line on the graph of the dependence of the intensity of flushing from the cube of the flow velocity.

The first attempt to explain this phenomenon was based on a fatigue approach to breaking bonds between soil particles. As is well known, in the process of fatigue disturbance, cyclic oppositely directed loads lead to the movement of material relative to each other at points with a structural defect, where cracks appear over time, and then the material is destroyed.

In the case of unidirectional cyclic loads, which is typical for particles lining the flow bed, reciprocating motion in the material can take place only if there are elastic properties in the material. If we assume that soil particles have the property of elasticity, then we can assume that the separation of particles by water flow occurs due to multiple cycles of reciprocating movements, which are possible only in the case when the minimum values of hydraulic loads are less than the elastic forces of the particles.

With an increase in minimum loads, the amplitude of reciprocating movements will decrease, which may be the cause of the relative slowing down of the intensity of the separation of particles with increasing flow velocity. With an unlimited growth of the flow rate, the minimum pulsating loads obviously exceed the elastic forces of the soil particles, as a result of which the reciprocating translational oscillations of the particles will cease and, accordingly, their separation will stop. However, the erosion of the model soil samples did not stop at a speed of more than 7 m/s. Thus, the idea of fatigue breaking of bonds between soil particles in the process of erosion and in this case does not work.

The case suggested the direction in which to look for a solution to the problem of breaking bonds between particles in the process of erosion by slope flows, shear stresses at the bottom of which are three orders of magnitude less than the resistance of the soil to breaking. Once, when conducting a series of experiments at a constant flow rate, the water temperature increased as a result of the pump operation from 13 to 25ºC, while the erosion rate of soil samples increased. In this regard, an experiment was conducted to study the effect of water temperature on the erosion rate of samples in the range from 0 to 25ºC with a step of 5ºC (Figure 3).

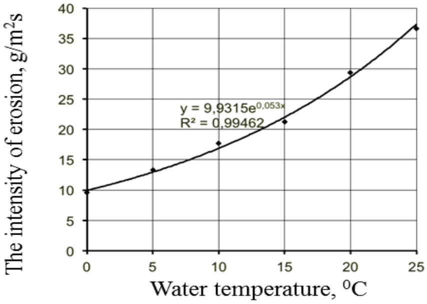

Figure 3. The effect of water temperature on the erosion rate of a soil sample

It turned out that when water temperature increases by 10ºC, the erosion rate (erodibility) increases by 1.5-1.6 times, which practically fits into the framework of the Van't Hoff rule, which reflects the influence of the speed of movement of molecules, reactants, frequency and force of their collisions. From this it follows that the destruction of bonds between soil particles is the result of the action not of hydraulic forces, but of the kinetic energy of water molecules.

In this regard, exploratory studies have been conducted, the purpose of which were: 1) developing a methodology for determining the rate of destruction of bonds between soil particles; 2) search for factors determining the rate of destruction of inter-aggregate bonds in model soil samples.

The destruction of bonds between soil particles is not visually determined; it is also impossible to mechanically separate particles that have lost contact with the soil mass. The most optimal is the use of water flow to remove particles that have lost contact with the underlying particles from the sample surface. In this regard, to assess the role of water as a substance that causes disruption of bonds between soil particles, the following method was adopted. Water was fed into the tray with pauses, in the continuation of which the water flow was absent, but the soil sample was under water with a layer of 1 cm. The experiment began with a pause.

At 1-2 cm below the sample, a blind jumper with a height of slightly more than 1 cm was placed and water was poured in an amount that provided a water depth of 1 cm. After a pause, the active phase of the experiment followed the jumper was removed and the pump was turned on. Water was fed into the tray for a specified time. Then the pump was turned off, the jumper was installed again, and the resulting container was filled with water. In the very first seconds, all particles that had lost contact with the sample were washed off the sample surface.

This process can be visually observed a mass of soil particles moves along the tray (Figure 4), while with the traditional method of testing samples, individual particles or groups of particles are rarely observed.

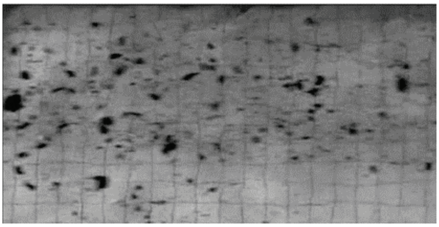

Figure 4. View of the tray below the sample. Dark spots are particles and groups of soil particles, torn off by a stream

With an increase in the duration of the pauses, the number of particles that have lost their bond increases with some delay (Figure 5). This can be explained in two ways. With the accumulation of particles lacking bonds, they all more strongly press down (press down) the underlying particles, which makes it difficult or completely stops the propagation of the process of breaking bonds deep into the sample (substance).

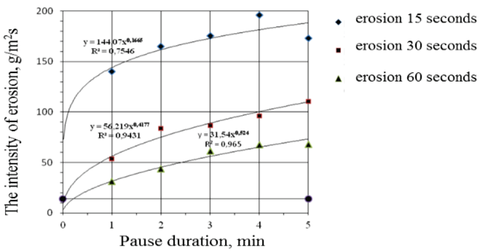

Figure 5. Slowing down the intensity of erosion of the sample with an increase in the duration of the pause

Another explanation may be that the molecular movement of water between soil particles slows down as the front breaks the destruction of bonds between particles inwards, which leads to a decrease in the rate of breakdown of bonds between particles.

Maximal significance decreases with increasing magnification, so that witnesses are shifted and destroyed by the extent of the active actives of the front. This type of character may, in any event, be in the same way that it has a role in disturbing the interplay between the players playing the forces of the non-military nature, namely, the forces of mediation. Attention is drawn to the fact that the circumstance, the gradient of the graphical dependencies and the intensity of the cubic velocities (Figure 2) can be considered as a follow-up. On the perimeter of the crown, the lines cut the graph of the speed of the destruction between the particles, which is under the action of the molecule of the water molecules, which is carried out in the water, which is free of charge. By increasing the speeds, the flow increases the intensity of the particle grows, and the increase in the acceleration of the water vaporizes the chemical substances as a layer of the soil, with the non-destructive effects between the particles of the soil. Otherwise, with the acceleration of freeze-thrust acceleration, it increases the speed of destruction between consoli-dirovannımy particles.

However, the ability to flow into the unconsolable parts, the speed of the deconsolation particles, the line of the graph (in Figure 2).

By increasing the speeds, the flow of the gyra-skiing stream is taking part in disturbing the intersection of the parts of the soil in a supreme layer of the image of the soil, and, hopefully, the hydraulic force is increased by the increase of the increase in the number of droughts. Check and quantify the value of this item in the top of the page.

In Figure 6 presents three options for placing fields on a bilateral slope.

The first option (a) provides for the location of 2 and 3 fields with long sides along the slope with tillage in the same direction, which is unacceptable for reasons of protecting the soil from erosion.

In the second variant (b) with tillage strictly along horizontals, the best protection of the soil from flushing is provided; however, this option is difficult to implement due to technological and economic considerations.

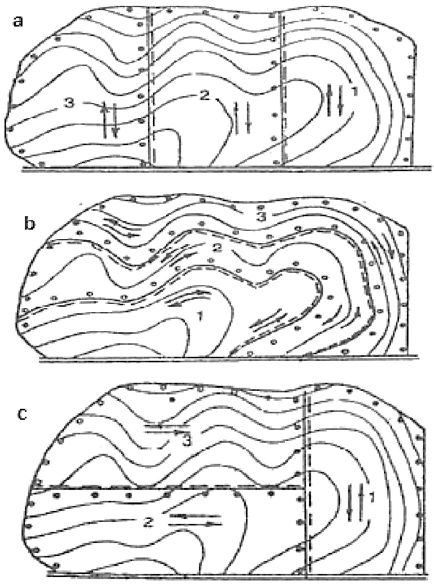

Figure 6 (a, b, c). Placement of fields on the slope and the direction of tillage

In the third variant (c), the soil is slightly worse protected than in the second, however, if we consider that the costs of tillage in the third variant are less than in the second, the third variant (c) will be optimal for the farm.
As a result of the fact that this is likely to be the result of a deterioration in the speed of destruction between particles under the water. It can be avoided, however, that it is not acceptable to use that model. The second part of the thesis is based on a detailed description of the size of the figure in the gypsum building, in which the roughness of the substructure is about 0 and in the other about 1 atm.
In the second case, speeding up the pattern of soil and, consequently, speeding up the destruction of particles between the particles of water and the chemical substances in the course of the chemical process, in the case of zero chemical reactions.
Finally, it can be assumed that the adhesion between the soil particles is ensured not only by Van-der-Waals forces, but also, for example, by polymer compounds. To confirm this hypothesis, several samples were dried, then moistened to their original condition and then subjected to erosion testing. And this assumption was justified. After drying, the samples were practically not eroded.
Since the soil is still washed off under natural conditions, it has been suggested that the bonds between the particles can also be destroyed by cyclic wetting and drying, as well as other weathering factors.
Thus, the research results allowed to reveal the mechanism of destruction of bonds between soil particles in the process of water erosion, which will serve as the basis for the development of physically sound models of erosion.

CONCLUSIONS

The first attempt to explain this phenomenon was based on a fatigue approach to breaking bonds between soil particles. As is well known, in the process of fatigue disturbance, cyclic oppositely directed loads lead to the movement of material relative to each other at points with a structural defect, where cracks appear over time, and then the material is destroyed.
It turned out that when water temperature increases by 10ºC, the erosion rate (erodibility) increases by 1.5-1.6 times, which practically fits into the framework of the Van't Hoff rule, which reflects the influence of the speed of movement of molecules, reactants, frequency and force of their collisions.
With an increase in minimum loads, the amplitude of reciprocating movements will decrease, which may be the cause of the relative slowing down of the intensity of the separation of particles with increasing flow velocity. With an unlimited growth of the flow rate, the minimum pulsating loads obviously exceed the elastic forces of the soil particles, as a result of which the reciprocating translational oscillations of the particles will cease and, accordingly, their separation will stop. However, the erosion of the model soil samples did not stop at a speed of more than 7 m/s. Thus, the idea of fatigue breaking of bonds between soil particles in the process of erosion and in this case does not work.
In this regard, exploratory studies have been conducted, the purpose of which were: 1) developing a methodology for determining the rate of destruction of bonds between soil particles; 2) search for factors determining the rate of destruction of inter-aggregate bonds in model soil samples.

As a result of the fact that this is likely to be the result of a deterioration in the speed of destruction between particles under the water. It can be avoided, however, that it is not acceptable to use that model.

The second part of the thesis is based on a detailed description of the size of the figure in the gypsum building, in which the roughness of the substructure is about 0 and in the other about 1 atm.

REFERENCES

Larionov, G.A. & Krasnov, S.F. (2000). Probabilistic model of soil erosion and cohesive soils. In: *Pedology*, No. 2, 616–624.

Mirtskhulava, T.E. (1966). Method for calculating canal deformation. In: River transport. No. 5, 27–36.

Mirtskhulava, T.E. (1970). Engineering methods for calculating and forecasting water erosion. M. Kolos, 240.

Tsytovich, N.A. (1963). Soil mechanics. M.: Gosstroyizdat, 636.

Grass, A.J. (1970). Initial instability of fine bed sand. In: J. Hydraulics Div. ASCE 96, 619–632.

Foster, G.R. (1982). Modeling the erosion process. Hydrlogic modeling of small watersheds, Eds. Haan, C.T., Johnson, H.P., Brakensiek, D.L. ASAE. Monogr., No. 5, St. Joseph, 296–380.

Nearing, M.A. (1991). A probabilistic model of soil detachment by shallow turbu-lent flow. Trans. ASAE, Vol. 34. 81–85.

Nearing, M.A., Parker, S.C., Bradford, J.M., Elliot, W.J. (1991). Tensile Strength of Thirty-Three Saturated Repacked Soils. *Soil Sci. Soc. Am. J., 55*(6), 1546–1551.

Rose, C.W. (1985). Developments in soil erosion and deposition models. *Advances in Soil Science, 2*. 163.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

MAIN CHARACTERISTICS OF FOREST SOILS ACROSS GETIC PIEDMONT (SOUTH-WESTERN ROMANIA)

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Abstract

In recent years, the importance of the forest soils started to gain an increasing attention both among forest specialists, policy makers and other stakeholders. This is mainly due to their importance for humanity, being the base for assuring several forest products and services. Since the 1980s, the Romanian soil specialists developed several soil taxonomy systems, based on the developments and data acquired at a certain moment. Knowing the type of the soil and its characteristics is crucial also for the foresters, for a sustainable forest management point of view. The aim of this study was to highlight the main characteristics of the forest soils across Getic Piedmont. Data from the forest management plans of twelve forest district that are responsible for managing the forests across the region were taken into account. The main representatives of Luvisols Class accounted for three quarters of the forest soils. In general, the characteristics of the forest soils are favorable for mixed-oak forest stands. In the future, special attention should be given to preserving the forests composed by autochthonous species, in order to maintain the fertility of the forest soils.

***Key words**: base saturation, forest soils, Getic Piedmont, humus, pH.*

INTRODUCTION

Soils are vital for mankind, providing several products and services, such as food and fresh water (Abrahams, 2002; Huang et al., 2018), mitigating the effects of the contemporary climate change (Lal, 2004) and conserving the biodiversity of the ecosystems (Crișan et al., 2017). In this context, it is mandatory to maintain their physical, chemical and microbiological characteristics to optimal levels (Morariu et al., 2018).

In the last decades, increasing attention was given to the impact of different human activities, such as agriculture or different forest operations, on soil characteristics (Worrell and Hampson, 1997; McFero Grace et al., 2006; Samuel et al., 2017). In particular, many foresters started to take into account the relation between the forest soils, the tree layer composition and the silvicultural measures in order to highlight the best combination from a sustainable development point of view (Enescu et al., 2018).

Moreover, the role of the forest soils as carbon sinks (Dixon et al., 1994; Lal, 2005; Edu et al., 2013; Chen et al., 2018), which, in general, recorded higher carbon concentrations in comparison with the agricultural soils, is gaining an increased interest among experts (Baritz et al., 2010).

Almost three decades ago, the European Commission started creating a soil database called LUCAS (Land Use and Land Cover Area Frame Survey), which is managed by the Eurostat (Montanarella et al., 2011). LUCAS is an in-situ survey (Panagos et al., 2012) aimed at assessing in a harmonized way the characteristics of the topsoil layer of the non-forest lands (Blujdea et al., 2016).

The database comprises more than 45,000 soil samples originating the EU Member States, which were analyzed (Tóth et al., 2013; Orgiazzi et al., 2018). In 2012, Romania added to the database 1,384 soil samples (Tóth et al., 2016).

In Romania, starting from 1980s, three soil classification systems were developed, namely SRCS-1980 (valid for the timeframe 1980-2002), SRTS-2003 (used in the period 2003-2012) and SRTS-2012, in use starting from 2013 (Enescu & Dincă, 2018).

The most common forest soils from Romania are represented by the dystric cambisols, haplic luvisols and eutric cambisols (Dincă et al., 2014) and the value of the organic C stock ranges between 70 and 360 t/ha (Dincă et al., 2015).

As regards the soil pH (soil reaction), at national scale, it was found that in the mineral horizon the pH values are slightly smaller in the coniferous forests in comparisons with the hardwood stands. Moreover, if we take into consideration the three historical provinces from Romania, it was reported that the pH values from the mineral horizon is 3.98 in Transylvania, 4.66 in Muntenia and 4.98 in Moldova, in the case of the soils with hardwood species, these values being higher than in the case of the coniferous-dominated sites (*i.e.* 3.50 in Transylvania, 3.44 in Muntenia and 4.04 in Moldova), respectively (Geambaşu et al., 2004). High amplitude of the pH values was also recorded on limited area, such as the one from Poiana Stampei Peat Bog, where the values ranged between 4.09 and 5.89 (Cazacu et al., 2018).

The aim of this study was to highlight the main characteristics of the forest soils from Getic Piedmont.

MATERIALS AND METHODS

Getic Piedmont, the largest piedmont of Romania (Bălteanu et al., 2010) and a relict geomorphologic relief unit (Stănilă et al., 2010), is located in the southern-western part of Romania (Figure 1), between the Danube (in the west and south) and Dâmboviţa River (in the east). It is a region characterized by intense human activities that generated several land-use changes (*e.g.* deforestation, fish fauna destructuring, exploitation of minerals) which occurred in the last two centuries (Tomescu, 2005; Spârchez et al., 2009; Bănăduc et al., 2013; Cărăboiu & Niţă, 2013; Anghelache & Burea, 2018).

One of the most visible effects of the deforestation is represented by the relief fragmentation (Ionuş et al., 2011).

The above-mentioned land-use changes were mainly generated by the favorable conditions for living, the number of the localities, according to the census made two decades ago, being more than one and a half thousand (Dumitrescu, 2003). Nowadays, most of the areas that were deforested, especially in the southern part (Popescu, 2009) are cultivated with several cereals, wheat and corn being the most common ones, but also large areas were reforested with fast-growing species, such as black locust (Enescu & Dănescu, 2013).

Getic Piedmont is part of Getic Depression, an area where the sedimentation process started in Triasic and continued until Cuaternary (Tomescu et al., 2008).

The area is characterized by a high variability in terms of precipitation (Pleniceanu & Alina, 2003), several hydrological hazards being reported in the last years especially in the eastern part of the region (Tanislav & Alexe, 2012). Getic Piedmont is also characterized by a dense gully system (Boengiu et al., 2012) and a significant percentage of its area is susceptible to landslides (Bălteanu et al., 2010).

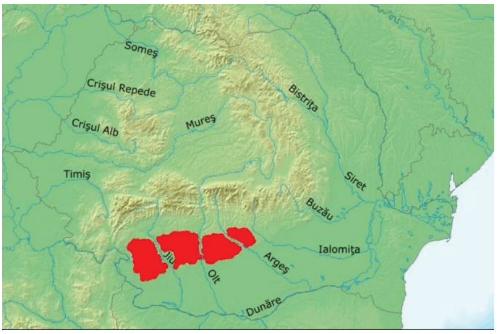

Figure 1. Location of Getic Piedmont
(Source: ro.wikipedia.org)

Data regarding the forest soils characteristics were gathered from the "Marin Drăcea" National Institute for Research and Development in Forestry. The database included the information available for the timeframe 1986-2014 for the twelve forest districts across Getic Piedmont, namely

Cotmeana and Poiana Lacului (Argeş Forestry Directorate), Cărbuneşti, Hurezani, Motru, Peşteana and Târgu Jiu (Gorj Forestry Directorate), Corcova and Topolniţa (Mehedinţi Forestry Directorate) and Băbeni, Horezu and Stoiceni (Vâlcea Forestry Directorate), respectively.
The main chemical characteristics were recorded for each pedogenetic horizon.
Afterwards, the data were centralized by the aid of Microsoft Office package, and the variation of the soil reaction (pH) and the base saturation degree (V%) was highlighted by using a Box and Wisker Plot model.

RESULTS AND DISCUSSIONS

A total of 566 soil profiles and 1796 pedogenetic horizons were analyzed. According to the centralized data, Luvisol and Preluvisol were the most common forest soils, accounting for three quarters out of the total forest soils across Getic Piedmont (Figure 2). The less common forest soils were the Fluvisol type.

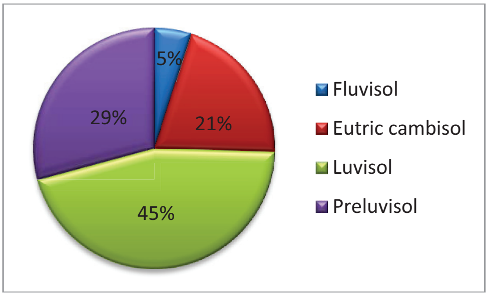

Figure 2. Main forest soil types from Getic Piedmont

A high variation was recorded for soil reaction (pH), as follows:
In the case of fluvisols, the minimum pH values were 4.20 and 4.41 in Ao and C horizons, respectively, while the maximum values recorded in the same horizons were 7.98 and 8.23, respectively (Figure 3).
As regards eutric cambisols, the minimum pH values were 3.67 and 4.11 in Ao and Bv horizons, respectively, and the maximum recorded value was 7.90 for both horizons (Figure 3).
In the case of luvisols, the minimum pH values were 4.03 in Ao horizon, 3.55 in El horizon 3.82 in Bt horizon, respectively. The average value for soil reaction ranged between 5.00 and 5.30 (Figure 3).
The minimum pH values for preluvisols were 3.52 and 4.17 in Ao and Bt horizons, while the maximum values recorded in the same horizons were 8.60 and 8.03, respectively (Figure 3).
In general, by taking into account the average values of the soil reaction, the soils are acid and weakly acid, being suitable for many hardwoods including the autochthonous oak species, such as Turkey oak (*Quercus cerris* L.), Hungarian oak (*Q. frainetto* Ten.), pedunculate oak (*Q. robur* L.) and sessile oak [*Q. petraea* (Matt.) Liebl.].
As regards the humus content (H, %), the highest values for A horizon were recorded in the case of luvisols, followed by eutric cambisols and preluvisols (Table 1).
In general, the forest soils across Getic Piedmont are moderately humiferous to intensely humiferous, being favorable for the main tree species from the region, especially for the oaks.

Table 1. Average humus content

Soil type (horizon)	Humus content (%)
Fluvisol (Ao)	3.33
Fluvisol (C)	1.05
Eutric cambisol (Ao)	4.94
Eutric cambisol (Bv)	1.59
Luvisol (Ao)	6.2
Luvisol (El)	2.21
Luvisol (Bt)	0.99
Preluvisol (Ao)	4.8
Preluvisol (Bt)	1.43

The variation of the base saturation degree is given in Figure 4. The highest values were recorded in the case of fluvisol, while the lowest values were recorded for luvisol.

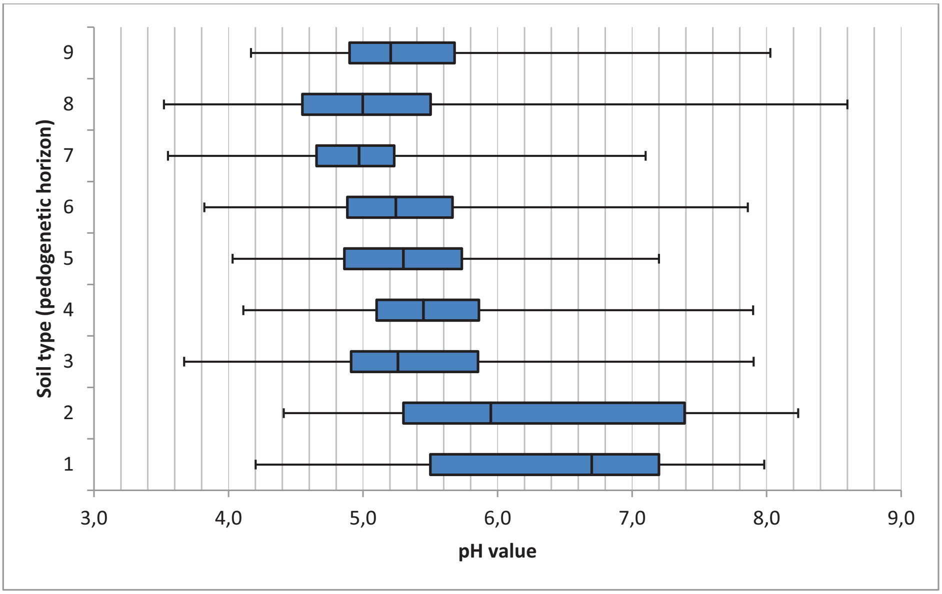

Figure 3. Box and Wisker Plot of the pH variation of genetic horizons for the most widespread forest soils from the Getic Piedmont: 1-Fluvisol (Ao), 2-Fluvisol (C), 3-Eutric cambisol (Ao), 4-Eutric cambisol (Bv), 5-Luvisol (Ao), 6-Luvisol (Bt), 7-Luvisol (El), 8-Preluvisol (Ao), 9-Preluvisol (Bt)

In the case of the fluvisols, the average value of the base saturation degree (V) was 74.30% in Ao horizon and 79.00% in C horizon, respectively, meaning that these soils were mesobasic-eubasic. Eutric cambisols had a value of 65.20% in Ao horizon and 75.80% in Bv horizon, respectively, being mesobasic soils. In general, luvisols and preluvisols were mesobasic soils (Figure 4).

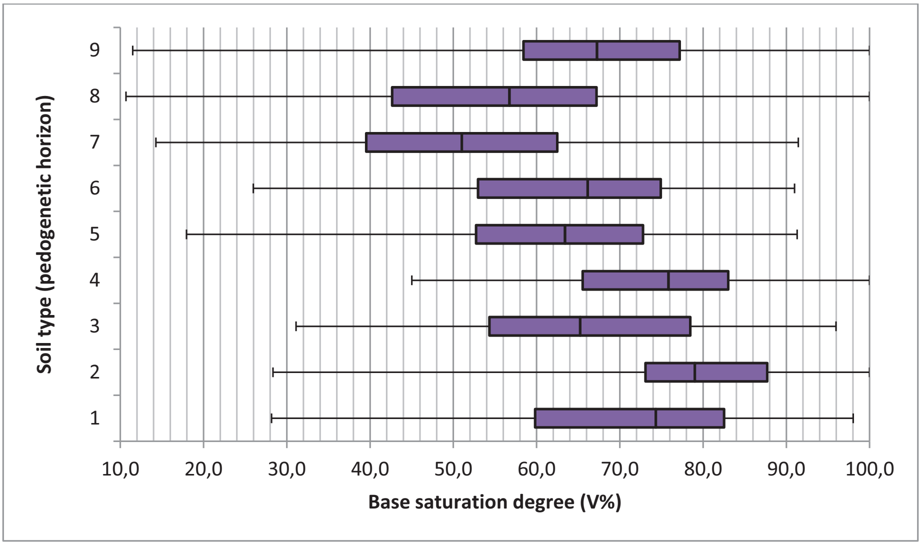

Figure 4. Box and Wisker Plot of the Base Saturation degree (V) variation of genetic horizons for the most widespread forest soils from the Getic Piedmont: 1-Fluvisol (Ao), 2-Fluvisol (C), 3-Eutric cambisol (Ao), 4-Eutric cambisol (Bv), 5-Luvisol (Ao), 6-Luvisol (Bt), 7-Luvisol (El), 8-Preluvisol (Ao), 9-Preluvisol (Bt)

CONCLUSIONS

Three quarters of the forest soils across Getic Piedmont belong to Luvisols class, being soils characterized by a high fertility for mixed forest stands dominated by oak species. In order to preserve their characteristics and fertility, future silvicultural measures should be focused on maintaining and/or increasing the area of oak-dominated stands, even if several challenges exist.

This study represents an overview of the main forest soils across Getic Piedmont and, in our opinion, its results should be interested both for practitioners, but especially for the policy makers who are responsible of maintaining and promoting a sustainable forest management, including preserving current forest ecosystems, composed by valuable autochthonous species. It is well known that in case of the forest soils were the forest was eliminated and land-use changes occurred (*i.e.* deforestation), their acidity increased, meaning that in the next couple of years those soils will be degraded and only a very limited species will be able to grow. This is what happened in the region in the last two hundred years, and as a result, nowadays the lands that were covered by mixed oak species are either degraded, either planted with black locust or other allochthonous fast growing tree species.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Abrahams, P.W. (2002). Soils: their implications to human health. *Science of The Total Environment*, *291*(1-3), 1–32.

Anghelache, C., Burea, D. (2018). Analiza evoluției industriei în România. *Revista Română de Statistică*, 10, 109–116.

Baritz, R., Seufert, G., Montanarella, L., Van Ranst, E. (2010). Carbon concentrations and stocks in forest soils of Europe. *Forest Ecology and Management*, *260*(3), 262–277.

Bălteanu, D., Chendeș, V., Sima, M., Enciu, P. (2010). A country-wide spatial assessment of landslide susceptibility in Romania. *Geomorphology, 124*(3-4), 102–112.

Bănăduc, D., Mărginean, M., Curtean-Bănăduc, A. (2013). Geographical and human impact elements influence on the fish fauna of the Olteț River (Romania). *Transylvanian Review of Systematical and Ecological Research*, *15*(2), 9–44.

Blujdea, V.N.B., Vinas, R.A., Federici, S., Grassi, G. (2016). The EU greenhouse gas inventory for the LULUCF sector: I. Overview and comparative analysis of methods used by EU member states. *Carbon Management*, *6*(5-6), 247–259.

Boengiu, S., Vlăduț, A., Marinescu, E. (2012). Conditions of gully development within piedmont areas with examples from the western part of the Getic Piedmont, Romania. *Journal of Environmental Biology*, *33*. 407–415.

Cazacu, B.C., Buzgar, N., Iancu, O.G. (2018). Geochemical and Spatial Distribution of Heavy Metals in Forest Soils Adjacent to the Tinovul Mare Poiana Stampei Peat Bog. *Rev. Chim.*, *69*(2), 434–438.

Căbăroiu, G., and Niță, L. (2013). Land quality classes and natural landscape of the mining area Valea Mănăstirii 2, Gorj County. *Research Journal of Agricultural Science*, *45*(4), 14–18.

Chen, S., Martin, M.P., Saby, N.P.A., Walter, C., Arrouays, D. (2018). Fine resolution map of top- and subsoil carbon sequestration potential in France. *Science of the Total Environment*, *630*. 389–400.

Crișan, V.E., Enescu, R.E., Bragă, C. (2017). The main characteristics of forest soils from Buzău forest administration County. *Journal of Horticulture, Forestry and Biotechnology*, *21*(3), 142–146.

Dincă, L., Spârchez, Gh., Dincă, M. (2014). Romanian's forest soil GIS map and database and their ecological implications. *Carpathian Journal of Earth and Environmental Sciences*, *9*(2), 133–142.

Dincă, L., Dincă, M., Vasile, D., Spârchez, Gh., Holonec, L. (2015). Calculating Organic Carbon Stock from Forest Soils. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, *43*(2), 568–575.

Dixon, R.K., Solomon, A.M., Brown, S., Houghton, R.A., Trexier, M.C., Wisniewski, J. (1994). Carbon Pools and Flux of Global Forest Ecosystems. *Science*, *263*(5144), 185–190.

Dumitrescu, D. (2003). Disparități temporal și spațiale în evoluția numeric a populației rurale din Piemontul Cândești. *Analele Universității "Valahia" Târgoviște, Seria Geografie*, *3*. 244–251.

Edu, E.M., Mihalache, M., Ionescu, M. (2013). Determination of organic carbon in forest soils by comparative analysis of methods: Walkley Black Method with the Gogoasa modification versus dry combustion Dumars Method. *Research Journal of Agricultural Science*, *45*(1), 13–20.

Enescu, C.M., Dănescu, A. (2013). Black locust (*Robinia pseudoacacia* L.) - an invasive neophyte in the conventional land reclamation flora in Romania. *Bulletin of the Transilvania University of Brașov, Series II: Forestry Wood Industry Agricultural Food Engineering*, *55*(20), 23–30.

Enescu, C.M., Dincă, L. (2018). Forest soils from Argeş County. *Current Trends in Natural Sciences*, *7*(14), 176–182.

Enescu, C.M., Dincă, L., Bratu, I.A. (2018). Chemical characteristics of the forest soils from Prahova County. *Scientific Papers Series Management, Economic Engineering in Agriculture and Rural Development*, *18*(4), 109–112.

Geambaşu, N., Surdu, A., Dănescu, F., Prigoreanu, C. (2004). Forest soil condition in Romania. Results of transnational grid 16x16km. *Analele ICAS*, *47*(1), 143–164.

Huang, J., Minasny, B., McBratney, A.B., Padarian, J., Triantafilis, J. (2018). The location- and scale-specific correlation between temperature and soil carbon sequestration across the globe. *Science of The Total Environment*, *615*. 540–548.

Ionuş, O., Licurici, M., Boengiu, S., Simulescu, D. (2011). Indicators of the Human Pressure on the Environment in the Bălăciţa Piedmont. Forum geografic. *Studii şi Cercetări de Geografie şi Protecţia Mediului*, *10*(2), 287–294.

Lal, R. (2004). Soil carbon sequestration to mitigate climate change. *Geoderma*, *123*. 1-22.

Lal, R. (2005). Forest soils and carbon sequestration. *Forest Ecology and Management*, *220*(1-3), 242–258.

McFero Grace, J., Skaggs, R.W., Cassel, D.K. (2006). Soil Physical Changes Associated with Forest Harvesting Operations on an Organic Soil. *Soil Science Society of America Journal*, *70*, 503–509.

Montanarella, L., Tóth, G., Jones, A. (2011). Soil Component in the 2009 LUCAS Survey. *Land quality and land use information in the European Union*, 209–219.

Morariu, S.I., Duceac, L.D., Luca, A.C., Popescu, F., Pavel, L., Gavrilescu, C.M. (2018). Soil Chemical Pollution and Aggressive Pathologies. *Rev. Chim.*, *69*(8), 2278–2282.

Orgiazzi, A., Ballabio, C., Panagos, P., Jones, A., Fernández-Ugalde, O. (2018). LUCAS Soil, the largest expandable soil dataset for Europe: a review. *European Journal of Soil Science*, *69*, 140–153.

Panagos, P., Meusburger, K., Alewell, C., Montanarella, L. (2012). Soil erodibility estimation using LUCAS point survey data of Europe. *Environmental Modelling & Software*, *30*, 143–145.

Pleniceanu, V., Alina, V. (2003). Annual variation of the mean monthly quantities of precipitations at the meteorological station of Craiova. *Revista Forum Geografic - Studii şi Cercetări de Geografie şi Protecţia Mediului*, *2*(2), 26–29.

Popescu, C. (2009). Researches on the Structure of the Preluvosoil from the Southern Part of the Getic Piedmont that Was Used Long Term as Arable. *Bulletin UASVM, Agriculture*, *66*(1), 522.

Samuel, A.D., Tit, D.M., Melinte (Frunzulica), C.E., Iovan, C., Purza, L., Gitea, M., Bungau, S. (2017). Enzymological and Physicochemical Evaluation of the Effects of Soil Management Practices. *Rev. Chim.*, *68*(10), 2243–2247.

Spârchez, Gh., Târziu, D.R., Candrea-Bozga, B., Cioc, C.V. (2009). Research regarding the ecological rehabilitation of the landfills resulting from opencast coal mining activities in the north-west of the Getic Plateau. *Revista Pădurilor*, *2*. 23–30.

Stănilă, A.L., Parichi, M., Cruceru, N. (2010). Pedogenetic soil enfranchisement of the relict Getic Piedmont. *Scientific Papers, Series A. Agronomy*, *LIII*. 41–46.

Tanislav, D., Alexe, R. (2012). Hydrological hazards in the Eastern part of the Getic Piedmont. *Proceedings of the Conference "Water resources and wetlands", 14-16 September, Tulcea-Romania*, 238–243.

Tomescu, V. (2005). Natural protected areas for biodiversity within Oltenia and their role in sustainable development. *Geographical Phorum - Geographical Studies and Environment Protection Researches, 4*(4), 116–121.

Tomescu, V., Popescu, L., Ciocan, E., Mara, C., Negreanu, Ş. (2008). Man induced impact on the environment as a result of hydrocarbons' exploitations within the Amaradia hills. *Annals of the University of Craiova - Series Geography*, *11*, 5–18.

Tóth, G., Weynants, M., van Liedekerke, M., Panagos, P., Montanarella, L. (2013). Soil databases in support of pan-European soil water model development and applications. *Procedia Environmental Sciences*, *19*, 411–415.

Tóth, G., Antofie, T.E., Jones, A., Apostol, B. (2016). The LUCAS 2012 topsoil survey and derived cropland and grassland soil properties of Bulgaria and Romania. *Environmental Engineering and Management Journal*, *15*(12), 2651–2662.

Worrell, R., Hampson, A. (1997). The influence of some forest operations on the sustainable management of forest soils - a review. *Forestry*, *70*(1), 61–85.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

STUDY OF PEDOGENETIC PROCESSES IN SOILS LONG IRRIGATED - MONITORING AND PROJECTING THEIR EVOLUTION

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Abstract

The soil and water resources of the Republic of Moldova are subjected to an intense anthropogenic press. As a result, such forms of soil degradation such as surface and deep water erosion, landslides, secondary salinisation and secondary salinisation as a result of irrigation, exasinary destructuring and compacting and others have intensified. The rational use of soil resources is based on the detailed knowledge of the main natural and anthropogenic factors that influence effective fertility and their quality status. In order to assess the change of the soil cover structure over the years, it is necessary to use the thematic materials previously elaborated. It should be noted that they do not fully reflect the complexity of the situation at the moment. This finding refers in particular to processes of soil degradation in basin landscapes (Rozloga, 2013; 2015) where pedogenic factors (soil, parental rocks, hydrology, lithology, hydrogeology and vegetation), interacting in various ways, lead to the emergence of extremely complex degradation forms. The anthropic impact on soil and surface waters is largely determined by the activity of the agrarian and industrial sectors. According to the latest estimates, the chemical composition and the soil and surface water quality indices record negative changes. This concerns (Filipciuc, 2007; Simakova et al., 2008), first of all, the agricultural soils (land) that are involved in the irrigation systems and the increase of the degree of mineralization, the ionic reaction and ionic composition of the inland river waters, and to the accumulation basins (lakes, ponds). All these constitute the limiting factor of using soils for irrigation with surface waters. In order to develop complexes of measures to prevent land degradation it is necessary to know the spatial spread and intensity of the manifestation of these processes on a more detailed scale. The achievement of this objective is possible with the use of the Geoinformatics System (GIS), which allows the operative and reasoned approach to inventory, analysis, planning and design issues (Sorokina, 2006). This system creates prerequisites for the development and implementation of measures to combat land degradation at a new quality level aimed at preserving and improving soil fertility.

Key words: *GIS, irrigation, pedogenic processes, soil.*

INTRODUCTION

The agro-industrial complex of the Republic of Moldova operates under risk conditions. By geographic location the territory of the republic falls within the area with insufficient and unstable humidity. The annual amount of atmospheric precipitation varies between 380 and 550 mm. Deposits register a marked decline from Northwest to South East (Rozloga, 2015; Andrieș, 2007). In the northern part of the republic, the volume of rainfall is attested as submijlociu, in the central pedoclimatic zone this indicator is considered to be low and in the south - as very low.

A peculiarity of the republic's climate is the phenomenon of drought. According to the analysis of observations data over the past 100 years, strong and long droughts occur over three years in northern and central areas and over two years in the southern area (Gavrilitsa, 1993). The result of this climate phenomenon is the reduction or compromise of agricultural production.

Through long-term research, it has been established that the main natural limiting factor in obtaining high and stable crops is low availability of affordable water plants (Andrieș, 2007). Under the conditions of the Republic, one of the most effective measures for optimizing the soil moisture regime is irrigation. Obviously, it does not exclude the application of agro-technical processes to water the soil.

Soil irrigation as a method of improving the water regime has been known since ancient times, but to date there are a number of complicated problems related to the reaction of some soil types to the change of the water regime and to the quality of the water used for irrigation. Thus, irrigation of chernozems with mineralized water (> 3000 mg/l) does not cause essential changes in the soil adsorption complex and does not lead to secondary salinisation (Kovda, 1981; Simakova et al., 2008). At the same time, the same author points out that the use of water with a degree of mineralization higher than 1000 mg/l on chernozem soils leads to secondary salinisation and solonization, but also their hydromorphic evolution. Because of the multiannual researches, V.A. Covda (Kovda, 1981; Sorokina, 2006) concludes that the development of water quality indicators for unified irrigation is virtually impossible due to the extremely wide variation in the pedological, geomorphological and geochemical conditions of the irrigated territories.

In the Republic of Moldova, chernozems make over 74% of the surface of irrigated soils (Filipciuc, 2007; 2014). They can be included in the irrigation regime only when using water with low salt content (up to 1000 mg/l) and a favorable chemical composition. Research on different chernozem subtypes shows that changing the natural water regime through the irrigation system, even with the use of good quality water, leads to structural degradation, compaction of the upper horizon and decalcification (Krupenikov et al., 1979; 1981; Podymov, 1976).

The use of highly mineralized water or inappropriate chemical composition in the irrigation of chernozems results in their salinisation and/or secondary solonization (Poznyak, 1997; Filipciuc et al., 1990; Filipciuc, 2014). In this context, it is useful to note that the process of solonization severely affects not only the physical and physico-chemical properties of the soil. It also produces degrading effects on the mineralogical composition by increasing the content of inflatable material in the smectite and illit-smectite group (Gavrilitsa, 1993). Some secondary soil processes induced by irrigation of chernozem with unsatisfactory quality water, such as clay, peptization of fine clay and illitisation, are irreversible and cannot be restored or restored by means of melioration.

Contrary to these findings, some authors argue that chernozem soils can be irrigated with water in which the soluble salts content is 1500-3000 mg/l (Rabochev, 1981; Bezdnina, 1990; Zimovets, 1993; Prikhodko, 1996).

V. Kovda, through the analysis of the global experience of the irrigation consequence, points out that the unification of water quality indicators for irrigation and the setting of limit values presents great difficulties or is impossible due to the diversity of climatic, geomorphological conditions. For the soil and climatic conditions of the Republic of Moldova, water quality indicators used for irrigation regulating the degree of mineralization, the reaction, the sodium adsorption ratio, the magnesium indicator, the chlorine content and the residual sodium carbonate (Filipciuc, 2007) have been established.

MATERIALS AND METHODS

In order to establish and evaluate the impact of irrigation of chernozem soils with water of different quality, land, laboratory and office methods were used.

On the ground, the following works were carried out: the opening of the soil profiles, the morphological and morphometric description; collecting soil samples from genetic horizons for the determination of chemical, physical, hydric and physico-mechanical properties.

Collecting water samples for irrigation in order to determine chemical composition and quality indicators;

The laboratory determined the hygroscopic water content; solid soil phase density; granulometric and microaggregate composition; structurally-aggregate structure and structure's hydrostability; hygroscopicity and cheering coefficients; the content and composition of humus; the content of calcium and magnesium carbonates; nutrient content of nitric nitrogen, mobile phosphorus and exchangeable potassium; the soluble salt content, the reaction and the ionic composition of the aqueous extract; the content of adsorbed cations; chemical composition and water quality indicators for irrigation.

At the office stage the calculations of the field and laboratory determinations were carried out, systematized and analysed the results obtained and elaborated the report for the current year stage.

RESULTS AND DISCUSSIONS

The works covered by the thematic plan were carried out on experimental polygons selected within irrigation systems and their immediate proximity (non-irrigated soils). The soil cover of the polygon in Egoreni commune, Soroca rayon, is represented by deeply humorous, humorous, humorous chernozem. The polygon is located on the first terrace of the Dniester River. The carbonate chernozem is irrigated with this river's water for 47 years. The experimental polygon in Singerei is located on a right slope with an east exposure and with a slope of 2-3%. Soil is a habitually profoundly moderate clay-humus humerus. Irrigated with pond water for 13 years. The experimental polygon in Cozăşti commune Singerei district is situated on a south-eastern slope with a slope of 2%. The soil is presented by the habitus of deeply silky clayey humerus. It has been under irrigation for 9 years. The water used is deep water with unfavorable chemical composition and strong alkaline reaction.

The evolution of secondary pedological processes and changes in physical and chemical properties in irrigated soils were determined by the "pairs profiles" method (irrigated soil - non-irrigated soil). This method is widely used in the pedoameliorative study and is considered practicable in the quantitative determination of soil characteristics (Poznyak, 1997; Ropot, 1991). Field determinations and laboratory analyses performed to determine irrigation-induced changes were performed using the methods.

Table 1. The chemical composition of the Dniester (Nistru) river water

Indicators	Unit of measurement	The content		
		minimum	Maximum	Medium
Mineralization	mg/l	240	670	455
The reaction (pH)	Units	7.53	8.45	7.99
CO_3^{2-}	me/l	0.07	0.84	0.46
HCO_3^-	me/l	2.49	4.92	3.71
Cl^-	me/l	0.57	3.10	1.84
SO_4^{2-}	me/l	1.00	3.33	2.16
Ca^{2+}	me/l	2.60	4.50	3.55
Mg^{2+}	me/l	0.58	3.33	1.96
Na^+	me/l	0.70	3.26	1.98

Table 2. The composition and quality indices of the Dniester (Nistru) river (year 2018)

Period	Mineralization, mg/l	pH	HCO_3^-	Cl^-	SO_4^{2-}	Ca^{2+}	Mg^{2+}	Na^+	SAR	P_{Mg}, %
			me/l mg/l							
May	496	8.31	3.42 209	1.60 57	1.40 67	3.72 74	1.20 14	1.50 34	0.9	24
August	240	7.95	2.60 159	1.16 41	0.31 15	3.30 66	0.12 2	0.65 15	0.5	19
September	254	8.05	2.80 171	1.36 48	0.23 11	3.02 61	0.72 9	0.65 15	0.5	17
Admissible values										
	< 1000	< 8.30	-	<3.00	-				<3.0	<50

Depending on the climatic conditions, especially the rainwater regime, the degree of mineralization and the chemical composition of the Nistru river water are marked by significant changes (Table 1).

Soluble salt content ranges from 240 to 670 mg/l over a multi-year cycle. The water reaction is judged to be slightly alkaline with pH values ranging from 7.53 to 8.45 units (Florea, 1976).

One of the main qualitative indicators of surface water in the Republic, used for irrigation, is the magnesia index. For water from the Dniester river, between May and September 2018, it oscillated between 17 and 24%. The calculated values of this indicator show that the possibility of the adsorption process of the magnesium cation by the colloidal soil complex in appreciable quantities is virtually excluded.

By chemical composition and quality indicators the water of the Nistru river is appreciated as good - very good for irrigation (Krupenikov, 1983). It can be used without limitation for irrigating any type of soil.

However, it should be noted that under certain thermal conditions, usually at high temperatures, or during the abundant development of phytoplankton, the water in the Dniester is potentially alkaline, and in its composition appears the CO_3^{2} anion. In such transformations, the water intensifies the decalcification process of the soil and changes the ratio of the exchangeable cations in the adsorbent complex. Such degrading impact of the Nistru river water was recorded at the Caragaşi irrigation system (Orlov, 1980).

During the reference year, the Dniester river water analysis was carried out in three rounds. The determinations show that the total content of soluble salts is low and compose 240-496 mg/l. According to river classification by degree of mineralization, sweet to low salted (Orlov, 1980). By mineralization, water used for irrigation does not pose a risk of secondary salinisation of the soil (Table 2). In the soluble salt composition predominates calcium compounds, which make up 58-71% of the dry residue (Annex 1). Toxic sodium and magnesium salts have a subordinate role.

Table 3. The main agrochemical properties of chernozem carbonate

Deph, cm	Humus, %	Humus reserve, t/ha	$CaCO_3$, %	N-NO_3	P_2O_5	K_2O
				me/100 g sol		
Not irrigated soil						
0-25	2.18	68	3.8	0.45	1.05	19.6
25-50	2.11	67	3.8	0.29	1.19	18.8
50-84	1.88	86	6.1	0.34	0.82	14.6
84-106	1.20	36	8.7	0.26	0.80	14.6
106-130	0.87	25	10.9	0.26	0.75	14.5
130-160	0.62	26	11.8	0.22	0.98	12.9
Irrigated soil						
0-28	2.56	90	2.3	0.45	1.85	20.8
28-48	2.58	71	3.5	0.34	0.89	19.6
48-76	2.16	83	5.4	0.32	1.02	19.6
76-107	1.38	58	8.8	0.36	0.69	14.6
107-138	1.02	43	10.7	0.31	0.65	14.5
138-180	0.81	46	12.0	0.30	0.82	13.8

Table 4. Changing the humus composition as a result of irrigation (plowing horizon)

C total, %	C, %			$\frac{C_{AH}}{C_{AF}}$	C faction AH,%		C in the residue, %
	Extract with $Na_4P_2O_7$+NaOH	AH	AF		free and bonded with R_2O_3	bonded with Ca	
Not irrigated soil							
1.73	0.88 / 50.9	0.63 / 36.4	0.25 / 14.4	2.5:1	3.2	96.8	0.85 / 49.1
Irrigated soil							
1.75	0.83 / 47.4	0,57 / 32.6	0.26 / 14.9	2.2:1	6.0	94.0	0.92 / 52.6

The water reaction is slightly alkaline with pH variations from 7.95 to 8.31 units.

Chlorine content is well below the maximum permissible limit of this element in irrigation water, being only 1.16-1.60 me/l.

The sodium adsorption ratio is very low with values ranging from 0.5 to 0.9, so the water does not pose a risk of soil degradation by secondary solonization.

The effect of long-term irrigation on the chemical and physico-chemical properties of carbon black chernozem

The main agrochemical properties of the non-irrigated carbon black chernozem and of the irrigation regime were determined on the genetic horizons of the soil profiles. The obtained results show that both soil variants are characterized by a medium content of organic matter in the plowed and substrate layers, this being 2.11-2.58%. In the transition horizons the humus content decreases slowly to 1.20-2.88%, being appreciated as low. The chernozem curing gradient contains 0.62-0.81% humus, a very low value. It should be noted that the distribution of humus on the irrigated soil profile is more uniform and its content is slightly higher compared to the non-irrigated soil. Reserves of organic matter in irrigated soil are also higher.

The investigated soil variants contain calcium and magnesium carbonates from the surface in average quantities of 2.3-3.8%. In depth the carbonate content gradually increases, reaching maximum values of 11.8-12.0% in the rock (Table 3).

By determining the available nutrients it was found that in the upper horizons of the non-irrigated soil the nitric nitrogen is contained in small amounts of 0.29-0.40 mg/100 g of soil and slowly decreases to 0.22 mg/100 g in depth. The distribution of N-NO3 on the irrigated soil profile is much more uniform. Thus, in the humus-accumulative horizon the nitric nitrogen content is 0.34-0.45 mg/100 g of soil. In the transition horizons and in the parental rocks he recorded an insignificant decrease of up to 0.30 mg/100 g of soil.

Table 5. The salt content, the reaction and the ionic composition of the aqueous extract of the carbonate chernozem

Depth, cm	Dry residue, %	pH	HCO_3^-	Cl^-	SO_4^{2-}	Ca^{2+}	Mg^{2+}	Na^+	$\frac{Ca+Mg}{Na}$	K_{SO}
			me/100 g soil							
Not irrigated soil										
0-25	0.046	8.30	0.51	0.06	0.17	0.63	0.07	0.04	18	0.7
25-50	0.044	8.25	0.54	0.07	0.14	0.63	0.08	0.04	18	0.8
50-84	0.043	8.15	0.55	0.07	0.14	0.66	0.06	0.04	18	0.8
84-106	0.040	8.05	0.55	0.06	0.10	0.56	0.09	0.06	16	0.8
106-130	0.051	8.10	0.52	0.16	0.15	0.58	0.17	0.08	9	0.8
130-160	0.053	8.15	0.60	0.11	0.18	0.60	0.15	0.14	5	0.8
Irrigated soil										
0-28	0.045	8.20	0.56	0.08	0.16	0.60	0.16	0.04	19	0.7
28-48	0.048	8.10	0.65	0.09	0.15	0.70	0.12	0.07	12	0.8
48-76	0.038	8.15	0.62	0.07	0.05	0.60	0.10	0.04	18	0.9
76-107	0.046	8.20	0.55	0.07	0.22	0.58	0.20	0.06	13	0.7
107-138	0.046	8.25	0.60	0.08	0.12	0.58	0.14	0.08	9	0.8
138-180	0.053	8.25	0.54	0.13	0.23	0.64	0.16	0.10	8	0.7

Table 6. The content of cation exchangeable in carbonate chernozem

Depth, cm	Ca^{2+}	Mg^{2+}	Na^+	Sum	Ca^{2+}	Mg^{2+}	Na^+
	me/100 g soil				% from sum		
Not irrigated soil							
0-25	21.17	2.11	0.55	23.83	89	9	2
25-50	21.06	2.11	0.19	23.36	90	9	1
50-84	19.16	2.21	0.36	21.73	88	10	2
84-106	17.92	2.58	0.36	20.86	86	12	2
106-130	15.76	3.97	0.19	19.92	79	20	1
130-160	12.85	4.85	0.36	18.06	71	17	2
Irrigated soil							
0-28	21.85	2.00	0.48	24.33	90	8	2
28-48	21.50	1.50	0.48	23.48	92	6	2
48-76	19.75	2.00	0.48	22.23	89	9	2
76-107	18.00	1.50	0.48	19.98	90	8	2
107-138	15.50	2.00	0.54	18.04	86	11	3
138-180	15.00	3.35	0.48	18.83	79	17	4

The content of mobile phosphorus is small to medium. After the exchangeable potassium content both soil variants are appreciated as moderately insured.

It is worth mentioning that irrigation of carbonate chernozem with water from the Dniester River influences the composition of humus (Table 4). It has been found that the irrigated soil reduces the carbon content in the humic acids fraction. This led to a reduction in the ratio of CAH: CAF from 2.5: 1, the humus type being the humatic one. Another change in the composition of organic matter refers to the increase in the fraction of free humic acids or bound to iron and aluminum sequestrants.

Therefore, the irrigation of the carbonated chernozem with water from the Dniester is increasing humus mobility.

This trend has been observed in other irrigation facilities (Poznyak, 1997; Novikova, 2009; Novikova et al., 2007).

Irrigation use of good quality water for 47 years does not essentially alter the saline indices of carbon black chernozem. From the results we can see that the anionic composition is clearly predominated by HCO_3^- (0.51-0.64 me), followed by SO_4^{2-} (0.12-0.23 me). Chlorine content is small and accounts for 0.06-0.13 me/100 g of soil. It is important to emphasize that in soil solution (irrigated and non-irrigated) prevails Ca^{2+} with a content of 0.56-0.70 me.

Annex 1

The composition of the soluble salts of irrigation water

Water source	Collection period	Na_2CO_3	$Ca(HCO_3)_2$	$Mg(HCO_3)_2$	$NaHCO_3$	$CaSO_4$	Na_2SO_4	NaCl	$MgCl_2$	$CaCl_2$
		me/l								
River Nistru	May	-	3.42	-	-	0.30	1.10	0.40	1.20	-
	July	-	2.60	-	-	0.31	-	0.65	0.12	0.39
	September	-	2.80	-	-	0.22	0.01	0.64	0.72	-
Lake	May	2.40	4.60	10.70	-	-	23.60	1.83	1.90	-
	September	4.80	7.34	7.66	-	-	33.92	-	4.41	-
Underground	May	4.80	1.08	2.44	1.30	-	2.87	1.73	-	-
	September	4.80	1.00	2.22	0.60	-	4.85	1.49	-	-

Annex 2

The composition of the soluble salts of carbonate chernozem

Depth, cm	$Ca(HCO_3)_2$	$CaSO_4$	Na_2SO_4	$MgSO_4$	$MgCl_2$
	me/100 g sol				
Not irrigated soil					
0-25	0.51	0.12	0.04	0.01	0.06
25-50	0.54	0.09	0.04	0.01	0.07
50-84	0.55	0.11	0.03	-	0.07
84-106	0.55	0.01	0.06	0.03	0.06
106-130	0.52	0.06	0.08	0.01	0.16
130-160	0.60	-	0.14	0.04	0.11
Irrigated soil					
0-28	0.56	0.04	0.04	0.08	0.08
28-48	0.65	0.05	0.07	0.03	0.09
48-76	0.62	-	0.04	0.03	0.07
76-107	0.55	0.03	0.06	0.13	0.07
107-138	0.58	-	0.08	0.06	0.08
138-180	0.54	0.10	0.10	0.03	0.13

Magnesium cations are contained in 0.06-0.20 meters, and Na^+ does not exceed 0.04-0.14 me/100 g soil. The ratio between bivalent and monovalent cations shows a significant decrease in soil profile at 19: 1 in the 5: 1 horizon in parental rock (Table 5). The sodium ratio in non-irrigated and irrigated soil is very stable on the soil profile, ranging from 0.7 to 0.9. Thus, during 47 years of irrigation in carbon black chernozem, no soda formation conditions are recorded (Krupenikov et al., 1981).

Determination of soluble salt content shows that this indicator does not change when using Dniester water. The profile of both dry soil variants ranges between 0.038 and 0.053%, characteristic for the subtype given by chernozem (Shaimukhamedov et al., 1990).

The actual soil response is estimated to be slightly alkaline with pH values of 8.05-8.30 units. By determining the composition of the soluble salts, it has been established that their main (predominant) components, both in the non-irrigated and the irrigated soil, are represented by Ca $(HCO_3)^2$ and $CaSO_4$. These compounds are harmless to soil and plants and make up 71-84% of dry residue (Annex 2).

One of the main indicators of the irrigated soils' soil condition is the changeable cation content. The impact of water quality on irrigation on the bases of exchange is very defiant. It is reported that when applying freshwater, the main changes refer to the increase in Mg^{2+} content and to the change in cation ratio. The research did not confirm this finding (Table 6).

The content of adsorbed calcium forms in the horizon of 21.17-21.85 me and gradually decreases in the transition horizons. In the rock of solification this cation has values of 12.85-15.00 me.

The magnesium cation distribution on the soil profiles is inverse. Its contents register an increase from 2.00-2.11 me in humus-accumulative horizon up to 3.35-4.85 me/100 g soil in parental rock. Adsorbed salt constitutes 1-2% of the sum of the bases and in the irrigation soil only it increases to 4% of the amount. According to the scale of the degree of irrigation of irrigated soils, the carbonate chernozem is certified as very weakly solonised.

CONCLUSIONS

The obtained results show that both soil variants are characterized by a medium content of organic matter in the plowed and substrate layers, this being 2.11-2.58%. In the transition horizons the humus content decreases slowly to 1.20-2.88%, being appreciated as low. The chernozem curing gradient contains 0.62-0.81% humus, a very low value. It should be noted that the distribution of humus on the irrigated soil profile is more uniform and its content is slightly higher compared to the non-irrigated soil. Reserves of organic matter in irrigated soil are also higher.

The investigated soil variants contain calcium and magnesium carbonates from the surface in average quantities of 2.3-3.8%.

Irrigation use of good quality water for 47 years does not essentially alter the saline indices of carbon black chernozem. From the results we can see that the anionic composition is clearly predominated by HCO_3^- (0.51-0.64 me), followed by SO_4^{2-} (0.12-0.23 me). Chlorine content is small and accounts for 0.06-0.13 me/100 g of soil. It is important to emphasize that in soil solution (irrigated and non-irrigated) prevails Ca^{2+} with a content of 0.56-0.70 me.

It has been found that the irrigated soil reduces the carbon content in the humic acids fraction. This led to a reduction in the ratio of CAH: CAF from 2.5: 1, the humus type being the humatic one. Another change in the composition of organic matter refers to the increase in the fraction of free humic acids or bound to iron and aluminum sequestrants.

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REFERENCES

Agroclimatic resources of the Moldavian SSR (1982). L.: Gidrometizdat (pp.198). Chisinau.

Alekseev, V.E., Filipchuk, V.F., Arapu, K.G. (1985). Transformation of clay minerals and the properties of greenhouse soils under the influence of irrigation water and chemical ameliorants. In: *Composition, properties and agroecological assessment of Moldavian soils* (pp. 80–93). Chisinau.

Andrieş, S. (2007). Optimization of soil nutrient regimes and productivity of crop plants (pp. 374). Chisinau.

Bezdnina, S.Ya. (1990). Regulation and improvement of irrigation water quality. In *Agropromizdat*, *Improving the quality of irrigation water.* (pp. 4–11). Moscow.

Ecological requirements for soil irrigation in Russia. (1996). M., 71.

Irrigation and Drainage Bulletin. (1976). FAO, 29. 56.

Gavrilitsa, A.O. (1993). Modern problems of sprinkling. Chisinau: Type. *SAUM*, 338.

Kovda, V.A. (1981). Soil cover, its improvement, use and protection. *M. Science*, 173.

Krupenikov, I.A., Podymov, B.P. (1978). Diagnostics and principles of classification of irrigated soils. In

Sat: *Soil-reclamation problems of irrigated agriculture*. Chisinau. *Science*, 8–13.

Krupenikov, I.A., et al. (1981). Statistical parameters of the composition and properties of Moldavian soils. Chisinau. *Science*, *2*. 251.

Krupenikov, I.A. (1983). New representatives of soils of chernozem type. *Pedology*, 6. 3-6.

Filipciuc, V.F. (2007). Soil and water supply to irrigation. In: *Drought and methods to minimize adverse consequences*. Chisinau, 10–11.

Filipciuc, V.F., et al. (1990). Ecopedological monitoring report (pedoameliorative). Edition I. Chisinau. *Agroinformreclama*, 50.

Filipciuc, V.F. (2014). Black soil degradation during irrigation and fertility restoration methods. In: *Natural and anthropogenic factors influencing the quality of soils and water resources of the Republic of Moldova*. Chisinau, 108–136.

Florea, N. (1976). Geochimia and the capitalization of the waters of the Northeastern Romania Plain. Bucharest. Ed / Romanian Academy, 201.

Orlov, D.S., Anikanova, E.M., Markin, V.A. (1980). Features of the organic matter of irrigated soils. In: *Problems of soil irrigation Chernozem zone*. *M. Science*, 136.

Novikova, A.V. (2009). Theoretical aspects of the prediction of secondary salinization, alkalization and co-expression in irrigated soils. In. *Studies of saline and sodic soils: genesis, melioration, ecology*. Kharkov, 464–519.

Novikova, A.V., Gavrilyuk, N.E. (2007). Sod manifestation and alkalization in the rainfed and irrigated soils of the south of Ukraine. Kharkov, 79 p.

Podymov, B.P. (1976). Moldavian soil and water in connection with the development of irrigation. In: *Agriculture of Moldova*. Chisinau, 5. 26–27.

Poznyak, S.P., Tortik, N.I. (1997). Irrigated chernozems of the southwest of Ukraine. Lvov, VNTL, 239.

Prikhodko, V.E. (1996). Irrigated steppe soils: functioning, ecology, productivity. M.: Intellect, 180.

Rabochev, I.S. (1981). Classification of mineralized water according to suitability for irrigation. - *Abstracts of the VI delegate meeting of the All-Union Society of Soil Science*. Tbilisi, 5. 64–65.

Ropot, V.M., et al. (1991). Problems of quality, use and protection of water resources of the SSR Moldova. Chisinau. *Science*, 282.

Rozloga, Iu. (2013). The spatial positioning of the land infrastructure within the upper Cogilnic catchment area with the use of GIS. In: *"Moldova's chernozems - evolution, protection and restoration of their fertility"*, Chisinau, "Recklama" Printing House, 290–295.

Rozloga, Iu. (2015). Application of the geoinformatics system for establishing and evaluating erosion processes. In: "*Akademos" Magazine of Science, Innovation, Culture and Art*, Chisinau: ASM, 4. 66–74.

Simakova, M.S., Rukhovich, D.I., Belobrov, V.P., and others. (2008). Guidance on medium-scale mapping of soils based on GIS. M.: Soil Institute in the name the V.V. Dokuchaeva, 243.

Shaimukhamedov, M.Sh., Kurakulov, S.N. (1990). The reaction of the exchange of Ca-Na in black soil and forecasting the effect of irrigation water on some of their properties. *Pedology*, 3. 88–103.

Sorokina, N.P. (2006). Methodology for compiling large agro-ecologically oriented soil maps. M.: Russian Agricultural Academy, 160.

Zimovets, B.A. et al. (1993). Methods for assessing and studying the suitability of water for irrigation. Moscow: Publishing house GOSNITI, 64.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

VERMICOMPOST PRODUCTION AND ITS IMPORTANCE FOR SOIL AND AGRICULTURAL PRODUCTION

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Abstract

The vermicompost (earthworm humus) is a particularly effective fertilizer for both vegetables and fruits, as well as for large crops. This fertilizer has, in a high percentage all of the 16 nutrients that are essential for plants. The vermicompost is obtained through the aid of Eisenia fetida (Savigny), also known as: "compost earthworm", "red earthworm", "redworm" "brandling worm". Eisenia fetida is a species of earthworm developed by researchers and geneticists in the 1950's and 1960's, having a much higher reproductive capacity and better adaptability to the living environment than other existing species. It was first used in the USA in farms where large quantities of biomass and natural fertilizers were needed. In addition, this species of earthworm processes large amounts of biodegradable organic waste more rapidly with maximum productivity of natural humus. Thanks to these benefits, farmers around the world began to grow earthworms and produce vermicompost in a growing number. One of the most important advantages of using the vermicompost is that it increases production, both in terms of quantity and quality.
Another advantage is that it helps in the suppression of harmful fungi in the soil. These microorganisms produce hormones, vitamins, nutrients, enzymes, amino acids and minerals that are important for plants.
The vermicompost has a positive effect not only on plants but also on the soil. It greatly improves the structure of the soil, decreasing its density, increasing the aeration and the absorption of nitrogen from the atmosphere. Therefore, it repairs the soils affected by the long-term use of chemical substances.

***Key words**: eartworms, vermicompost, nutrients, soil fertility.*

INTRODUCTION

"Nobody and nothing can be compared with earthworms and their positive influence on the whole living Nature. They create soil and everything that lives in it. They are the most numerous animals on Earth and the main creatures converting all organic matter into soil humus providing soil's fertility and biosphere's functions: disinfecting, neutralizing, protective and productive" (Igonin, 2004).
Aristotle called the earthworms "intestines of the earth" and Charles Darwin wrote a book about earthworms and their activities, saying that there may not be another creature that played such an important role in the history of life on Earth (Bogdanov, 1996).
Vermiculture is the culture of earthworms. Its goal is to continuously increase the number of earthworms in order to achieve a sustainable population. The earthworms are either used to expand a vermicompost operation or are sold to customers who use them for the same or for other purposes (Munroe, 2007).
Vermicomposting is the process where earthworms are used to convert organic materials (usually waste) into humus-like material known as vermicompost (Munroe, 2007).

MATERIALS AND METHODS

It is estimated that there are over 1800 species of earthworms worldwide (Edwards & Lofty, 1972), but only one species, *Eisenia fetida* (Savigny), also known as: "*compost earthworm*", "*red earthworm*", "*redworm*" "*brandling worm*" is being discussed in this paper.
Eisenia fetida is a species of earthworms developed by researchers and geneticists in the 1950's and 1960's, with a much higher reproductive capacity and a better adaptability to the living environment than the existing species. It was first used in the USA on farms where large amounts of biomass and natural fertilizers were needed. In addition, this earthworm species processes much more

rapidly large amounts of biodegradable organic waste with a maximum productivity of natural humus. Due to these benefits, farmers around the world began to grow earthworms and to produce vermicompost in a growing trend.

In the present, the largest vermicompost production centers are in India and Cuba, countries that are currently leaders in this field. In India, it is estimated that around 200,000 farmers practice vermicomposting and a network of 10,000 farmers produces 50,000 metric tons of vermicompost each month. Over the last decade, farmers in Australia and the USA'a West Coast began to use vermicompost in large quantities, fueling the development of the vermicompost industry in these regions. At the same time, scientists from several universities in the USA, Canada, India, Australia and South Africa began to document the benefits associated with the use of the vermicompost, providing facts and figures to support the observations of those who actively used the material (Munroe, 2007).

Composting earthworms live in "nests" estimated at 100,000 individuals (Figure 1).

Figure 1. Soil composting with earthworms

They are hermaphrodites, and they double their number in a cycle of around 100 days. The nest has a surface of 2 m^2 and a thickness of 25 cm. For a nest it is necessary to have an area of 2 m^2, in the shape of an alley, on which the bedding is formed.

Preparing the bedding

One of the main activities in raising earthworms is bedding preparation. The substrate of the bedding is a layer of organic materials rich in cellulose, where the earthworms build their habitat from where they will go to the top to feed themselves (Duşan, 2013). Over this substrate there is the food (manure) layer, which should not be disturbed during the earhworms' habitation (Figure 2).

Figure 2. Preparing the bedding

The bedding's substrate must meet two basic requirements:

a. To have the property of maintaining moisture (the earthworms require a high degree of moisture in the environment in which they live and multiply because they breathe through their skin and take oxygen out from the water;
b. It must be porous, aggregated soil represents the amorphous surface in the bedding and it prevents the penetration of oxygen into the substrate (Duşan, 2013).

If available, shredded paper or paperboard are an excellent bedding material (Georg, 2004), especially when combined with typical organic farm resources, such as straws and hay.

Optimum oxygen quantities facilitate the decomposition of garbage into nutrients. A well-fed earthworm in a well-protected substrate lives peacefully and multiplies intensely producing cocoons with more hatchlings in them (a cocoon contains 10-12 hatchlings) (Duşan, 2013).

It is very important that the substrate, in addition to being aerated, it should also allow the formation of bacteria, which make the primary food preparation, without which, regardless of the quantity or quality of food, the earthworms die (Duşan, 2013).

The natural habitat for earthworms is, in general, as known, given by the deposits of manure and vegetal residues. All the research and experience gained so far shows that the more the artificial habitat resembles the natural one, the more the success in growing earthworms will be greater. The best earthworm food and the best biohumus are obtained if manure is given (Table 1) from horses, cows, sheep, rabbits, goats, provided that it is pre-composted and that it passed the fermentation phase.

Table 1. Common Worm Feed Stocks (Munroe, 2007)

Food	Advantages	Disadvantages	Comments/Notes
Cattle manure	Good nutrition, natural food, therefore it requires little adaptation	Weed seeds make pre-composting necessary.	The manure is partially decomposed and therefore ready for consumption by the earthworms.
Poultry manure	High nitrogen content, has as result a good nutrition and a high-value product	High levels of protein can be dangerous for earthworms, so they should be used in small quantities; there is a need for a major adaptation process for the earthworms that are not used to this raw material. It can be pre-composted but it's not a must, if used with caution.	Gaddie and Douglas (1975) suggest that poultry waste is not suitable for earthworms, because it is too "hot"; however, researchers from Nova Scotia (Georg, 2004) have shown that earthworms can adapt if the original proportion of poultry manure in the bedding is 10% of the total volume or less.
Sheep/Goat manure	Good nutrition	Requires pre-composting (due to weed seeds); small-sized particles can lead to agglomeration, therefore it requires additional bulking materials.	With the right additives to increase the Carbon/ Nitrogen ratio, this fertilizer is also good as a bedding.
Pig manure	Good Nutrition; produces an excellent vermicompost	It is ually in liquid form, it should therefore be dehydrated or used with large amounts of highly absorbent bedding.	Scientists from Ohio State University have discovered that vermicompost made from pig manure has outperformed all other vermicomposts and commercial fertilizers as well.
Rabbit manure	The nitrogen content is similar to that of poultry manure, so it provides good nutrition; it contains a very good mixture of vitamins and minerals and can be an ideal food for earthworms (Gaddie and Douglas, 1975)	It must be leached before use due to the high urine content; it can overheat if the quantities are too high, it's also not usually available.	Many rabbit breeders in the USA place the earthworms under the rabbit cages in order to catch the pellets as they fall through the wire mesh cage floors.

In the earthworm's diet there can also be used: fresh food scraps (egg shells, other waste from food preparation, food waste from commercial food processing), pre-composted food waste, biosolids (human waste), seaweed, legume hays, grains (feed mixtures for animals, such as poultry feed concentrates), corrugated cardboard (including waxed cardboards), fish, poultry meat residues, blood waste, dead animals. If the resulting vermicompost is used in organic farming, some of the residues listed above can not be used in earthworm diet.

Earthworm care

Earthworms usually do not get ill (they are not attacked by viruses and bacteria), so the only enemy that can cause them harm can only prove to be man through its negligence. Their care requires little work at fairly long intervals, therefore, in case of forgetfulness or other reasons, they can remain unfed (food is given every 15 days), or even dehydrated below the optimal humidity level.

Humidity

In regards to moisture, the range within which the ideal moisture content can be found for materials in conventional composting systems is between 45-60% (Rink et al., 1992), but in contrast to the conventional practice, the ideal humidity range for vermicomposting or the vermiculture process is of 70-90%. In this wide range, researchers found slightly different optimal values: Dominguez & Edwards (1997) found that the best range would be of 80-90%, with 85% being optimal, while researchers in Nova Scotia found out that 75-80% humidity produced the best growth and the highest reproduction rate (Georg, 2004). In Romania, Duşan (2013) found that the optimal humidity would be of 82.5% (Table 2), or, if empirically controlled, it can be tested by collecting the manure in the hands and squeezing it, this action should result in a few drops running through the fingers, if the humidity level is appropriate.

Aeration

Aeration is a basic factor in growing earthworms by allowing the air to enter their habitat. Therefore, it should be taken into consideration, when preparing the bedding, for the bedding material to be porous, and if there are cases in which aggregated material is used as food, then this material should be placed either in the middle or at the edge of the bedding in order to allow ventilation through the sides. The earthworms should be disturbed as little as possible, but once a month, the first layer at the surface, down to 8-10 cm should be aerated with a fork, while taking care not to go deeper than that (Duşan, 2013).

Table 2. Action of earthworms in the habitat, depending on the pedoclimatic conditions (Duşan, 2013)

	Senescence	Lethargy	Produces only humus	Reproduction reduced to a minimum	Optimal conditions	Reproduction reduced to a minimum	Produces only humus	Lethargy	Death
pH level	6<	6	6.5	6.8	7	7.2	7.5	7.8	<7.8
Temperature (°C)	0<	0-7	7-14	14-19	19-20	20-27	27-33	33-42	<42
No. of hatchlings in the cocoon		-	-	2-10	10-20	2-10	-	-	
Humidity (%)	70<	70	75	80	82.5	84	88	90	<90

As far as the pH level is concerned, the best range would be of 6.8-7.2 with a neutral reaction, if the pH is too acidic, their acidity can be reduced by the scattering of calcium carbonate, and if it is alkaline, it can be reduced by washing with water or by adding grounded peat (Duşan, 2013). Other researchers say that earthworms can survive in a pH range of 5 to 9 (Edwards, 1998). Eitherway, most experts believe that earthworms prefer a pH of 7 or slightly higher, but those in Nova Scotia have found that the range of 7.5 to 8 is the optimum range (Georg, 2004).

In regards to the salt content, earthworms are very sensitive to sodium, preferring a sodium content of less than 0.5% (Gunadi et al., 2002).

Temperature

The optimal temperature in which the earthworms are most active is around 20°C, respectively between 15°C and 20°C. At temperatures below 0°C the earthworms die but the cocoons can withstand up to -20°C and can withstand this temperature for a long time until the conditions allow the hatchlings to come out. If the temperature is too high, of 30°C and above, it will cause the earthworms to leave the bedding and if they cannot leave it, they will die (Munroe, 2007).

Stimulating the reproduction

When the earthworm population's density becomes higher than 5 kg/m^2 the need for reproduction begins to slow and a fierce competition for food and space occurs. Although it is possible to obtain a population density of even 20 kg/m^2 (Edwards, 1999), the most common densities found in practicing vermicomposting are between 5-10 kg/m^2. Earthworm growers usually tend to practice a density of 5 kg/m^2 (Bogdanov, 1996) and then "split the beddings" where density has doubled, while assuming optimal reproduction densities have been exceeded from that point onwards.

In terms of the food used as a stimulant in reproduction, the following foods are mentioned: fresh horse manure, rabbit or young cattle manure in weighed amounts, sugar-sweetened water (a small cup of sugar for 3 liters of water) that is sprinkled on 2 m^2 every 3-4 days (Duşan, 2013). Sexual maturity in earthworms occurs after 60 to 90 days (Table 3), but reproduction itself begins only after 7-10 months (Duşan, 2013).

Table 3. Earthworm life cycle (Duşan, 2013)

Parent copulation	Cocoon laying	Hatchlings emerging from cocoons	Appearance of the clitellum	Full adulthood/ maturity	End of the cycle

↑ 7-10 days ↑ 14-21 days ↓ 60-90 days ↑ 7-10 months ↑ over 15 years ↑

Earthworm reproductive organ development

RESULTS AND DISCUSSIONS

The vermicompost, the same as the conventional compost, offers many benefits to agricultural soil, including increased water retention capacity, improved nutrient retention, improved soil structure, and enhanced microbial activities (Munroe, 2007).

By using it, the vermicompost helps to improve the physical, chemical and biological properties of the soils. It contributes in increasing the soil's porosity and respectively in increasing the diameter of the pores in the soil from 50 to 500 µm, which results in better aerohydric conditions for crops (Marinari et al., 2000). It also contributes to stabilizing the soil's reaction, increasing the microbial population and the enzyme activities in the soil (Makeswarapa et al., 1999).

Accessibility of nutrients for plants

Atiyeh et al. (2000) discovered that a classical compost is richer in ammonium, whereas the vermicompost tends to be richer in nitrates, which is the most accessible form of nitrogen for plants.

Similarly, Hammermeister et al. (2004) indicated that "vermicompost has higher N availability than the conventional compost on a weight basis and the supply of several other plant nutrients".

Other researchers (Short et al., 1999; Saradha, 1997; Sudha & Kapoor, 2000) found that the supply rate with more nutrients, including phosphorus, potassium, sulfur and magnesium became higher through vermicomposting compared to the conventional composting.

The analyzes carried out in 2012 on the vermicompost produced in Romania showed the following values: pH - 7.8, humidity (%) - 62.74, organic matter (%) - 82.09, humus (%) - 32.30, ash (%) - 17.91, total N (%) - 1.68, total P_2O_5 (%) - 1.44, total K_2O (%) - 1.42 (Panici, 2012).

Table 3. Nutrient content of vermicompost and compost (Nagavallemma K.P. et al., 2004)

Nutrients	Vermicompost (%)	Compost (%)
pH	7.0-7.5	6-7.2
Organic carbon	9.8-13.4	12.2
Nitrogen	0.51-1.61	0.8
Phosphorus	0.19-1.02	0.35
Potassium	0.15-0.73	0.48
Calcium	1.18-7.61	2.27
Magnesium	0.093-0.568	0.57
Sodium	0.058-0.158	<0.01

It appears that vermicomposting tends to lead to higher levels of availability of most nutrients for plants, than the conventional composting process does.

The level of benefic microorganisms

Specialised literature has less information on this topic than on nutrient availability, but it is believed that the vermicompost far exceeds conventional compost with respect to beneficial microbial activity levels. Clive Edwards of Ohio State University (Edwards, 1999) stated that the vermicompost can have 1,000 times more microbial activity than conventional compost, although this figure is not always attained. The vermicompost plays a role in transforming nutrients into forms that are easier to process by plants in comparision to the conventional compost.

The ability to stimulate plant growth

Atiyeh et al. (2000) carried out a thorough examination of the literature on this phenomenon. The authors said: "Most of these studies confirmed that vermicomposts have beneficial effects on plant growth. Vermicomposts, whether used as soil additives or as components of horticultural media, improved seed germination and enhanced rates of seedling growth and development." These beneficial effects on plant growth showed to appear independently of nutritional transformations and availability, increasing plant growth and productivity rates more than it would be possible through simple nutrient conversion of minerals in more available forms for plants. This is the area where most interesting and exciting results have been obtained. Many researchers have found that the vermicompost stimulates the subsequent growth of plants even when plants already receive optimal nutrition (Munroe, 2007).

Ability to suppress diseases

In recent years, a considerable number of tests have been made regarding the vermicompost's ability to protect plants against various diseases. The theory behind this statement is that high levels of beneficial microorganisms in the vermicompost protect plants by competing with pathogens for available resources, while also blocking access to the plant's roots by occupying all available spaces. This analysis is based on the concept of "soil trophic, a soil-based approach (Munroe, 2007). Edwards and Arancon (2004) show that, through the research of vermicompost commercial products for plants attacked by: *Pythium* on cucumbers, *Rhizoctonia* on greenhouse radishes, *Verticillium* on strawberries and *Phomopsis* and *Sphaerotheca fulginae* on grapes, the incidence of diseases decreased significantly. The authors continue to say that pathogen suppression disappeared when the vermicompost was sterilized, indicating that the mechanism involved was the microbial antagonism.

Ability to reject pests

Research in this area is relatively new and the results so far have been inconsistent. A theory is advanced by George Hahn, a vermicompost producer in California, who claims that his product rejects many different insect pests. He believes this is due to the earthworm's production of the chitinase enzyme, which decomposes chitin from insect exoskeleton. However, the independent testing of its product has produced inconsistent results (Biocycle, 2001; Edwards & Arancon, 2004).

CONCLUSIONS

Earthworms play an extremely important role in counteracting the loss of biodiversity. They increase the number and types of microbes in the soil by creating conditions in which these creatures can grow and multiply. The earthworms's intestine has been described as a small "bacteria factory", expeling more microbes than it igests.

By adding vermicompost and earthworm cocoons on the farm's soil, the microbial community of the soil largely multiplies itself. This underground biodiversity is a basis for an increased biodiversity above ground, as soil creatures and the plants that they help to grow are at the heart of the entire food chain.

By growing earthworms, many of the environmental problems can be solved. Their ability to consume all that is biodegradable can help in getting rid of many environmental waste (manure from livestock farms, sewage sludge, leaves and dry grass, sawdust from nonaromatic wood, paperboard and paper).

The vermicompost has a positive effect not only on plants but also on the soil. It greatly improves the soil structure, decreasing its density, increasing the porosity and absorption of nitrogen from the atmosphere. It thus remedies the soils affected by the long-term use of chemicals. Another advantage is given by the suppression of harmful fungi in the soil. These microorganisms produce hormones, vitamins, nutrients, enzymes, amino acids and minerals needed for plants.

Plants treated with vermicompost have a well-developed immune system, making them more resistant to disease and pest attacks.

Moreover, the vermicompost is the only fertilizer accepted in the European Union as an amendment in organic farming.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Atiyeh, R.M., Subler, S., Edwards, C.A., Bachman, G., Metzger, J.D. and Shuster, W. (2000). Effects of vermicomposs and composts on plan growth inhorticultural container media and soil. In *Pedobiologia, 44*. 579–590.

Bogdanov, P. (1996). *Commercial Vermiculture: How to Build a Thriving Business in Redworms*. Ed. Vermico Press, Oregon.

Cracas, P. (2000). *Vermicompostare în stil cubanez.* Compediu despre râme, No. 25.

Dominguez, J. & Edwards, C.A. (1997). Effects of Socking Rate and Moisture Content on the Growth and Maturation of *Eisenia andrei* (Oliogochaeta) in Pig Manure. *Soil Biol. Biochem., 29*(3,4), 743–746.

Duşan, P. (2013). *Practical guide: Earthworm farming*. Ed. Măiastra, Târgu Jiu.

Edwards, C.A. & Lofty, J.R. (1972). *Biology of Earthworms*. London: Chapman and Hall Ltd.

Edwards, C.A. & Arancon, N. (2004). Vermicomposts Suppress Plant Pest and Disease Attacks. In Rednova News: http://www.rednova.com/display/?id=55938.

Edwards, C.A. (1998). *The Use of Earhworms in the Breakdown and Management of Organic Wases* (pp. 327–354). In: Edwards, C.A. Ed. Earthworm Ecology. St. Lucie Press, Boca Raton.

Edwards, C.A. (1999). Interview with Dr. Clive Edwards. In: Casting Call; Peter Bogdanov, Ed. VermiCo, Merlin, Oregon 4(1), June Issue.

Gaddie, R.E. (Senior) & Douglas, D.E. (1975). *Earthworms for Ecology and Profit*. Vol. 1, Scientific Earthworm Farming. Bookworm Publishing Company, California.

Georg (2004). Feasibility of Developing the Organic and Transitional Farm Market for Processing Municipal and Farm Organic Wastes Using Large-Scale Vermicomposting. Good Earth Organic Resources Group, Halifax, Nova Scotia. Available at: http://www.alternativeorganic.com.

Gunadi, B., Charles, B., Edwards, C.A. (2002). The growth and fecundity of *Eisenia fetida* (Savigny) in cattle solids pre-composted for different periods. In *Pedobiologia, 46*. 15–23.

Hammermeister, A.M., Warman, P.R., Jeliazkova, E.A., Martin, R.C. (2004). Nutrient supply and lettucegrowth in response to vermicomposed and composted catle manure. Submitted to Bioresource Technology, December.

Md. Abul, Kashem, Ashoka, Sarker, Imam, Hossain, Md. Shoffikul, Islam (2015). Comparison of the Effect of Vermicompost and Inorganic Fertilizers on Vegetative Growth and Fruit Production of Tomato (*Solanum lycopersicum* L.). Open Journal of Soil Science, 2015, 5. 53–58. Published Online Feburary 2015 in SciRes. http://www.scirp.org/journal/ojss; http://dx.doi.org/10.4236/ojss.2015.52006.

Mila, M.P. (2010). *Research paper on vermiculture and vermicomposting undertaken by bachelor of secondary education, biological science major third year students at Ramon Magsaysay Technological University, San Marcelino, Zambales.* In Partial Fulfillment of the Requirements for Major 6 (Ecology).

Munroe, G. (2007). *Manual of On-Farm Vermicomposting and Vermiculture.* Available at: https://www.researchgate.net/publication/268254767_Manual_of_On-Farm_Vermicomposting_and_Vermiculture.

Nagavallemma, K.P, et al. (2004). Vermicomposting: Recyclinng Wastes into Valuable organic fertilizer. *Global Theme on Agroecosystems*, *8*. 1–2.

Radovich, T., Smith, J., Render, A., & Deenik, J. (2007). *Vermicompost in Hawaii: Its Production and Use.* College of Tropical Agriculture and Human Resources, Virtual FieldDay. http://www.ctahr.hawaii.edu/sustainag/video/vermicompost_r.html.

Rink, R. (Editor), van de Kamp, M., Wilson, G.B., Singley, M.E., Richard, T.L., Kolega, J.J., Gouin, F.R., Laliberty, L.Jr., Kay, D., Murphy, D.W., Hoitink, H.A.J., Brinton, W.F. (1992). *On-Farm Composting Handbook*. Natural Resource, Agriculture and Engineering Service (NRAES-54), Ithaca, NY.

Saradha, T. (1997). The culture of earthworms in the mixture of pond soil and leaf litter and analysis of vermifertilizer. *J. Ecobiology, 9*(3), 185–188.

Short, J.C.P., Frederickson, J., Morris, R.M. (1999). Evaluation of traditional windrow-composting and vermicomposting for the stabilization of wase paper sludge (WPS). In Diaz Cosin, D.J. Jesus, J.B. & Trogo D. (Eds), 6th International Symposium on Earthworm Ecology, Vigo, Spain, 1998. *Pedobiologia, 43*(6), 735–743.

Sudha, B. & Kapoor, K.K. (2000). Vemicomposting of crop residues and cattle dung with *Eisenia fetida*. In *Bioresource Technology*, 73, 95–98.

***Biocycle, 2001. *Vermicompost as Insect Repellent.* Jan 01, 19.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE CELLULOLYTIC ACTIVITY DEPENDING ON SOIL TILLAGE AND INFLUENCE OF FOREST STRIP UNDER WINTER WHEAT AGROCOENOSES

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Abstract

The aim of the study was to determine cellulolytic activity depending on soil tillage in a long-term crop rotation under winter wheat agrocoenoses. There also were determined the cellulolytic activity in dependence of experimental plots location toward forest strip. The study was carried out at the Didactic Experimental Station "Chetrosu" of the State Agrarian University of Moldova on carbonate chernozem with sandy loam texture. The cellulolytic activity was determined according to the Misustin E., 1978, method based on the principle of cellulose decomposition under aerobic conditions by incorporating linen tissue in the 0-30 cm soil layer. The research has shown that soil cellulolytic activity is characterized with very low index level under both Plowing and No-till variants during June of 2017-2018 crop year - that registered dry weather conditions. The presence of forest strip near the crop land has revealed its beneficial influence on soil moisture preserve that facilitated cellulolytic activity on the plots located besides the forest strip under both soil tillage variants. It has been confirmed that forest strip (shelterbelt) form a microclimate with benefits for crop land.

Key words: carbonate chernozem, cellulolytic activity, forest strip, soil moisture, soil tillage, weather conditions.

INTRODUCTION

Soils that are involved in agricultural production are influenced by powerful anthropogenic factors modifying the soil biological activity, physicochemical properties, and ultimately fertility. The study of the nature of the impact on soil microflora facilitates the selection process techniques, helping to improve soil fertility and its properties, makes it possible to predict changes in the level of cultivation. The most sensitive indicators, responsive to changing water and air, heat, soil nutrient regimes are soil microorganisms. However, despite the obvious need to study the soil microbocenosis, this issue is given little attention. Determination of the intensity of soil-biological processes under the influence of anthropogenic change will allow establishing the level of biological activity, in order to select human impacts that have a positive impact on soil fertility and crop yields (Churkina Galina et al., 2012).

Cellulose is the most abundant renewable natural product in the biosphere (Bakare et al., 2005). The degradation of cellulosic biomass represents an important part of the carbon cycle (Spence et al., 2010) at both local and global scales (Lynd et al., 2002). A large number of cellulolytic microorganisms are involved in the decomposition of plant material in soil (Spence et al., 2010) so bio-sequestered carbon return to soil environment (Lynd et al., 2002).

Cellulolytic microorganisms are fundamental for the transformation of cellulose into sugars that are essential nutrients for various organisms (Bhat & Bhat, 1997).

More than 90% of the global amount of cellulose is degraded in well aerated agricultural, grassland or forest soils (Bastian et al., 2009; Kurka, 2001).

Soil microbial community is very dynamic and promptly affected by different soil uses regarding management, frequency and amounts of applied fertilizers, or any other disturbance (Cardoso et al., 2013). The preservation of soil microbial diversity is crucial for a balanced agro-ecosystem, especially under increasing agricultural intensification. In the past, agricultural practices have failed to promote populations of microorganisms, limit production yields and threaten sustainability. Agricultural practices such as organic farming, reduced soil tillage, crop rotation, intercropping, and land use extensification may help soil microorganisms to become more abundant and active (García-Orenes et al., 2013).

Microbial abundance and activity are promoted by reduced tillage and fertilization, while incorporation of crop residues demonstrated a higher effect in comparison with other cropping technologies. Increase in microbial number and activity most likely occurred due to increases in organic matter and changes in organic matter quality. Development of sustainable agricultural strategies, based not only on crop productivity but also on ecological principles, is crucial to maintaining soil microbial communities as well as soil quality and fertility (Marinkovic, Jelena et al., 2018).

The objective of this study was to assess the cellulolytic activity in carbon chernozem under different soil tillage. Also the presence of forest strip near the experimental plots allowed to find out its influence on the agroecosystem soil moisture and respectively cellulolytic activity.

MATERIALS AND METHODS

The present study was carried out on carbonate chernozem with sandy loam texture at the educational and experimental station of the State Agrarian University of Moldova DES Chetrosu in a long-term crop rotation (Photo 1).

Fhoto 1. Carbonate chernozem DES Chetrosu

The cellulolytic activity has been determined depending on: soil tillage - conventional (plowing) and conservative (No-till - 5 years old) and the spatial location of experimental plots against old forest strip - the plot no. 3 is located near forest strip and the plot no. 7 in the middle of the experimental field.

The cellulolytic activity was determined during June of 2017-2018 crop year according to the Misustin E., 1978, method based on the principle of cellulose decomposition under aerobic conditions by incorporating linen tissue in the 0-30 cm soil layer for 30 days, under winter wheat agrocoenoses (Photo 2).

Photo 2. Experimental field with winter wheat

The results of cellulolytic activity was evaluated according to Table 1.

Table 1. Values of cellulolytic activity on chernozems (Misustin E., 1978)

Index level	Cellulolytic activity
Very low	< 36
Low	36-52
Middle	52-68
Great	68-84
Very great	> 84

There were also determined soil moisture (May, June) and resistance to penetration (May).

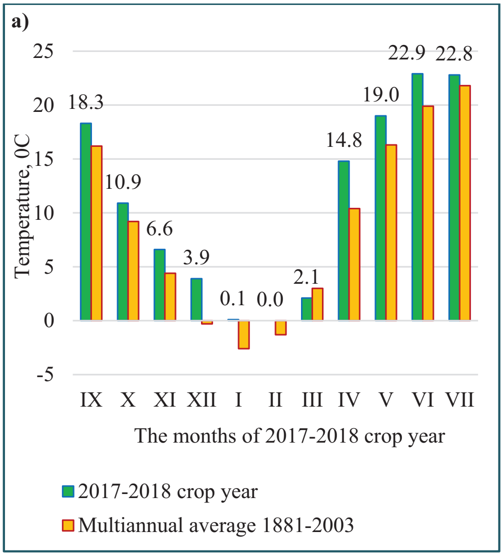
Chemical and physical soil properties were determined according to methods recommended and used in the pedological monitoring of Republic of Moldova (V. Cerbari, 1997; 2010).

The soil of researched agroecosystem is carbonate chernozem characterized by the sub-moderate humus content (2.4% in 0-30 cm soil layer), the sum of Ca^{++} and Mg^{++} in arable layer is about 22.0 me/100 g soil. The carbonates are present throughout the profile

Ap-Ahk-Bhk-BCk-Ck, ranging from 1% in the upper layer to 8% at a depth of 110-120 cm. Soil reaction is slightly alkaline.

The weather conditions registered for 2017-2018 crop year (Figure 1) show that the months in which occurred research (May, June) was characterized with average temperatures over 3°C to multiannual average (1881-2003).

The amount of rainfall registered deficit to multiannual average: during May the precipitation deficit has been 28.7% and for June - 58.4%.



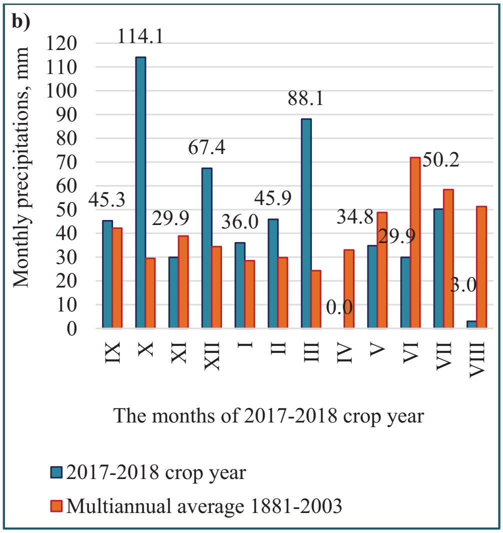


Figure 1. Meteorological conditions of 2017-2018 crop year, DES Chetrosu: a) temperature; b) precipitation

RESULTS AND DISCUSSIONS

The different cropping systems, inclusive soil tillage, can have a positive or negative effect on microbiological number and activity, which directly reflect the fertility of the soil (Falkowski et al., 2008).

Soils exposed to disturbance by tillage can be more susceptible to reductions in soil microorganisms due to desiccation, mechanical destruction, soil compaction, reduced pore volume, and disruption of access to nutrient resources (Huang et al., 2013). Ploughing and tillage operations facilitate aeration in soil and exposure of soil to light and thereby increase the biological activity of organisms, particularly of bacteria (Roger-Estrade et al., 2010).

Non-agricultural soils as no-tillage systems under native vegetation have a stimulatory effect on the microbial parameters (Marinkovic, Jelena et al., 2018).

With the purpose to know physical soil conditions on which largely depends cellulolytic activity there have been studied – soil moisture and resistance to penetration under Plowing and No-till variants (Table 2).

The research occurred in mid-May under winter wheat agrocoenoses has shown the same tendency regarding soil moisture and settlement of 0-50 cm layer for both investigated variants (Plowing and No-till). So the 0-20 cm soil layer has distinguished with 18% moisture and with close values of resistance to penetration - 13.2 kgf/cm^2 for Plowing and 16.4 kgf/cm^2 for No-till variant.

The underlying 20-50 cm soil layer has been very different from previous one: moisture has decreased to 13% and resistance to penetration has grown much to 30 kgf/cm^2 values under both variants.

The observed soil moisture difference could be caused by small amount of precipitation that has penetrated only surface soil layer, while more favourable soil resistance to penetration registered in 0-20 cm layer, beside good moisture conditions is due to roots abundance of winter wheat - concentrated mainly in the upper soil layers.

The research of cellulolytic activity (June) parallel spent with soil moisture determination (Tables 3, 4) shows the close dates under both soil tillage variants (Plowing and No-till). The index level of cellulolytic activity during June is very low - the tissue breaking (% to the initial mass) has registered the limits of 28.0-34.0% (Figure 2). This has been coused by dry weather conditions of June caracterized with unsuficient precipitations (deficit of 58.4% to multiannual average) and high temperatures that registered over 3°C to multiannual average (Figure 1). So the main limiting factor that has inhibited cellulolytic activity was soil moisture that registered limits of 12.3-15% in arable layer (0-30 cm) (Photo 3).

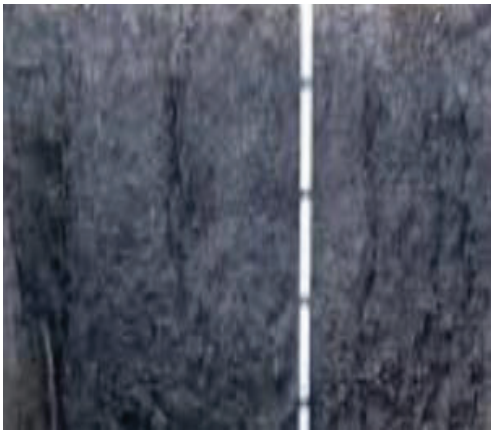


Photo 3. The arable layer (0-30 cm) of carbonate chernozem

The presence of forest strip near the crop land has revealed its beneficial influence on soil moisture preserve (Table 3) - on the plot no. 3, situated near the forest strip, has been registered higher soil moisture content (15%) in arable layer comparatively with the soil moisture (12.3%) of experimental plot no. 7 - situated in the middle of the field. The differences of 3% soil moisture recorded between these plots has facilitated cellulolytic activity near forest strip (Table 4) under both soil tillage variants.

It has been confirmed that around forest strip (shelterbelt) is formed a microclimate with many benefits for crop land: give a protection from wind, reduce evapotranspiration from crop lands, reducing transpiration from plants, controls blowing snow, habitat for wildlife, shelter for honeybees, energy saving, aesthetic value (Molla Mekonnen Alemu, 2016).

One of the main tasks of agricultural and microbiological science is to solve the problem of soil fertility and plant productivity. Research has so far proved that microorganisms present one of the main factors of soil fertility and growth plants. Soil microflora and, in particular, that part of it that develops around the roots, acts directly on plant growth and development. These microorganisms perform differently functions depending on their own particularities and host plants. Some are able to optimize soil fertility dissolving its mineral and releasing some of the nutrients into forms accessible to plants (Кожемяков, Тихонович, 1998; Onofraş et al., 2003).

Table 2. Physical soil conditions under winter wheat, May 2018

Soil tillage	Depth, cm	Moisture, %	Penetration resistance, kgf/cm^2
Plowing	0-10	19.07	9.0
	10-20	17.14	17.3
	20-30	12.52	29.9
	30-40	13.45	30.0
	40-50	13.64	30.0
No-till	0-10	20.11	11.9
	10-20	15.46	20.8
	20-30	11.67	28.9
	30-40	12.87	29.6
	40-50	13.16	30.0

Table 3. Soil moisture under winter wheat agrocoenoses depending on soil tillage and experimental plot, June 2018

Depth, cm	Plowing				No-till			
	Plot no. 3 (near forest strip)		Plot no. 7 (in the middle of the experimental field)		Plot no. 3 (near forest strip)		Plot no. 7 (in the middle of the experimental field)	
0-10	17.51	14.9	12.21	12.3	15.52	15.0	11.37	12.4
10-20	15.46		12.38		15.99		12.93	
20-30	11.67		12.36		13.36		12.78	
30-40	12.87	12.8	12.90	13.0	12.98	13.4	12.33	13.6
40-50	13.16		13.11		12.31		12.86	
50-60	13.41		13.34		12.07		12.98	
60-70	12.10		12.63		13.43		13.89	
70-80	12.97		12.97		13.48		14.18	
80-90	12.64		13.67		14.16		14.36	
90-100	12.37		12.94		14.25		14.78	
100-110	12.89		12.17		14.28		13.68	

Table 4. Cellulolytic activity (%) depending on soil tillage and experimental plot, June 2018

Soil tillage	Plot nomber	Depth, cm	Tissue breaking, % to the initial mass		Index level
Plowing	3	0-10	32.1	31.9	Very low
		10-20	32.8		
		20-30	30.9		
	7	0-10	28.0	29.3	
		10-20	29.5		
		20-30	30.3		
No-till	3	0-10	33.0	32.9	
		10-20	34.0		
		20-30	31.8		
	7	0-10	28.6	29.7	
		10-20	29.8		
		20-30	30.8		

Figure 2. Tissue breaking, June 2018

CONCLUSIONS

Determination of physical soil conditions - soil moisture and resistance to penetration (mid-May) under winter wheat agrocoenoses has shown the same tendency for both soil tillage variants (Plowing and No-till). The resistance to penetration is high (about 30 kgf/cm^2) in the 20-50 cm soil layer and favourable in 0-20 cm layer, that beside good moisture conditions, may be caused by roots abundance of winter wheat - concentrated mainly in the upper soil layers.

The weather conditions registered for May, June of 2017-2018 crop year are characterized with average temperatures over 3°C to multiannual average (1881-2003) and with precipitation deficit of 28.7% for May and of 58.4% for June.

The research of cellulolytic activity, parallel spent with soil moisture determination, showed the close dates under both soil tillage variants (Plowing and No-till).

The index level of cellulolytic activity during June is very low - the tissue breaking (% to the initial mass) has registered the limits of 28.0-

34.0%. The cellulolytic activity was inhibited by drought.

The presence of forest strip near the crop land has revealed its beneficial influence on soil moisture preserve. The plot situated near the forest strip, has been registered higher soil moisture content (15%) in arable layer comparatively with the soil moisture (12.3%) of experimental plot situated in the middle of the field. The differences of 3% soil moisture content recorded between these plots has facilitated cellulolytic activity near the forest strip under both soil tillage variants. It has been confirmed that around forest strip (shelterbelt) is formed a microclimate with benefits for crop land.

REFERENCES

Bakare, M., Adewale, I., Ajayi, A., Shonukan, O. (2005). Purification and characterization of cellulase from the wild-type and two improved mutants of *Pseudomonas fluorescens*. *Afr. J. Biotechnol., 4*. 898–904.

Bastian, F., Bouziri, L., Nicolardot, B., Ranjard, L. (2009). Impact of wheat straw decomposition on successional patterns of soil microbial community structure. *Soil Biology and Biochemistry, 41*. 262–275.

Bhat, M.K., Bhat, S. (1997). Cellulose degrading enzymes and their potential industrial applications. *Biotechnol Adv., 15*. 583–620.

Cardoso, E.J.B.N., Vasconcellos, R.L.F., Bini, D., Miyauuchi, M.Y.H., dos Santos, C.A., Alves, P.R.L., de Paula, A.M., Nakatani, A.S., de Moraes Pereira, J. & Nogueira, M.A. (2013). Soil health: looking for suitable indicators. What should be considered to assess the effects of use and management on soil health? *Scientia Agricola, 70*. 274–289.

Cerbari, V. (1997). *Metodica instituirii monitoringului funciar in Republica Moldova*. Chisinau.

Cerbari, V. (2010). *Monitoringul calitatii solurilor Republicii Moldova.* Chisinau: Pontos.

Churkina, G., Kunanbayev, K., Akhmetova, G. (2012). The taxonomic composition of soil microorganisms in the ecosystems of southern chernozems of Northern Kazakhstan. *Applied Technologies & Innovations, 8*(3), 13–19.

Falkowski, P.G., Fenchel, T., Delong, E.F. (2008). The microbial engines that drive Earth's biogeochemical cycles. *Science, 320*. 1034–1039.

García-Orenes, F., Morugán-Coronado, A., Zornoza, R., Scow, K. (2013). Changes in soil microbial community structure influenced by agricultural management practices in a mediterranean agro-ecosystem. *PLOS ONE, 11*(3), E0152958

Huang, M., Jiang, L., Zou, Y., Xu, S., Deng, G. (2013). Changes in soil microbial properties with no-tillage in Chinese cropping systems. *Biology and Fertility of Soils, 49*. 373–377.

Kurka, A. (2001). The use of cellulose strips to study organic matter decomposition in boreal forested soils. *Boreal Environment Research, 6*. 9–17.

Lynd, L.R., Weimer, P.J., van Zyl, W.H., Pretorius, I.S. (2002). Microbial cellulose utilization: fundamentals and biotechnology. *Microbiol.Mol.Biol. Rev., 66*. 506–577.

Marinkovic, J., Bjelic, D., Šeremešic, S., Tintor, B., Ninkov, J., Živanov, M., Vasin, J. (2018). Microbial abundance and activity in chernozem under different cropping systems. *Ratar. Povrt., 55*(1), 6–11.

Onofraş, L., Todiraş, V., Prisacari, S. (2003). Eficacitatea şi perspectiva utilizării microorganismelor de rizosferă. *Bul. al AŞ a Moldovei. Şt. Biol., Chim. şi Agr., 2*(291), 108–111.

Roger-Estrade, J., Anger, C., Bertrand, M., Richard, G. (2010). Tillage and soil ecology: Partners for sustainable agriculture. *Soil and Tillage Research, 111*. 33–40.

Spence, K.L., Venditti, R.A., Habibi, Y., Rojas, O.J., Pawlak, J.J. (2010). The effect of chemical composition on microfibrillar cellulose films from wood pulps: mechanical processing and physical properties. *Bioresour Technol, 101*. 5961–5968.

Мишустин Е., Емцев В. (1978). *Микробиология*. Москва: Из. Колос.

Кожемяков, А.П., Тихонович, И.А. (1998) Использование инокулянтов бобовых и биопрепаратов комплексного действия в сельском хозяйстве. Докл. Россельхозакадемии. Москва, 6. 7–18.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

BIODEGRADATION OF WOOL WASTE: THEORY AND PRACTICAL ASPECTS

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Abstact

The elimination of waste from every industry is one of the critical problems facing manufacturers. Different technologies for different types of waste can be used in a way that is as convenient as possible for the manufacturer. Textile industry is one of the fields of activity that produces a series of biodegradable waste. Among these, the waste generated by the industrialization of wool is a product whose disposal in the environment requires sustainable treatment processes. Therefore, scientific literature has proposed the transformation of waste wool by composting and its direct use as an amendment for agricultural and gardening soils.

Key words: *waste wool, biodegradation, recycling, agriculture, composting.*

INTRODUCTION

The textile industry affects the environment by: high consumption of water used for wool washing, energy consumption and pollutants. Also for the solubilization of wool cloth substances that provide protection for animals against precipitation, as well as the large amount of waste generated by the felt processing technology.

Wastes from the wool processing industry, respectively from the felt factory, are a secondary problem, but at the same time important, which is generally solved by the accumulation of large quantities of waste deposited in the special waste disposal site, being transported in the final landfill.

In addition, Directive 75/442/EEC establishes a waste hierarchy. The most desirable is the prevention of waste and the minimization of waste generation, by these measures being understood also the transformation into compost (raw material as an organic fertilizer for agriculture). Waste is thus transformed into raw material for composting that no longer generates waste, being a beneficial product for agricultural production.

European Directive (1999/31/EC) on the landfill of waste for the collection and processing of biodegradable waste as well as the European Directive (2008/98/EC) laying down measures for the protection of the environment and the health of the population. Composting is a means of achieving minimal expense and maximum benefit if used as a concentrated organic fertilizer applied to crops with long-lasting vegetation.

MATERIALS AND METHODS

The compost burst was built on the field of University of Agronomic Sciences and Veterinary Medicine of Bucharest. In the summer of 2017 wool wastes from the felt factory, wheat straw and cattle dung from the Prince Mill. Waste is considered as a residual polluting product by the felt industry but can be used as raw material and not as waste in the form of compost, as an organic fertilizer and as an amendment to soil fertility, generating benefits at minimal cost.

In order to build the composting pile under ideal conditions and to obtain sufficiently high temperatures, for the sterilization of weed seeds and pathogenic microbial agents, other organic waste was used to stimulate the composting process (Wiese et al., 1998; Hustvedt G. et al., 2016).

Due to the high organic nitrogen content of wool fibers (waste), we used the supra unit fraction to obtain the proper proportions of composting materials composting:

$$R = \frac{M_1[C_1(100-U_1)] + M_2[C_2(100-U_2)] + M_3[C_3(100-U_3)]}{M_1[N_1(100-U_1)] + M_2[N_2(100-U_2)] + M_3[N_3(100-U_3)]}$$

where:
R - C/N ratio;
M_1, M_2, M_3 - mass of materials subject to composting (g, kg);
C_1, C_2, C_3 - carbon of materials (%);
N_1, N_2, N_3 - nitrogen of materials (%);
U_1, U_2, U_3 - moisture of materials (%).
Following mathematical calculations, we obtained a C/N ratio of 20-25 (Table 1).

Table 1. The C/N ratio in 2017 to the construction of the compost pile

The material	H_2O %	Mass (g, kg)	C %	N %	Report C/N (20-25)
Straw	10.19	10	48	0.5	22.448
Wool waste	21.47	1	50	17	
Cattle buckwheat	45	3	25	3	

The composting pile contains 10 kg of straw, 1 kg of wool waste and 3 kg of bovine manure in successive layers. The mixture was made in the stamps, so as to obtain uniformity of the materials subjected to composting (Figure 1).

Figure 1. Laying in successive layers of materials subject to composting

The compost burst was built according to P. Pfeiffer's recommendations in 1966 so it is presented in (Figure 2).
The layering of the materials subject to composting has been done manually for a better distribution of materials.
The waste of wool was evenly distributed along with straw and bovine manure.
The height of the compost pile ranged from 1.5 to 2 m depending on the degree of decomposition of the materials.

Figure 2. Compost piles after Pfeiffer P., 1966

This height allowed the heap to generate enough heat to inactivate pathogens (including weed seeds). Removal of the composting pile was done monthly, manually, from top to bottom to ensure that the exposed exposed surfaces were buried inside the pile each time the pile was reshaped. This allowed all weed seeds and pathogens to be exposed to high temperatures inside the heap. Humidity was provided by spraying.

RESULTS AND DISCUSSIONS

In the first month of the pile construction, the temperature was measured in the middle of the pile once every 2-3 days, approximately the same hour, in three rehearsals.
For this, temperature evolution was tested by 18 determinations. Using the Excel application, linear regression (Figure 3) and square regressions (Figure 4) and cubic (Figure 5) were determined for which the correlation ratios (Table 2) were determined to determine the function that best approximates the composting process.

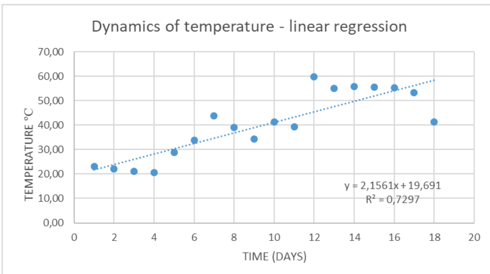

Figure 3. Regression of the temperature relative to the incubation period in the composting piles in the Experimental Field of the Faculty of Agriculture - Bucharest in 2017

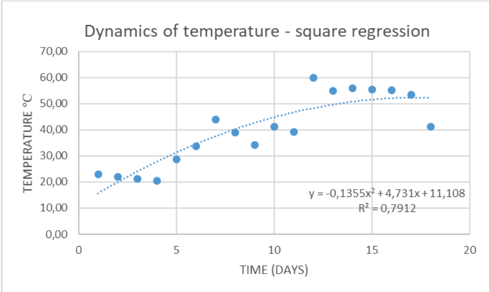

Figure 4. The quaternary regression of the temperature from the incubation period in the composting piles in the Experimental Field of the Faculty of Agriculture - Bucharest in 2017

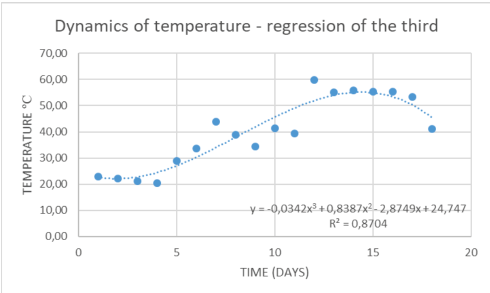

Figure 5. The third degree regression of the temperature from the incubation period in the composting piles in the Experimental Field of the Faculty of Agriculture - Bucharest in 2017

Table 2 shows that the highest correlation correlation coefficients of 0.932 were achieved at cubic regression, although at 18 pairs of values both linear regression with the correlation ratio of 0.854 and the square with 0.888 of the test showed them very significant.

Table 2. Regressions and values of correlation ratios that related to the temperature and incubation period in the incubation process

The type of equation	The value of R under the radical	Semnificaţion
$y = 2{,}156x + 19{,}691$	0.854	***
$y = -0{,}135x^2 + 4{,}731x + 11{,}108$	0.889	***
$y = 0{,}034x^3 + 0{,}838x^2 - 2{,}874x + 24{,}747$	0.932	***

The mixture was monitored and remixed once a month as the heap temperature began to decrease, averaging 41.23°C.
This low average temperature is due both to the depletion of the sources of nutrition of heterotrophic microorganisms and to the reduction of compostable organic matter, as well as to the decrease of atmospheric temperatures.
One month after the founding of the pile, we found molds on straw but also on wool waste (Figure 6).

Figure 6. Appearance of molds on wool waste under composting, 2017

As can be seen in Figure 6, we find that actinomycetes and fungi micelles are colonized on the material subjected to composting (prehumic materials) with heterotrophic bacteria.
Some of these are thermogenic (heat), which contributes to the increase of the temperature from the compost stack to the average of 59.83°C, as self-biosterilization agent, when the humification and mineralization processes begin to satisfy both the release of mineral molecules and molecules of related pre-human substances.
The color of materials subject to composting becomes more and more dark. The wool (waste) wool due to the keratinous outer cover is hardly biodegradable and therefore we have introduced as a stimulator for the biodegradation of the keratin layer, the bovine waste.
The introduction of the bovine manure into the composting mixture was done to stimulate the growth of the number of heterotrophic microorganisms necessary to stimulate the keratinolytic factors, which would degrade the keratin embodying the wool yarn.
The aspect of biodegradation of wool waste was followed by a microscope, obtaining the photographic image (Figures 3, 4, 5) of the appearance of keratin biodegradation.

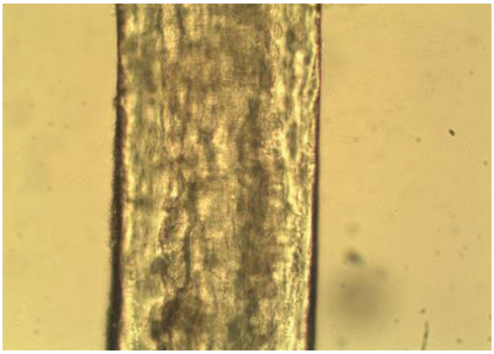

Figure 3. Appearance of uncomposed wool waste

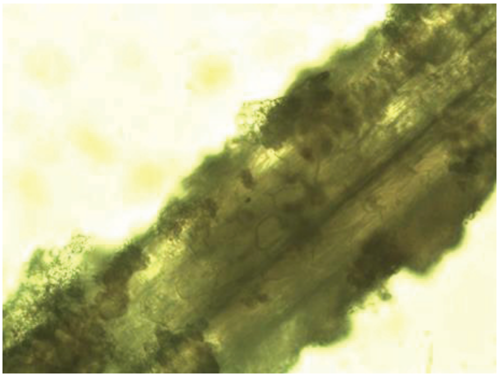

Figure 4. Appearance of compost wool waste in the first month of reshuffle

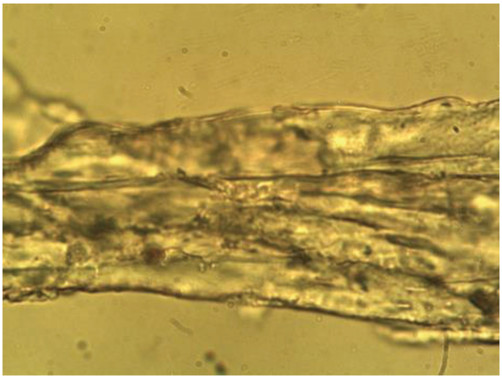

Figure 5. Appearance of wool debris with degraded keratin layer

After maturation, compost produced from wool waste, cattle manure and wheat straw was sifted manually.
The compost samples were then sent for analysis to the Chemistry Laboratory of the Academy of Agricultural and Forestry Sciences “Gheorghe Ionescu-Şişeşti” (Bucharest).

Since the compost pile is diminishing over time in size, the collected samples were inside the pile, for the analyzes to be representative of all materials subject to composting. According to the analysis report, the chemical test method of the mixture of compost materials is shown in (Table 3), as follows:

- Organic carbon (Corganic), wet oxidation;
- Organic matter (MO) through loss at calcination; Methodology I.C.P.A. (1981), Vol. 1, Cap. 23, PT 44;
- Azot Kjeldahl (Nt), SR EN ISO 20483: 2007, PTL 11;
- Nitric nitrogen (N-NO3) determined potentiometrically, ICPA Methodology (1980), ch. 4 PT 98.

Table 3. Analytical results

No	Identification	$C_{organic}$ %	MO %	N_t %	N-NO_3 mg/kg
1	Repetition 1	17.3	34	1.302	178
2	Repetition 2	18.0	37	1.318	177
3	Repetition 3	19.4	36	1.323	178
Average		18.2	35.6	1.314	177.6

The results of these tests indicate that, on average, compost produced from wool waste is suitable for use in agriculture.

CONCLUSIONS

The experiment presented in this chapter was an attempt to understand the use of wool waste for agricultural purposes as an extremely valuable organic fertilizer as raw material for obtaining superior, unpolluted and non-waste agricultural produce.

ACKNOWLEDGEMENTS

Thank you to the laboratory of the Gheorghe Ionescu-Siseşti Academy of Agricultural Sciences and Forestry (Bucharest) for the chemical analysis of the compost.

REFERENCES

Cooperband, L. (2002). The Art and Science of Composting. A resource for farmers and compost producers. Center for Integrated Agricultural Systems, University Wisconsin - Madison.

Das, K., Tollner, E.W., Annis, P.A. (1997). In: Zheljazkov V. D., Stratton G. W., Pincock J., Butler S., Jeliazkova E.A., 2009. Wool-waste as organic nutrient source for container-grown plants. *Waste Management, 29*, 2160–2164.

Directive 2009/28/EC on the promotion of the use of energy from renewable sources and amending and subsequently repealing Directives 2001/77/EC and 2003/30/EC.2009.04.23. *Off J. Eur Union*, *L*(140), 16-62.

Evangelou, M.W.H., Ebel, M., Koerner, A., Schaeffer, A. (2008). In: Zheljazkov V.D., Stratton G.W., Pincock J., Butler S., Jeliazkova E.A., 2009. Wool-waste as organic nutrient source for container-grown plants. *Waste Management, 29*, 2160–2164.

Górecki, R.S., Górecki, M.T. (2010). Utilization of Waste Wool as Substrate Amendment in Pot Cultivation of Tomato, Sweet Pepper, and Eggplant. *Polish J. of Environ. Stud., 19*(5), 1083–1087.

Govi, M., Ciavatta, C., Sitti, L., Gessa, C. (1998). Influence of organic fertilisers on soil organic matter: a laboratory study. 16th World Congress of Soil Science. http://natres.psu.ac.th/Link/SoilCongress/bdd/symp 40/974-r.pdf (5. avg. 2010).

Hustvedt, G., Meier, E., Waliczek, T. (2016). The Feasibility of Large-Scale Composting of Waste Wool. S.S. Muthu and M.A. Gardetti (eds.), Green Fashion, Environmental Footprints and Eco-design of Products and Processes, DOI 10.1007/978-981-10-0111-6_4.

Ionescu-Sisești, V., Papacostea, P., Ștefanic, G. (1980). *Compostul - îngrașământ din deșeuri organice*. Editura Științifică și Enciclopedică, București.

Ionescu, A., Jinga, I., Ștefanic, Gh. (1985). *Utilizarea deșeurilor organice ca îngrășamânt.* Editura Ceres, București.

Juarez, M.F., Prähauser, B., Walter, A., Insam, H., Franke-Whittle, I.H. (2015). Cocomposting of biowaste and wood ash, influence on a microbially driven-process. In: Onwosi C.O., Igbokwe V.C., Odimba J.N., Eke I.E., Nwankwoala M. O., Iroh I. N., Ezeogu L.I., 2017. Composting technology in waste stabilization: On the methods, challenges and future prospects. Review. *Journal of Environmental Management*, *190*, 140–157.

Kraft von Heinitz (2012). Compostul în gospodărie. Editura Casa, Oradea.

Li, Z., Lu, H., Ren, L., He, L. (2013). Experimental and modeling approaches for food waste composting: a review. In: Onwosi C.O., Igbokwe V.C., Odimba J. N., Eke I.E., Nwankwoala M.O., Iroh I.N., Ezeogu L.I., 2017. Composting technology in waste stabilization: On the methods, challenges and future prospects. Review. *Journal of Environmental Management*, *190*, 140–157.

Nustorova, M., Braikova, D., Gousterova, A., Vsileva-Tonkova, E., Nedkov, P. (2006). Chemical, microbiological and plant analysis of soil fertilized with alkaline hydrolysate of sheep's wool waste. *World Journal of Microbiology and Biotechnology 22*(4), 383–390.

Onwosi, C.O., Igbokwe, V.C., Odimba, J.N., Eke, I.E., Nwankwoala, M.O., Iroh, I.N., Ezeogu, L.I. (2017). Composting technology in waste stabilization: On the methods, challenges and future prospects. Review. *Journal of Environmental Management, 190*, 140–157.

Plat, J.Y., Sayag, D., Andre, L. (1984). In: Zheljazkov V. D., Stratton G. W., Pincock J., Butler S., Jeliazkova E.A., 2009. Wool-waste as organic nutrient source for container-grown plants. *Waste Management, 29*, 2160–2164.

Poston, T., 2006. In: Hustvedt G., Meier E., Waliczek T. (2016). The Feasibility of Large-Scale Composting of Waste Wool. S.S. Muthu and M.A. Gardetti (eds.), Green Fashion, Environmental Footprints and Eco-design of Products and Processes, DOI 10.1007/978-981-10-0111-6_4.

Poston, T., 2006: In Hustvedt G., Meier E., Waliczek T. (2016). The Feasibility of Large-Scale Composting of Waste Wool. S.S. Muthu and M.A. Gardetti (eds.), Green Fashion, Environmental Footprints and Eco-design of Products and Processes, DOI 10.1007/978-981-10-0111-6_4.

Proietti, P., Calisti, R., Gigliotti, G., Nasini, L., Regni, L., Marchini, A. (2016). Composting optimization: integrating cost analysis with the physical-chemical properties of materials to be composted. In: Onwosi C.O., Igbokwe V.C., Odimba J.N., Eke I.E., Nwankwoala M.O., Iroh I.N., Ezeogu L.I., 2017. Composting technology in waste stabilization: On the methods, challenges and future prospects. Review. *Journal of Environmental Management*, *190*, 140–157.

Răuță, C. (1980). Metodologia I.C.P.A. vol. 1, cap. 23, PT 44.

Răuță, C. (1981). Metodologia I.C.P.A. cap. 4, PT 98.

Storino, F., Arizmendiarrieta, J.S., Irigoyen, I., Muro, J., Aparicio-Tejo, P.M. (2016). Meat waste as feedstock for home composting: effects on the process and quality of compost. In: Onwosi C.O., Igbokwe V.C., Odimba J.N., Eke I.E., Nwankwoala M.O., Iroh I.N., Ezeogu L.I., 2017. Composting technology in waste stabilization: On the methods, challenges and future prospects. Review. *Journal of Environmental Management*, *190*, 140–157.

Verville, R.R., 1996. In: Zheljazkov V.D., Stratton G.W., Pincock J., Butler S., Jeliazkova E.A. (2009). Wool-waste as organic nutrient source for container-grown plants. *Waste Management, 29*, 2160–2164.

Vončina, A., and Mihelič, R. (2013). Sheep wool and leather waste as fertilizers in organic production of asparagus (*Asparagus officinalis* L.). *Acta Agriculturae Slovenica, 101*(2), 191–200.

Wang, Y. (2010). Fiber and Textile Waste Utilization. Waste Biomass Valor 1:135–143. DOI 10.1007/s12649-009-9005-y.

Wang, Y., Ai, P., Cao, H., Liu, Z. (2015). Prediction of moisture variation during composting process: a comparison of mathematical models. In: Onwosi C. O., Igbokwe V. C., Odimba J. N., Eke I. E., Nwankwoala M. O., Iroh I. N., Ezeogu L. I., 2017. Composting technology in waste stabilization: On the methods, challenges and future prospects. Review.

Journal of Environmental Management, *190*, 140–157.
Zheljazkov, V.D., Silva, J.L, Patel, M, Stojanovic, J., Youkai, L., Kim, T., Horgan, T. (2008). Human hair as a source of horticultural crops. *HorTech., 18*(4), 592.
Zheljazkov, V.D., Stratton, G.W., Pincock, J., Butler, S., Jeliazkova, E.A. (2009). Wool-waste as organic nutrient source for container-grown plants. *Waste Management*, *29*, 2160–2164.
Zheljazkov, V.D., et al. (2009): In Hustvedt G., Meier E., Waliczek T., 2016. The Feasibility of Large-Scale Composting of Waste Wool. S.S. Muthu and M.A. Gardetti (eds.), Green Fashion, Environmental Footprints and Eco-design of Products and Processes, DOI 10.1007/978-981-10-0111-6_4.
Zoccola, M. (2014): In Hustvedt G., Meier E., Waliczek T., 2016. The Feasibility of Large-Scale Composting of Waste Wool. S.S. Muthu and M.A. Gardetti (eds.), Green Fashion, Environmental Footprints and Eco-design of Products and Processes, DOI 10.1007/978-981-10-0111-6_4.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF PLASTIC MULCH SYSTEM ON AGRO-CLIMATIC FACTORS AND STRAWBERRIES DISEASES IN ORGANIC SYSTEM

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Abstract

The desire for sustainable agriculture has begun to spread in the las decades. The main issues of growing strawberries in organic system are represented by the control of diseases and pests and weeds control. Their appearance and development is directly influenced by agro-climatic factors, being, in this case, necesarry to use the most effective techniques and methods to control them. Thus, for the control of weeds the soil was mulched with black plastic. The soil temperature and humidity was monitorized in fieldd and tunnel using two HOBO microstations. The sensors were placed at 10-15 cm depth. In field were registered the rainfalls and air temperature at a heigh of 1m. Durring the study (March-June 2018), the highest values of soil temperature and moisture were recorded in tunnel. From phytosanitary point of view, the fluctuations of agro-climatic factors influenced both the variety and the amount of strawberries affected by fungal diseases. The highest quantity of strawberries was affected in field.

***Key words**: agro-climatic factors, monitoring, organic system, soil, strawberry disease.*

INTRODUCTION

Due to the high demands of consumers for organic products, especially in North America and Europe, gouvernments of many countries make efforts to encourage farmers to make the transition from conventional farming to organic farming system (Willer et al., 2009).

Sustainable agriculture is based on the intensification of ecological processes, thus prohibiting the use of all synthetic inputs (FAO/WHO Codex Alimentarius Commission 1999).

Globally, agricultural area under organic management has increased significantly, with an area of 50.9 milion ha in 2015, compared to 11 milion ha in 1999 (Willer & Lernoud, 2016; 2017).

Increasing interest for this type of agriculture has led to a deepening of research, being developed effective practices for the development of sustainable agriculture (Graf & Häselli, 1991; Röser, 1992; Neuweiler, 1997a;1997b).

Thus, the control of diseases, pestst and weeds is a major concern for farmers who are practicing this type of agriculture.

The strawberry crop (*Fragaria x annanasa* Dutch.), due to the high content of C vitamin, ß-caroten and other nutrients that are beneficial for health, has become one of the most popular crops worldwide, the total surface cultivated in 2017 being estimated, according to FAO, 395.844 ha (Monda, 2017).

The performances of different mulch systems on plant health and fruit yield are often inconsequent, and their effect seems to be related to the changes in microclimate (Passos, 1997; Maas, 1998).

Having a high susceptibility to water stress, the use of black plastic mulch plays an important role in soil moisture conservation, weed control and keep the fruits clean (Kasperbauer, 2002). In the same time adjusts the soil temperature, reduces water loss caused by evaporation, thus decreasing irrigation frequency, enhance nutrients absorption and provides a better growth and development of the roots compared to organic mulch or white polyethylene (Gupta

& Acharya, 1993; Khadas, 2014; Sharma, 2002; Ramakrishna, 2006).
Compared to other irrigation systems, to ensure the water requirements, drip irrigation presents a number of advantages which contribute to avoid formation of a humid microclimate favorable to diseases development, especially the fungal ones (Howard et al., 1992; Madden et al., 1993; Tanaka et al., 2005; Tanaka, 2002).
An important management tool against diseases and pests is soil solarisation, which is a process that increases the soil temperature due to the solar radiation using a plastic material. High temperatures will therefore be at the expense of pathogens from the soil (Katan, 1981).
The purpose of this paper is represented by the influence of mulch system on development of fungal diseases on strawberry crop and on agro-climatic factors.

MATERIALS AND METHODS

The experience was set up at the research station **Landwirtschaftskammer Nordrhein-Westfalen Köln-Auweiler**, Germany.
Köln area is bordered in West by the Bergisches Land mountainous region and in South by Eifel mountains and is characterized by a temperate climate with high humidity during the summer and gentle winters with oceanic influences. The pluviometric regime is characterized by an annual rainfall average of 676 mm.
According to german literature, the type of soil present in Köln-Düsseldorf region is represented by "Parabraunerde". This type of soil has been formed by the terraced deposition of alluviums, followed by the flooding of the area, by Rhein river, the upper layer having a clayey texture (Ameldung et al., 2018; Leitgeb et al., 2013).
In Federal Republic of Germany "Parabraunerde" together with "Fahlerden" belong to Luvisol (Lessivés) class within "Terestriche Böden" department (Landböden). "Parabraunerden" may also be known as loamy-illuvial brown soil.
These are characterized by an A humic horizon, intermediate horizon (AlBv) with a low content of clay, Bt loamy horizon and C horizon which is limestone with rich sediments of quartz and silicate.
After the analise of the soil profile of Landwirtschaftskammer Nordrhein-Westfalen Köln-Auweiler, the 0-200 cm layer is composed of roughly equal amounts of clayey and loamy soils.
The biological material used in the trial was represented by the varieties Rumba, Sonata and Clery, which belong to *Fragaria* x *ananassa* Dutch. species, wide spread in countries with advenced agriculture.
Planting was done on asparagus dam on 14.07.2017. Due to the large width of the dam, planting was done on two rows. The distance between plants was 30 x 30 cm and the distance between dam was aproximately 170 cm. To combat weeds between plants, the dam was covered with black polyethylene. Between dams, after plantig, was used against weeds, My-Pex, which is a waterproof material.
During the winter, in periods of near-frost temperatures, the plants were coated with a white microporous textile, to protect the strawberries against frost. In January 2018, one of the experimental fields was covered with transparent polyethylene, which is intended for protected field.
In March 2018 two microstations HOBO-MAN-H21-002 were installed, in both fields. The sensors for temperature and moisture were placed at 10 cm depth in soil. In field were recorded air temperature and rainfall too.
The recording of climatic parameters took place every 10 minutes and the data was downloaded at the end of the harvest period.
During the harvest period, observations were made on the following: the occurrence and evolution of strawberry diseases analyzing the fruits at each harvest, the monitoring of agro-climatic factors using the HOBO microstations, the amount of fruits affected by *Sphaeroteca macularis, Botrytis cinerea, Phytophtora fragariae* and *Colletotrichum fragariae.*

RESULTS AND DISCUTIONS

A. Climatic parameters

The thermal regime of the soil is influenced by a complex of factors, especially by the intensity of solar radiation and its periodic variations over time. In addition of these factors, the physical properties of the soil, composition, texture, degree of moisture or dryness of the soil, specific heat, thermal conductivity and the degree of soil cover with vegetation influence the evolution of soil temperature.

The soil temperature present significant variations between day and night in field, throughout the study period (figure 1). Thus, the black plastic mulch under the influence of solar radiation favors the increase of soil temperature during the day but its capacity to conserve it over night is low.

In tunnel, a better temperature preservation occurs, with monthly averages, higher than in the field. This is influenced by the presence of the second layer of plastic, used to create the protected space.

In tunnel, the temperature average in March starting with the station's installation date was 10.70°C, with a maximum of 16.25°C and a minimum of 7.16°C. In field, the mothly average recorded a value of 6.87°C while the maximum temperature was 21.17°C and the minimum was 0.88°C. In April, the monthly average recorded in field was 14.93°C, while in tunnel was with 1.30°C higher. The differences are also visible from the point of view of minimum and maximum temperatures.

The therrmal parameter of soil had also changed in May, when the monthly average was 20.23°C in tunnel and 19.71°C in field. Maximum temperatures registered have values between 21.61°C and 30.20°C, while the minimum temperatures are between 8.69°C and 12.61°C.

In June, monitoring and recording of agro-climatic factors did not took place throughout the entire month due to the end of harvest period. Thus, the monthly average recorded in tunnel until the 9th of June 2018, when the field were abolished, is 23.57°C with a maximum of 29.11°C and a minimum of 20.17°C. In open-field, until the crop abolish (16th June 2018), the monthly average was 21.58°C, with a minimum of 15.10°C and a maximum of 29.92°C.

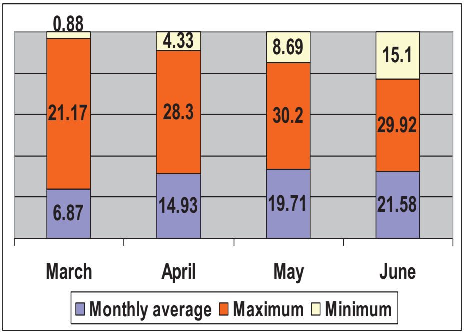

Figure 1. Evolution of soil temperature (°C) (Field, 2018)

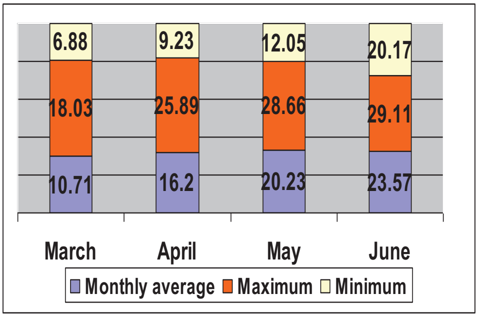

Figure 2. Evolution of soil temperature (°C) (Tunnel, 2018)

In this situation we can say that in case of tunnel, the use of second layer of polyethylene has the effect of reducing the temperature losses inside the dam both during the day and during the night, compared to field.

As regarding the soil moisture, there is a decrease of recorded values in both experimental fields starting with the microstation's installation date and ending with the moment of abolishing the fields, even if a constant irrigaton occurred.

The monthly averages recorded in tunnel had values between 0,197 m^3/m^3 and 0,174 m^3/m^3,while in field the values were between 0,191 m^3/m^3 and 0,153 m^3/m^3 (Figure 3). The influence of the second layer of plastic in case of the tunnel can also be observed in the evolution of soil moisture. Thus, the high temperatures inside the dam, in field have a direct impact on soil's moisture evolution.

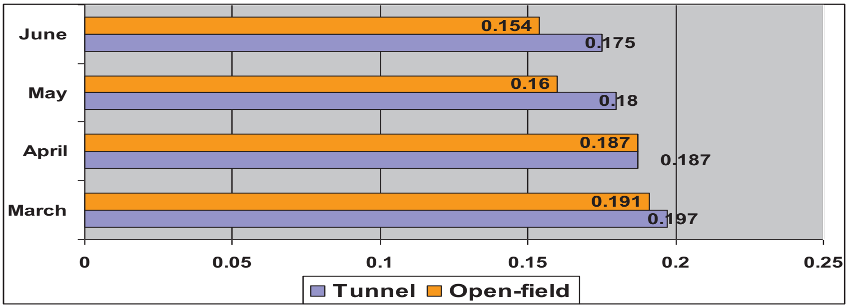

Figure 3. Monthly average of soil moisture (m^3/m^3) - 2018

The rainfalls peaked a maximum in March (56.40 mm), followed by April (48.60 mm) and June (42.31). From this point of view, the lowest amount of rainfall was recorded in May, with a volume of 18.60 mm.

The influence of precipitations on soil's humidity can be observed in April, when, with an amount of 48.60 mm, the soil moisture in field was close, as value, to the tunnel, compared with May and June.

The air temperature was monitorized only in field at a hight of 1 m. The monthly averages were between 6.17°C in March and 19.67°C in June. In April and May the monthly averages had values of 13.53°C respectively 17.73°C (Figure 4). The highest daily average was recorded in May (23.55°C).

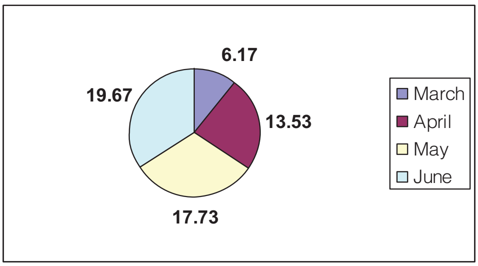

Figure 4. Air temperature in open field °C (2018)

B. Phytosanitary analysis

The most common strawberry diseases are caused by pathogens of a fungal nature, such as: leaf scorch, powdery mildew, anthracnose, grey mould, strawberry crown rot, strawberry red core. Their severity depends on the susceptibility of the variety, meteorological conditions and the degree of source infection (Müller, 1995; Jarvis, 1964; Meszka & Bielenin, 2011).

In the present study, during the vegetation period, were made observations on the attack of: *Botrytis cinerea, Phytophtora fragariae, Sphaerotheca macularis* and *Colletotrichum fragariae.*

According to the literature, the optimal conditions for germination of conidia and development of viruses are temperatures between 15-25°C, high relative moisture during the flowering and fruiting period, rainfall and excesive irrigation (Howard, 1972; Devaux, 1978).

Taking into account the mulching with black plastic, the higher incidence of diseased plants could be associated to a higher soil temperature compared with the clear plastic. For example, in case of anthracnose *(Colletotrichum fragariae)*, the high degree of attack using this system of mulching was attribute to the high soil temperature compared to other studied mulch systems (Camargo & Igue, 1973; Passos, 1997).

Fluctuations of agro-climatic factors influence the appearence and development of fungal diseases from a harvest to the other varying both the amount, in grams (g), and strawberry variety, in both trials (Tables 1, 2).

Due to the agro-climatic variations, the bigest amount of strawberries affected by *Phytophtora fragariae* and *Sphaeroteca macularis* were in tunnel. In field, due to the rainfall, was favorized the development of *Botritys cinerea*, being recorded the bigest amount of strawberries affected, followed by the attack of *Colletotrichum fragariae*.

Table 1. Response of the studied strawberry varieties to the diseases appeared in TUNNEL

Variety	Date of harvest	*Phytophtora fragariae* (g)	*Colletotrichum fragariae* (g)	*Botrytis cinerea* (g)	*Sphaeroteca macularis* (g)
Sonata	07.05.2018	290	0	64	0
Rumba	07.05.2018	507	0	0	0
Clery	07.05.2018	382	312	0	0
Sonata	10.05.2018	308	0	0	0
Rumba	10.05.2018	360	188	28	6
Clery	10.05.2018	44	158	10	0
Sonata	17.05.2018	94	142	34	118
Rumba	17.05.2018	105	203	21	10
Clery	17.05.2018	24	26	0	0
Sonata	29.05.2018	0	30	26	1,002
Rumba	29.05.2018	16	64	0	196
Clery	29.05.2018	0	12	0	0
Sonata	01.06.2018	0	62	0	184
Rumba	01.06.2018	0	10	0	6
Clery	01.06.2018	0	0	0	0
Sonata	05.06.2018	0	0	0	6
Rumba	05.06.2018	0	0	11	0
Clery	05.06.2018	0	0	0	0

Table 2. Response of the studied strawberry varieties to the diseases appeared in FIELD

Variety	Date of harvest	*Phytophtora fragariae* (g)	*Colletotrichum fragariae* (g)	*Botrytis cinerea* (g)	*Sphaeroteca macularis* (g)
Sonata	24.05.2018	57	47	74	21
Rumba	24.05.2018	442	26	90	24
Clery	24.05.2018	20	194	158	0
Sonata	29.05.2018	55	473	277	0
Rumba	29.05.2018	94	166	342	0
Clery	29.05.2018	0	286	568	0
Sonata	29.05.2018	55	473	277	0
Rumba	29.05.2018	94	166	342	0
Clery	29.05.2018	0	286	568	0
Sonata	01.06.2018	0	42	120	0
Rumba	01.06.2018	0	90	144	0
Clery	01.06.2018	10	180	412	0
Sonata	05.06.2018	178	575	824	5
Rumba	05.06.2018	48	126	1,018	0
Clery	05.06.2018	0	0	3,284	0
Sonata	08.06.2018	331	427	83	16
Rumba	08.06.2018	228	414	132	14
Clery	08.06.2018	372	230	148	2
Sonata	12.06.2018	0	6	96	0
Rumba	12.06.2018	0	18	292	0
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

In tunnel, during the harvest period, the bigest ammount of fruits were affected by *Phytophtora fragariae* at the begining of ripening due to the high moisture from the soil, while at the middle of harvest period, due to the increase of soil and ai temperature, were observed the highest attack of *Sphaeroteca macularis*. Comparing the field with the tunnel can be observed that, due to the rainfall, the ammount of strawberries affected by *Sphaeroteca macularis* is lower, while the attack of *Botrytis and Colletotrichum* is more pronounced to all varieties studied. The influence of agro-climatic factors and mulching system on the evolution of the studied diseases, is more pronounced in field over the entire period, from quatitive point of view (grams) as shown in Table 3. Also it vary, in the same time, the variety affected by each diseases studied.

Table 3. Total amount of strawberries affected by phytopathological agents

Variety	***Phytophtora cactorum* (g)**		***Colletotrichum fragariae* (g)**		***Botrytis cinerea* (g)**		***Sphaeroteca macularis* (g)**	
	Tunnel	**Open field**	**Tunnel**	**Open field**	**Tunnel**	**Open field**	**Tunnel**	**Open field**
Sonata	692	621	234	**1,571**	**124**	1,474	**1,310**	**41**
Rumba	**988**	**812**	465	840	60	2,018	218	38
Clery	450	402	**508**	890	10	**4,624**	0	0
TOTAL	**2,130**	**1,835**	**1,207**	**3,301**	**194**	**8,116**	**1,528**	**79**

CONCLUSIONS

Mulching with black plastic influences directly the evolution of soil moisture and temperature both in field and tunnel.

The conservation capacity of the studied agro-climatic factors and their fluctuations are influenced, in case of the tunnel conditions, by the presence of the second layer of plastic used to create the protected space.

Analyzing the optimal conditions for development of phatogenic agents, the use of black plastic favors their evolution in field, due to the highest temperatures over the day in relation with atmospheric humidity.

From phytosanitary point of view, the most pronounced attack of *Phytophtora fragariae* and *Sphaeroteca macularis* was in tunnel, while the strongest attack of *Colletotrichum fragariae* and *Botrytis cinerea* was in field.

The greatest amount of strawberry affected by fungal diseases was recorded in field compared to the tunnel.

ACKNOWLEDGEMENTS

This work was supported by a grant of the Ministery of Research an Innovation, CCCDI-UEFISCDI, project number PN-III-P1-1.2-PCCDI-2017-0056: Functional collaboration model between public research organizations and the economic environment for the provision of high-level scientific and technological services in the field of bio-economy, within PNCDI III.

REFERENCES

Amelung, W., Blume, H.P., Fleige, H., Horn, R., Kandeler, E., Kögel-Knabner, I., Kretschmar, R., Stahr, K., Wilke, B.M. (2018). *Scheffer/Schachtschabel Lehrbuch der Bodenkunde*. 17. Auflage. Heidelberg, DE.

Camargo, L.S.; Igue, T. (1973). Brangatia: Experiência sobre efeito da cobertura do solo na produção do morangueiro, 32, 149–169.

Devaux, A. (1978). Etude epidemiologique de la moississure grise der fraies et essais de lutte. *Phytoprotection, 59*, 19–27.

Graf, B., & Häseli, A. (1991). Eignung einiger Erdbeersorten für den Biologischen Anbau. 4. Internat. Erfahrungsaustausch über Forschungsergebnisse zum Ökologischen Obstbau, 75–78.

Gupta, R., & Acharya, C.L. (1993). Effect of mulch induced hydrothermal regime on root growth, water use efficiency, yield and quality of strawberry. *Journal of Indian Society of Soil Science*, *41*, 17–25.

Howard, C.M. (1972). A strawberry fruit rot casued by *Colletotrichum fragariae*. *Phytopathology, 62*, 600‒602

Howard, C.M., Maas, J.L., Chandler, C.K., Albregts, E.E. (1992). Anthracnose of strawberry caused by *Colletotrichum* complex in Florida. *Plant Disease*, *76*, 976‒981.

Jarvis, W.R. (1964). The effect of some climatic factors on the incidence of grey mould of strawberry and raspberry fruit. *Hortic. Res., 3*, 65‒71.

Katan, J. (1981). Solar heating (solarization) of the soil for control of soilborne pests. *Annual Review of Phytopathology, 19*, 211–236.

Kasperbauer, M. (2000). Strawberry-Yield over red versus black plastic mulch. *Crop' Science, 40*, 171‒174.

Khadas, O.A. (2014). Effect of different irrigation levels on growth and yield of strawberry under silver black mulch. Ph.D. dissertation, College of Agricultural Engineering and Technology, Dr. BSKKV, Dapoli, India.

Leitgeb, E., Reiter, R., Englisch, M., Lüscher, P., Schad, P., Feger, K.H. (2013), *Waldböden. Ein Bildatlas der wichtigsten Bodentypen aus Österreich, Deutschland und der Schweiz.* Wiley-VCH Verlag GmbH, Weinheim, DE.

Maas, J.L. (1998). *Compendium of strawberry diseases*. 2.ed. St. Paul: APS Press, 128.

Madden, L.V., Wilson, L.L., Ellis, M.A. (1993). Field spread of anthracnose fruit rot of strawberry in relation to ground cover and ambient weather conditions. *Plant Disease, 77*, 861‒866.

Meszka, B.M & Bielenin, A. (2011). Activity of laminarin in control of strawberry diseases, www.aupoznan.pl/ptfit/pdf/P62/P62_02.pdf.

Monda, D. (2017). Possibility of Strawberry Cultivation in Bangladesh. An Assignment on Strawberry, Department of Business Administration, Northern University, Dhaka, Bangladesh, https://www.scribd.com/doc/.../Possibility-of-Strawberry-Cultivation-in-Bangladesh.

Müller, H.W.K. (1965). The present state of grey mould (*Botrytis cinerea*) control in strawberry growing. *Rev. Appl. Mycol., 44*(8), 408.

Neuweiler, R. (1997a). Erdbeersorten - Fruchtqualität und Robustheit stehen im Vordergrund. *Schweizerische Zeitschrift für Obst- und Weinbau, 133*, 62–64.

Neuweiler, R. (1997b). New strawberry cultivars in Swiss integrated fruit production. *Acta Horticulturae*, *439*, 951–955.

Passos, F.A. (1997). Influência de alguns sistemas de cultivo na cultura do morango (Fragaria X ananassa Duch.). Piracicaba: *USP/ESALQ, 106* (Doutorado).

Ramakrishna, A., Hoang, M.T., Wani, S.P., Ding, T.L. (2006). Effect of mulch on soil temperature, moisture, weed infestation, and yield of groundnut in northern Vietnam. *Field Crop. Res., 95*, 115.

Röser, I. (1992). Leistungsvergleich bei neuen Erdbeersorten mit und ohne Pflanzenschutzbehandlung im 1 jährigen Anbau 1992. 5. Internat. Erfahrungsaustausch über Forschungsergebnisse zum ökologischen Obstbau, 114‒117.

Sharma, R.R. (2002). Growing Strawberries, International Book Distributing Co., Lucknow, India.

Tanaka, M.A.S. (2002). Controle de doenças causadas por fungos e bactéria em morangueiro. In: Zambolim, L., Vale, F.X.R., Monteiro, A.J.A., Costa, H. (Ed.). *Controle de doenças de plantas - Fruteiras. Viçosa: UFV*, 69‒139.

Tanaka, M.A.S., Betti, J.A., Kimati, H. (2005). Doenças do morangueiro - *Fragaria* x *ananassa* Duch. In: Kimati, H., Amorim, L., Bergamim Filho, A., Rezende, J.A., Camargo, L.E.A. (Ed.). *Manual de fitopatologia.* 3.ed. São Paulo: Agronômica Ceres, 2, 556–571.

Willer, H., Rohwedder, M., Wynen, E., (2009). Current statistics. In: Willer H, Kilcher L, editors. The World of Organic Agriculture - Statistics and Emerging Trends. Bonn: IFOAM, 25–58.

Willer, H., Lernoud J., (eds.) (2016). The world of organic agriculture. Statistics and emerging trends 2016. Research Institute of Organic Agriculture (FiBL), Frick, IFOAM - Organics International, Bonn, Germany.

Willer, H, Lernoud, J. (eds.) (2017). The world of organic agriculture. Statistics and emerging trends 2017. Research Institute of Organic Agriculture (FiBL), Frick, IFOAM - Organics International, Bonn, Germany. Version 1.3 of Feb 20.

*** Codex Alimentarius Commission, Joint FAO/WHO Food Standards Programme (1999). Guidelines for the production, processing, labelling and marketing of organically produced foods. GL32-1999. Rome, Italy.

*** http://www.fao.org/faostat/en/#data/QC

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EVALUATION OF PRECIOUS ECOLOGICAL CONDITIONS FOR FRUIT TREES PLANTATIONS IN THE MIDDLE FARM FIELD

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Abstract

Research over three years has allowed us to highlight the extension of the fruit plantations in Glodeni district. In the area of fruit plantations, the apple showed 98.1% of the total area of the trees. The structure of the species of fruit plantations does not correspond to the requirements of the national economy and requires optimization through the ecological reconstruction of the existing orchards. All orchard soils have at the top of the profile a dilapidated layer with an average depth of 57.3%, non-homogeneous by color, structure and other morphological indices, with physicochemical properties and nutrient assurance, which mostly determines the spreading the root system and the productivity of the fruit plantations. It was highlighted that the 3 years average values of real production were lower than the potential one, which requires more favorable conditions for fruit plantations.

***Key words**: agro ecology, tree species, fertility, pretability.*

INTRODUCTION

The efficient exploitation of the ecological, technological and socio-economic conditions of each unit or area is one of the main objectives of agricultural science and practice, fruit growing being an important place in the creation of agricultural resources, being known from ancient times one of the branches main and economically efficient agriculture in our country. Fruit growing previously occupied approx. 6-7% of the area of agricultural land, providing 20% the benefit from the marketing of agricultural production. Average fruit production amounted to 950 thousand tons per year, while the average production per hectare was - 7.3 tons.

Assessing the condition created in this basic branch of the agro-industrial complex, there is a sharp decrease in the global fruit harvest averaging 2.5 times. Under these conditions, it is necessary to carry out profound research on the substantiation of the sustainable development prospects of the fruit growing branch (Babuc, 2012; Dadu, 2011).

Currently in the Republic of Moldova most of the fruit plantations have exceeded the age of 20-30 years, they can be more easily affected by changes in the environment: climate, relief, hydrology, soil etc., with low production potential and reduced exploitation value. In accordance with the concept of sustainable fruit growing, the strategic direction consists in the efficient exploitation of existing orchards with the potential for unproductive productivity and their gradual replacement with new type orchards with modern assortment and modern technologies adapted to the concrete production conditions for efficient use of the ecological, biological, technological, economic potential of each land sector and agricultural enterprises (Babuc, 2012; Dadu, 2011; Ursu, 2006; 2011).

The purpose of the research was to monitor the ecological conditions and to highlight their suitability for the fruit trees in the Glodeni district. Research objectives: highlighting the structure of existing fruit plantations; the characterization of ecological conditions and the assessment of the productivity of fruit trees for the scientific argumentation of ecological micrororionation of fruit growing, which contributes to the more efficient utilization of the natural resources of the Glodeni district located in the Middle Prut Plain.

MATERIALS AND METHODS

Research methods are accepted in organic research and fruit growing. For the characteri-zation of climatic indices, published materials of the State Hydro meteorological Service of the Republic of Moldova (BSHS, 2015-2017; Pedological Report, 2011) were used. The elements of the relief were established as a result of the expeditionary observations in the field and on the basis of the pedological research materials of the Research Institute for Territorial Organization of the Republic of Moldova (Pedological Report, 2011). The morphological characteristics of the soils in the fruit plantations have been studied in the field. The soil samples in the laboratory were determined: hygroscopic humidity; soil texture; the content of humus by the Tiurin method in the modification of Simacov; carbonate content - gas-volumetric method; determining the content of Ca^{++} and Mg^{++}; bulk density-the metal cylinders method (Andriuca, 2015; Ursu, 2006).

Global harvest by species was determined 15-20 days before harvesting after the amount of fruit normally grown on 25 typical trees per hectare. On each tree, choosing 1-2 skeleton branches, which is equivalent to a crown sector (1/4) on which the fruit was weighed. Then the fruit mass was calculated at a tree and in tones' per hectare (Babuc, 2012).

RESULTS AND DISCUSSIONS

The plots of fruit plantations in the Glodeni district extend over an area of 2239.2 ha. Of the total area of the fruit plantations, the weight of the apple species is 91.8% (2057 ha), the cherry species is only 6.6% (150 ha) and the cherry 0.9% (19 ha) and the apricot 0.6% (13 ha). The proportion of fruit trees in existing orchards differs from the proportion of national economy requirements, which provide for the northern fruit plant: apple - 68%, hair - 12%, plum - 12%, cherry - 4%.

All fruit trees are privately owned - in the agricultural cooperative production (CAP) - 22.5 ha, in limited liability companies (LLC) - 1561.3 ha, peasant farms - 328.6 ha.

Relief and hydrographic network. The fruit plantations are located on various relief elements, plateau, slope with different north-western, west, south-western, southern, 1-3°, 3-5°, 5-7° inclined to the upper, middle and lower the slopes, the valleys of the Prut River, the smaller rivers Camenca, Căldăruşa, Ustia. The altitude of the relief elements varies from 90 m to 200 m above sea level. Reluctant relief causes the distribution of environmental factors. On flat fields, light, heat, water, nutrients, wind intensity are almost evenly distributed. On the sloping land, biotope elements record variations, which in turn generate variations in growth, tree metabolism, productive potential - quantitative and qualitative etc. As a whole, the relief elements of the locality cause the formation of three types of microclimate: moderate cold with the sum of the active temperatures Σ ta $\geq$ 10°C and above 2888-2934°C on the altitudes of 180-200 m above sea level; moderately warm with the sum of the active temperatures Σ ta $\geq$ 10°C and above 2957-3049°C on the slopes: E, 1-3°, upper, altitude 130-170 m, SE, 3-5°, middle slope altitude 130-170 m; SW, 5-7° mid, altitude 130-170 m; warm with the sum of active temperatures 3072-3164°C on the SW slopes, 5-7° medium, altitude 80-120 m.

The deviation of the fruit trees on the slopes from those on the plateau was up to 253°C. Soil varieties on the fruit trees are also different: argiloiluvial chernozem, chernozem leveled strongly deep lute-clayey on the plateau with an altitude of 180-200 m. On the slopes or highlighted leached chernozems weak and moderately eroded clay-clayey and clayey; moderate and mildly moderate to moderate deep chernozems, and deeply clay-clayey chernozems. On all the soils of the fruit plantations, a very pronounced layer degraded around the depth of 0-60 cm, which is very inhomogeneous according to their color and structure, with the succession of the genetic horizons interrupted due to their techno genic mixture, ie they are technogenode soils - formed with precipitations physic-chemical modifications.

Characteristic of the ecological conditions of the fruit plantations in the Glodeni district.

Climate conditions over the five years of research have been characterized by different thermal humidity regimes. The average annual temperature increased from 9.3°C (2014) to 10.5°C (2015), and in the following years it remained at the same level of 9.8°C. The humidity regime of Glodeni ranged from 382 mm (2015) to 69 mm (2014) years. In 2017 the rainfall was recorded with a moderate value of 578 mm compared to 2016-602 mm and 2018-615 mm (Figure 1).

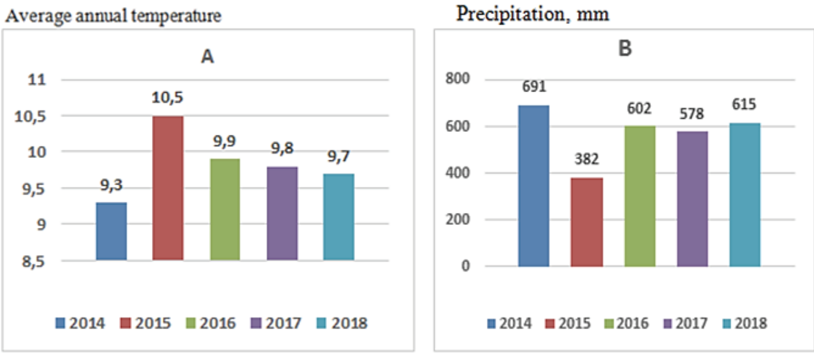

Figure 1. The thermal (A) and humidity (B) of the territory, Glodeni district, 2014-2018 (Meteorological Station Bălți)

Based on these results, the most dry years - 2015, wetter - 2014 and moderate - 2017, after the heat and humidity regime, 2017 were highlighted.

According to the particularities of these years the thermal moisture regimes on the territory of the rayon in the vegetation period (Figures 2 and 3).

The deviation of the fruit trees on the slopes from those on the plateau was up to 253°C. Soil varieties on the fruit trees are also different: argiloiluvial chernozem, chernozem leveled strongly deep lute-clayey on the plateau with an altitude of 180-200 m.

On the slopes or highlighted leached chernozems weak and moderately eroded clay-clayey and clayey; typical mild chernozems and strongly humid strongly profound and deeply clay-clayey chernozems.

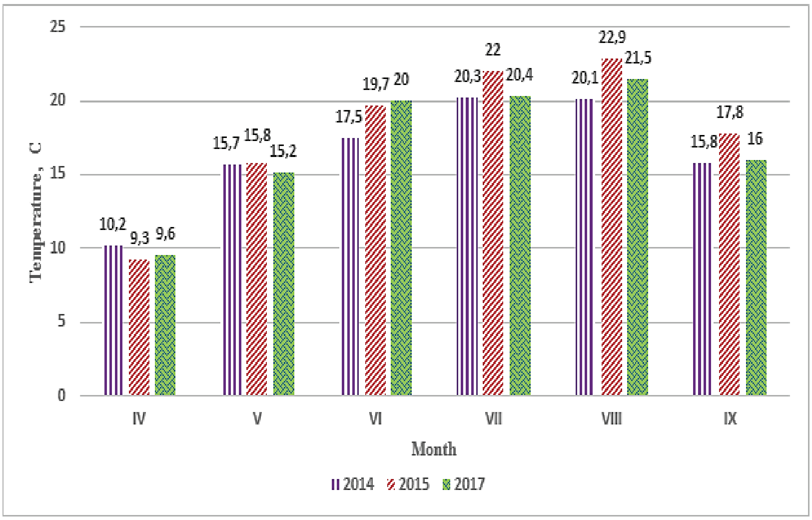

Figure 2. The thermal regime of the territory of Glodeni district during the vegetation period

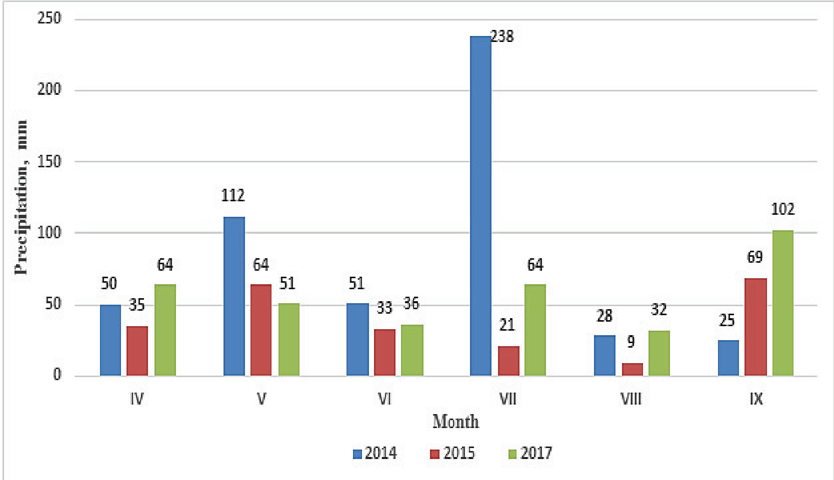

Figure 3. The precipitation regime during the vegetation period in the Glodeni district

On all the soils of the fruit plantations, a very pronounced layer degraded around the depth of 0-60 cm, which is very inhomogeneous according to their color and structure, with the succession of the genetic horizons interrupted due to their techno genic mixture, ie they are technogenode soils - formed with precipitations physico-chemical modifications.

From the data analysis, it is observed that the strongly clay-clayey chernozem is character-rized by a content of physical clay within the limits of 53.1 without significant changes in profile. The bulk density in the loose layer was recorded at a level of 1.25 g/cm^3 optimal for the penetration and penetration of the fruit roots, moderate humus of 3.35%. Ensured with nitric nitrogen Na-NO_3, phosphorus - P_2O_5 and potassium - K_2O is relatively optimal for fruit crops.

As a result of the complex analysis of the climatic, microclimatic, relief and soil condi-tions of the fruit plantations, three ecological fruit trees were found, which showed a different level of fruit productivity (Table 1).

Table 1. Productivity of the fruit species depending on the ecological conditions of the fruit trees in the Glodeni district, 2014-2018

Species	*Ecopedological conditions*	*Harvest, t/ha*		
		minimum	*maximum*	*average*
I microraion				
Apple 327 ha	Plate 200 m, Σ t ° ≥ 2888 °C, moderate cold, argiloiluvial chernozem, strongly deep lute-clayey	11.1	16.5	13.9
Cherry tree 46 ha		5.6	7.7	6.7
Cherry 19 ha		4.7	5.3	5.0
Apricots 13 ha		5.2	6.6	6.0
II microraion				
Apple 439 ha	Versions NV, V, SE, SV upper and middle 1-3° and 3-5°, 130-175 m Σ t ° ≥ 2950 - 3050 °C, moderately warm, weakly and moderately eroded lemon chernozem, typical moderate chernozem and low humidity, deep clay and lotus	7.4	13.8	10.2
Cherry tree 42 ha		6.5	8.6	7.5
III microraion				
Apple 80 ha	Versions S, Lower, 1-3°, 80-125 m, warm Melting point: 3072-3164°C, strong deep clay, clay chernozems	7.2	7.9	7.5
Cherry tree 10 ha		6.3	8.2	7.1

The deep-lute chernozems in the fruit plantations have a 49.8% physical clay content, the bulk density values of 1.23-1.29 g/cm^3, indicate good conditions for the penetration and development of the fruit tree roots. Higher humus content - 4.15% (increased), the nitrate content is estimated to be high (over (4.5 mg), relatively optimal for phosphorus (5.4 mg) and potassium (29 mg/100 g soil). Moderately eroded lemon chernozems have the 50.0% clay content of the clay-loamy texture of the soil. Bulk density values (1.29 g/cm^3) are optimal plant growth limits (1.1-1.3 g/cm^3). The humus content is 2.82%. The content of mobile forms of plant-accessible substances of moderate nitrates (3.3 mg), low phosphorus (2.3 mg) and moderate potassium 11.8 mg/100 g soil.

Typical moderate humid humorous moderate profound clay-clayey clay has a 47.9% physical clay content characteristic of the clay-clay soil variety. The bulk density in the loose layer - 1.20 g/cm^3 is considered to be optimal for root expansion and development, as the restrictive limits are 1.4-1.5 g/cm^3. Humus content 4.67% (increased). The content of mobile forms of substances is moderate for NO_3 (6.2 mg), P_2O_5 (3.2 mg), K_2O (15.2 mg/100 g soil).

The typical low humus-rich clay-humus typical chernozem has a physical clay content of the variety (45-60%). Bulk density 1.22 g/cm^3 - optimal for penetration and development of fruit plants. The humus content - 3.15% in the sloped layer is considered to be relatively optimal for the fruit plants. The sum of their exchange cations (basics) in the value of 31.93 me/100 g soil condition the good humus fixation and the long-lasting maintenance of its quantity and quality. Reaction of the soil solution, pH 7.5, low alkaline pH in the loose layer. The nitrification capacity in the high depleted layer over 4.5 mg/100 g of soil (6.5 mg/100 g of soil), the moderate mobile phosphorus content (2.6 mg/100 g of soil), the potassium content 12.9 mg/100 g soil also moderate.

The deeply clay-clayey chernozem is characterized by a clay content within the limestone-clay range (45-60%). The bulk density of 1.24 g/cm^3 is optimal for fruit plants. Low humus content (2.57%). The amount of exchange bases 30.75 me/100g soil, characteristic of the soil subtype and the chernozem layer of humerus. Reaction of weak alkaline soil to moderately alkaline soil. Content of mobile NO_3 - increased form (6.9 ml/100 g soil).

One of the main clues to characterize the efficiency of using natural resources and the application of performing processes is the productivity of cultivated plants. The harvests of the fruit crops in the Glodeni district during the years 2015-2017 were varied depending on ecological conditions.

In micronorade III the fruit maturation was recorded 8-11 days earlier compared to the micro world I on the plateau. The level of lower apple and cherry yields under the conditions of micorotherion II may be due to the degree of erosion of soils, which is characterized by the reduction of the humus-accumulative fertile horizon to the unroasted soil and the moderate thickness of some soil soils.

In the ecological conditions of the plateau the altitude 180-200 m, the moderate cold microclimate, the argiloiluvial chernozem soils and the strongly dewormed clay-clayey chernozem, the apple harvest was higher (13.9 t/ha) compared to the apple on the slopes (microdoraion II and III).

CONCLUSIONS

The fruit plantations in the Glodeni district are located on various relief elements (plateau, slopes with 1-3-3...1-3 slope, northern, eastern, southern, western, 80-200 m altitudes) in different thermal microclimate, moderately hot and warm, chernozems soils: moderate and weak humid, strong and moderately deep carbonate, weakly and moderately eroded, which can be divided into three ecological fruit trees.

The productivity of fruit plantations has been established in the years of research (2014-2018), which has been characterized by different quantitative indices on species, depending on the ecological conditions of microdorations.

Some limiting factors of fruit tree productivity have been highlighted: physical clay content, compaction, depth and carbonate content, degree of erosion.

Fruit plants are very complex biological systems and to use them in the interest of man, it is necessary to know them. In order to establish the appropriate crop technology necessary to obtain maximum yields for each species, it is necessary to know the organography and the requirements of each fruit tree species.

Sustainable development of fruit growing requires very large investments, which will be covered by the private sector, through allocations from the state budget and foreign investments. Current programs too provides for the creation of a state fund for the development of fruit production in Republic of Moldova.

Sustainable development of fruit growing requires gradual replacement of orchards exhausted with intensive and super intensive orchards with productivity potential and fruit quality 1.3-1.5 times higher than the previous level in the base rational use of ecological, biological, technological, and economic resources characteristic of each field sector and fruit growers.

REFERENCES

Andrieș, S., Cerbari, V. (2001). *Programul Național complex de sporire a fertilității solului*. Chișinău, Pontos, 76–92.

Andriuca, V. (2015). *Lucrări metodice la disciplina Ecopedologie* pentru specialitatea - 424.1 Ecologie. Chișinău, C.E. UASM, 54.

Babuc, V. (2012). *Pomicultura*. Chișinău, 80–105.

Cerbari, V. (2010). *Monitoringul calității solurilor Republicii Moldova*. Chișinău, Pontos, 45–57.

Dadu, C. (2011). Reflecții economice privind dezvoltarea pomiculturii. În: *Agricultura Moldovei,* Chișinău, *8-9*. 5–7.

Ursu, A. (2006). Raioanele pedogeografice și particularitățile regionale de utilizare și protejare a solurilor. *Chișinău, Știința, 30-51*, 64–69.

Ursu, A. (2011). Solurile Moldovei. *Chișinău, Știința, 36*, 205–206, 217–225.

***Buletinul Serviciului Hidrometeorologic de Stat al Republicii Moldova (2015-2017). Chișinău, 72–78.

***Materialele dărilor de seamă a primăriei localităților raionului Glodeni (2015-2017). Secția Agricolă Glodeni.

**Raportul Pedologic. Solurile raionului Glodeni și utilizarea lor rațională (2011). Chișinău, ICTOTRF, 102.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SPATIAL DISTRIBUTION OF HEAVY METALS IN AGRICULTURAL SOILS OF ROMANIA: REVIEW OF THE CURRENT SITUATION

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Abstract

Contamination of soils and crops by heavy metals is considered one of the most serious environmental problem due to their non-biodegradable nature, the long biological half-life and also their potential accumulation in different body parts of plants. In Romania there are areas of thousands of hectares of agricultural land polluted with heavy metals. The distribution of heavy metals in agricultural soil can serve as a basis for a better management of soil quality, as well as to protect human health and the soil environment. This paper aims to present an overview based on the data found in other studies and to obtain a mapping of each the most common heavy metals met in soils of Romania. Due to the fact that there are not enough studies for the whole country, some areas will be estimated and also, is possible that others will not be covered at all.

Key words*: heavy metals, mapping, pollution, Romania, soil contamination.*

INTRODUCTION

It is a well-known fact that heavy metals are part of the soil structure, and they occur there naturally. Depending on their type and concentration, these metals can be harmful to environment, and further, to human health.
Besides the natural causes (erosions, volcanic eruptions) (Cyraniak et al., 2014), the heavy metal pollution can also originate from human activities like mining industry, transportation, urbanization, agriculture and others (Dumitrel et al., 2013). With a few exceptions (some sites with high levels of natural resources), the pollution in Romania was caused by population, through unmonitored industry centers (Boros et al., 2015; Muntean et al., 2010; Sur et al., 2012).
Although in general Romania has safe areas from agricultural point of view, there are still some sites where the heavy metal pollution exceeds the limits provided by laws in force. Even more, soil is considered as a key source of socio-economic development (Dimitriu, 2014).
The only study that offers complete information about the levels of heavy metals in all areas of Romania was conducted by ICPA (National Research and Development Institute for Soil Science, Agrochemistry and Environment) in 2000. The network of soil monitoring sites uses 942 investigation points spread in all the country, as 16 x 16 km squares. For this reason, the points rarely hit areas with severe problems regarding pollution. Yet, this kind of approach is most representative for soil characterization.
A similar report was published in 2011 by the same institution (Soil Quality Monitoring in Romania, Dumitru et al., 2011) but it does not provide the same detailed information.
Using this study as a base, the current review aims to update the existing data with others found in most recent studies, and also to add the plots with high values of pollutants found in other studies as well, to form some maps as accurately as possible.

MATERIALS AND METHODS

The most common heavy metals that are polluting the soil are cadmium, lead, nickel, chromium, zinc, cobalt, copper, arsenic (Masindi et al., 2017).
In order to create the map distributions for some of these metals, some parameters that describe the levels of interest from current law were used (Table 1).

Using the data from ICPA report, six distribution maps were created, one for each element shown above in the Table 1. Also, a table with some statistical data was created for each element. Only two studies that have expanded their research on the entire country were found.

Table 1. Reference values for trace chemical elements in soil (Order 756, 1997)

Element	Reference value (ppm)	Alert threshold sensitive soils (ppm)	Intervention threshold sensitive soils
Zn	100	300	600
Cd	1	3	5
Cu	20	100	200
Pb	20	50	100
Ni	20	75	150
Co	15	30	50

Apart from ICPA report, another study was performed with the same scope, in 2012 (Moldoveanu, 2014). It is interesting that the second study analyses a large range of heavy metals from both urban and rural areas. Unfortunately, there are some areas that are not covered at all. Since it not provides any numeric values, a direct comparison can hardly be made.

Other assays that have limited their research on a small area provided some valuable information to update the map distribution. All the studies that have been analysed had the same mode for preparing the soil samples: depth 0-20 cm, dried, grinded, sieved and analysed through different detection methods.

RESULTS AND DISCUSSIONS

Each metal has been analysed separately in order to structure the information efficiently.

Zinc

Zinc is an essential micronutrient required by plants and it has multiple role in their development (Hassan et al., 2017). In many cases, the zinc deficiency is a very important problem that requires different amendments to raise its concentration (Berbecea et al., 2011). But there are some cases that due to different anthropogenic activities such as mining or industrial emissions the situation can be reversed (Dumitrel et al., 2013).

In Romania exists a few areas where the concentration of zinc exceeds both, alert or intervention threshold.

A series of studies highlights the zones who have encountered this problem and also offers some quantitative values (Buruiana et al., 2016; Dumitrel et al., 2013; Ene et al., 2010; Marin et al., 2010; Munteanu et al., 2012; Sur et al., 2012; Elekes, 2014; Nimirciag, 2012; Albulescu et al., 2012; Popa et al., 2016; Big et al., 2012; Gămăneci et al., 2011; Morar et al., 2010; Stefu et al., 2013; Ungureanu et al., 2017; Fâciu et al., 2012; Cojoc, 2011; Lăcătușu et al., 2009). These levels are shown in the table below, together with the average values from ICPA report (Table 2). The table contains only the counties were zinc levels were studied in recent works.

Table 2. Zinc concentration in different areas (ppm)

County	ICPA study					Other studies					
	Mean	Number of sites	Median	Min	Max	Mean	Number of sites	Median	Min	Max	Year
Alba	153.59	27	100	41	540	834	15	725	100	3997	2013
Bacau	51.27	26	50.5	20	111	75.48	95	-	24.97	126.38	2013
B Nasaud	84.19	21	65	35	275	578.14	35	197.5	58.7	5144.2	2012
Braila	84.38	21	60	32	235	88.80	3	92.75	70.80	102.86	2009
Caras-Severin	138.42	33	83	45	500	98.61	9	53.9	29.1	294	2012
Dambovita	99	18	7538		215	125.5	5	36.5	22	600.4	2014
Galati	155.06	16	170	25	250	57.66	8	58.16	33.62	72.69	2009
Gorj	68.91	22	65	37	110	61.56	17	52	31.8	133	2016
Iasi	83.05	22	49	23	405	45.36	1030	30.31	11.6	702.61	2008
Maramures	210.20	25	230	5	360	311.44	10	144.38	89.04	1110.2	2012
Mures	91.96	27	70	40	270	69.87	5	56.6	53.4	99.43	2010
Sibiu	95.75	20	68.5	35	255	463.4	94	-	64.04	1938	2011
Suceava	89.60	35	68	23	285	95.8	63	-	33.6	332.8	2013
Timis	193.06	34	200	36	500	60.43	18	57.62	39.82	98.72	2013
Vaslui	62.74	23	56	34	118	68.22	193	63	31	192	2017

Comparing the average values from ICPA study with the average values from other studies, it shows a significant difference between these set of values (T test result: 0.15). The difference is somehow explainable regarding the fact that most of studies directed their research on areas with problems. These areas are pointed out in the figure below (Figure 1). From a total of 1043 investigated sites, 767 (73.53%) have a smaller concentration of zinc than the reference value, 246 (23.58%) between reference value and alert threshold, 79 sites (7.57%) exceeds alert threshold and only 18 (1.72%) exceeds intervention threshold. These top values represent some well-known high polluted areas: Zlatna (Alba): 3997 ppm, Certej (Hunedoara): 2200 ppm.

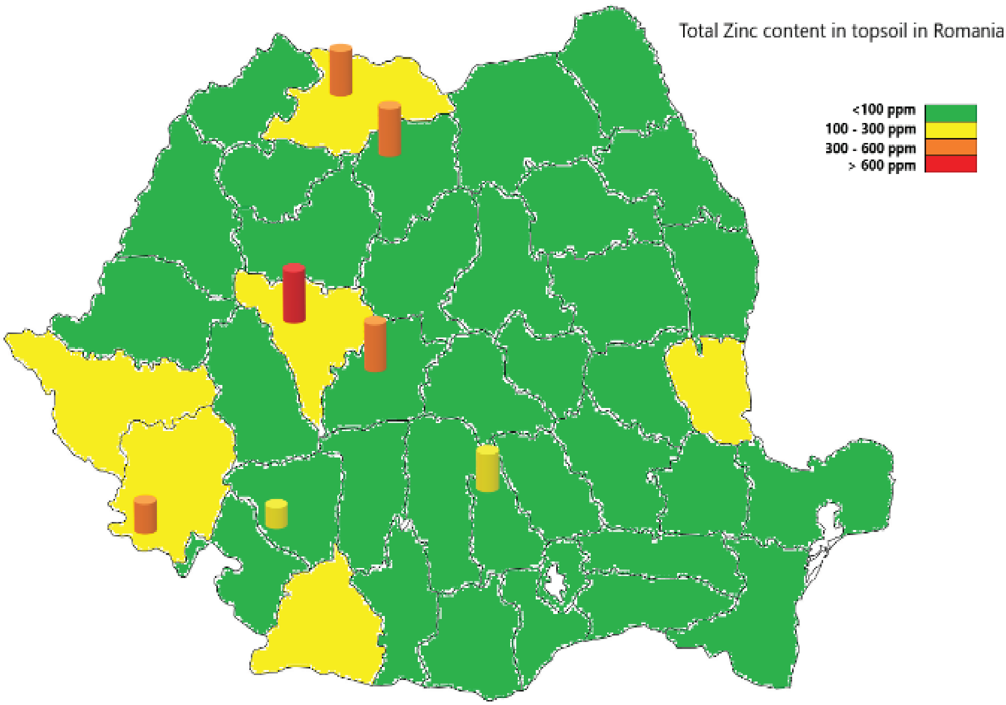

Figure 1. Zinc content in topsoil of Romania counties

Copper

Alongside zinc, copper is an important element in development of plants as well. Copper has an important role in metabolism and also participates to the respiration process (Laţo et al., 2012). Generally, a level of 4-20 ppm Cu is sufficient for any type of culture (Sutradhar et al., 2017).

A deficiency of copper can be possible as well, and this is one of the major constraints for crop productivity in many countries of the world (Corches et al., 2017).

The availability of copper depends on soil characteristics, such as organic carbon content, texture and pH and threshold values have been established as functions of these soil properties (Ballabio et al., 2018).

In Romania, almost all areas have sufficient quantities of copper, in some of them being in excess.

In the table below are presented the counties that have been mentioned in recent studies (Dinu et al., 2018; Suciu et al., 2008; Zgripcea, 2013; Popescu et al., 2013; Morar et al., 2010; Buruiana et al., 2016; Dumitrel et al., 2013; Ene et al., 2010; Marin et al., 2010; Munteanu et al., 2012; Sur et al., 2012; Elekes, 2014; Nimirciag, 2012; Albulescu et al., 2012; Popa et al., 2016; Big et al., 2012; Gămăneci et al., 2011; Morar et al., 2010; Stefu et al., 2013; Ungureanu et al., 2017; Hura et al., 2013; Iancu et al., 2008) to have this problem (Table 3).

Table 3. Copper concentration in different areas (ppm)

County	ICPA study					Other studies					
	Mean	Number of sites	Median	Min	Max	Mean	Number of sites	Median	Min	Max	Year
Alba	24.80	25	25	6	46	223.95	16	82.18	18.7	914.1	2013
Bacau	16.90	29	15	9	33	35.84	89	-	12.1	82.1	2013
B Nasaud	25.33	21	20	10	68	43.37	33	36.9	15.2	102.8	2012
Braila	31.14	21	25	12	125	33.87	6	31.28	25.06	52.78	2009
Caras-Severin	25.15	33	23	10	60	58.92	9	56.66	25.06	52.78	2012
Cluj	21.62	26	20	10	32	46.26	18	47.44	28.2	52.84	2008
Dambovita	20.47	17	20	6	50	125.5	5	36.5	22.0	600.4	2014
Galati	25.06	16	22.5	15	75	24.54	14	23.96	18.38	31.59	2009
Gorj	27.75	20	21	15	114	11.31	12	7.67	1	34.34	2015
Hunedoara	21.81	32	15	10	59	100.21	6	44.85	15.3	378	2018
Iasi	23.30	23	21	8	46	45.36	1030	30.31	11.6	702.61	2008
Maramures	19.52	25	20	5	40	112.98	10	80.11	33.29	310.82	2012
Mures	26.56	27	25	10	43	22.41	18	17.5	1.28	72.8	2008
Sibiu	26.38	21	25	10	49	31.73	56	-	9.03	114.6	2011
Suceava	23.42	36	20	8	75	36	63	-	17.85	112.75	2013
Timis	19.14	35	17	8	45	26.22	18	30.89	0	49.44	2013
Valcea	33.14	22	25	10	100	32.86	6	33.85	10.48	53	2013
Vaslui	23.26	23	21	8	45	29.83	193	27	14	300	2017

The average values from ICPA study and the average values from other studies are significantly different (T test result: 0.034).

Some major differences are shown on the map below (Figure 2).

A total of 1103 points were analysed. Less than half, 418 (37.89%) had a copper level smaller than the reference value, 663 (60.10%) points were situated between reference value and alert threshold, 14 (1.26%) points exceeds alert threshold but not the intervention threshold, and 7 (0.63%) exceeds intervention threshold. The highest recorded value is 914.1 ppm, near Zlatna region.

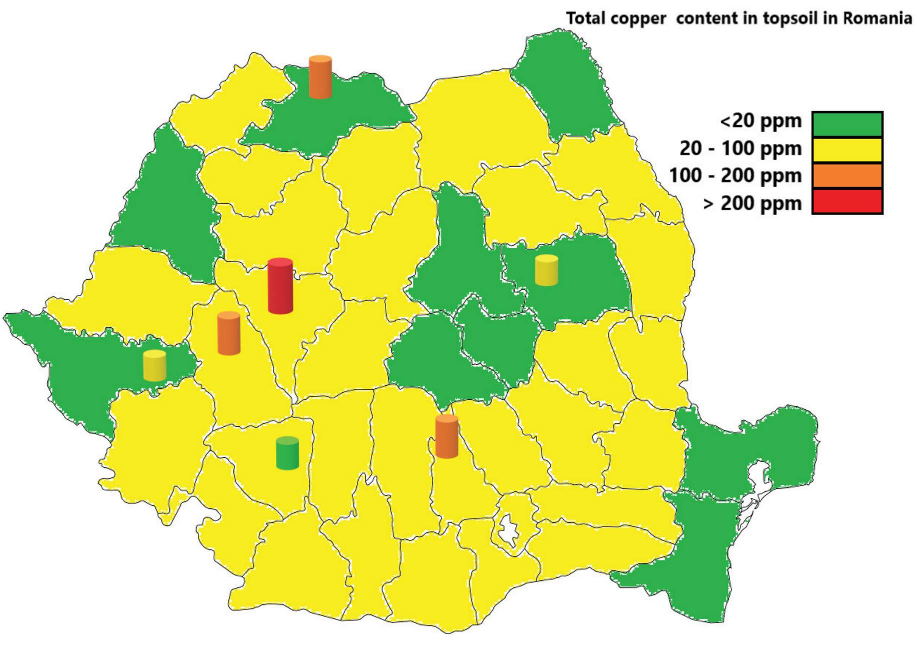

Figure 2. Copper content in topsoil of Romania counties

Lead

Lead is well known to be toxic and his harmful effects were intense studied (Roba et al., 2015). Unlike other heavy metals like zinc or cooper, lead has no role for plant or animal development. His presence in soils has only negative effects. Still, in a lot of areas the high concentration of lead causes serious problems, especially in zones with big industry centers. Some of these areas are mentioned in the table below (Table 4), according to the data presented in a set of studies (Mihali et al., 2013; Damian et al., 2013; Curcă, 2011; Dumitrel et al., 2013; Elekes et al., 2014; Bird et al., 2005; Buruiana et al., 2016; Sirbu et al., 2012; Dinu et al., 2018; Suciu et al., 2008; Zgripcea, 2013; Popescu et al., 2013; Morar et al., 2010; Buruiana et al., 2016; Dumitrel et al., 2013; Ene et al., 2010; Marin et al., 2010; Munteanu et al., 2012; Sur et al., 2012; Elekes, 2014; Nimirciag, 2012; Albulescu et al., 2012; Popa et al., 2016; Big et al., 2012; Gămăneci et al., 2011; Morar et al., 2010; Stefu et al., 2013; Ungureanu et al., 2017; Chirilă, 2013).

Table 4. Lead concentration in different areas (ppm)

County	ICPA study					Other studies					
	Mean	Number of sites	Median	Min	Max	Mean	Number of sites	Median	Min	Max	Year
Alba	32.11	19	30	13	84	207.24	18	0	0	1537	2013
Bacau	38.58	26	39	20	58	21.370	89	-	2.94	56.73	2013
B Nasaud	50.80	20	47	19	145	242.79	35	78.7	3.5	3687.2	2012
Braila	32.13	23	30	19	56	19.27	3	18.15	16.47	23.18	2009
Caras-Severin	43.19	32	40	7	98	31.00	9	25.59	21.63	47.08	2012
Cluj	30.41	27	30	15	51	75.36	18	0	0	735	2008
Dambovita	25.79	19	25	6	40	76.4	5	43.3	0.6	294.3	2014
Galati	19.81	16	20	10	30	20.17	8	18.54	6.50	35.72	2009
Gorj	39.50	22	35.5	28	67	10.89	12	1	1	64	2015
Harghita	43.31	26	35	20	84	102.08	12	67.5	30	260	2013
Hunedoara	41.86	29	30	15	99	373.5	6	270	120	888	2018
Iasi	32.65	23	28	14	69	27.73	1030	20.04	4.5	1995.43	2008
Maramures	32.77	26	30	10	81	365.27	10	261.88	163.28	804.09	2012
Mures	29.41	29	25	10	64	13.87	9	14.4	7.05	19.1	2010
Prahova	31.55	20	31.5	2	102	11.79	9	3.5	0	68.9	2006
Sibiu	43.05	19	35	20	170	680.9	56	-	19.61	2863	2011
Suceava	34.14	35	30	10	95	24.9	63	-	14.75	102.4	2013
Timis	21.51	35	20	11	40	6.58	18	0	0	26.09	2013
Valcea	33.45	22	33	15	45	16.90	4	14.84	12.43	25.5	2013
Vaslui	32.83	23	28	14	69	25.27	193	24	16	84	2017

The two sets of average values also differ significantly (T test value 0.049). The map shown in Figure 3 express some of the differences.

A total of 1115 points was used to obtain the map. Only 179 (16.05%) points had a smaller level than the reference value. The majority of sites, 759 (68.07%) had a value situated between reference and alert threshold. Surprisingly, a high number of sites, 131 (11.74%) have a high level of lead contamination, between alert and intervention threshold. Still, an impressive amount of 46 (4.12%) sites comes with a very high level of contamination, over intervention threshold. Considering that some studies were focused on these areas, the results are expected. The highest value was found around Rodna mining perimeter (3687.2 ppm).

Cadmium

Cadmium is found among the most toxic elements. Like lead, cadmium is not an essential microelement and his presence in the environment in concentrations that exceed the normal values may seriously affect living organisms (Oprea et al., 2011). The normal content of Cd in soil is 1 ppm as defined by the Romanian regulations.

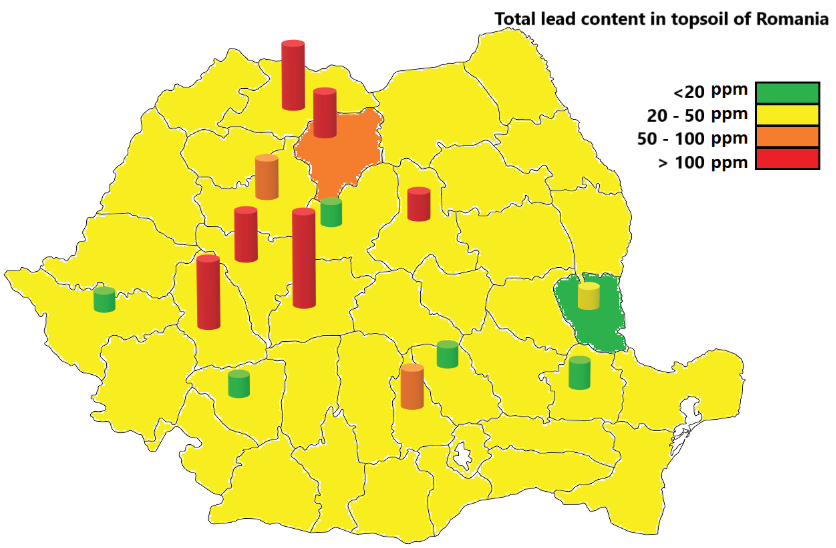

Figure 3. Lead content in topsoil of Romania counties

The cadmium pollution is described in the Table 5 (Buruiana et al., 2016; Ene et al., 2010; Popa et al., 2016; Albulescu et al., 2012; Damian et al., 2008; Stefu et al., 2013; Dumitrel et al., 2013; Oprea et al., 2011; Muntean et al., 2008; Fâciu et al., 2012; Iordache et al., 2015; Roşu et al., 2011; Chira et al., 2014; Sur et al., 2012; Nimirciag, 2012; Ungureanu et al., 2017; Dinu et al., 2018; Buzatu et al., 2018; Mandoc et al., 2013; Trîmbiţasu et al., 2006).

Table 5. Cadmium concentration in different areas (ppm)

County	ICPA study					Other studies					
	Mean	Number of sites	Median	Min	Max	Mean	Number of sites	Median	Min	Max	Year
Alba	1	26.00	1.00	0.1	2	0.53	4	0.42	0.35	0.95	2008
Bacau	0.9	27.00	0.83	0.5	1.1	0.595	89		0	1.45	2013
Braila	0.5	21.00	0.82	0.5	1.8	0.82	17	0.9	0.3	1.2	2009
Caras-Severin	1.5	34.00	1.54	0.9	3	2.06	9	1.85	1.28	3.22	2012
Dolj	1	29.00	0.82	0.4	1.6	0.6	2	0.6	0.05	1.15	2018
Galati	1	16.00	1.00	1	1	0.31	23	0.4	0	0.81	2009
Gorj	1	21.00	1.06	0.5	2.7	0.47	16	0.4	0	1.4	2015
Hunedoara	1	29.00	1.09	0.2	1.9	3.93	6	2.7	1.01	11.4	2018
Iasi	0.7	22.00	0.71	0.4	1.4	0.49	1030	0.36	0	15.44	2008
Maramures	1	25.00	1.19	0.5	2	3.8	10	2.4	1.52	12.6	2012
Prahova	2	21.00	1.88	1.1	2.5	0.59	9	0.5	0.2	1.05	2006
Sibiu	1.25	20.00	1.39	0.5	5	9.54	95	-	0.774	52.0	2011
Suceava	1	35.00	1.07	0.5	2	0.76	63	-	0.2	1.34	2013
Timis	1	35.00	1.11	0.4	1.5	0	18	0	0	0	2013
Valcea	1	23.00	1.12	0.5	1.8	1.96	5	2.1	1	2.1	2013
Vaslui	0.8	23.00	0.73	0.4	1	0.32	193	0.31	0.02	0.8	2017

In the cadmium analysis, the average values from ICPA study and the other studies are significantly different (T test = 0.307), as before. Form a total of 1035 point analysed, 708 (68.40%) are smaller than the reference value, 317 (30.62%) are situated between reference and alert threshold, just 6 points (0.57%) exceeds alert threshold to intervention threshold, and only 4 (0.38%) exceeds intervention threshold. The maximum value for cadmium contamination was 52.01 ppm, found in Sibiu County, near Copşa Mica, one of the world most polluted city.

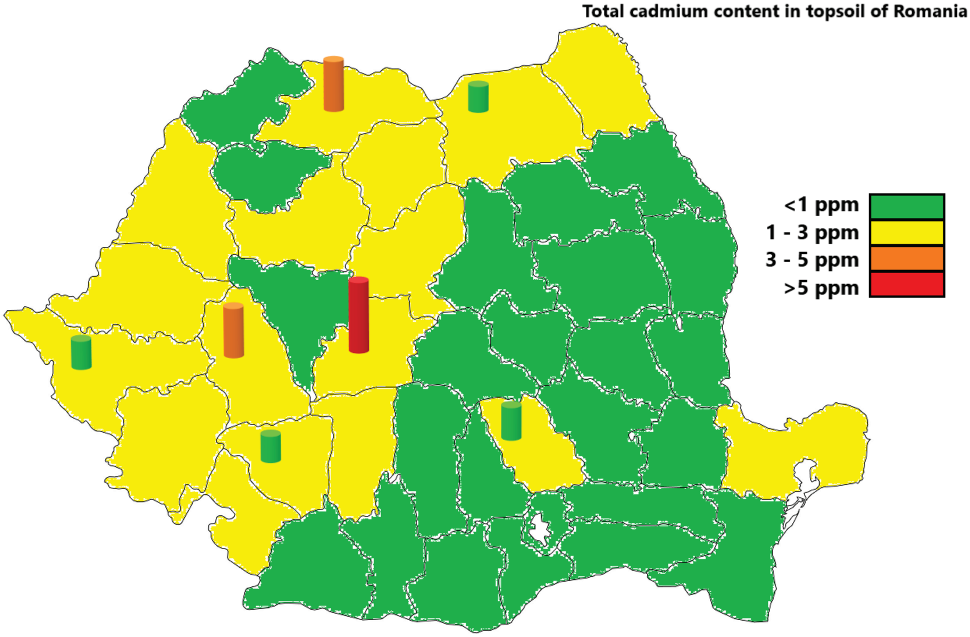

Figure 4. Cadmium content in topsoil of Romania counties

Giving an overview to the analysed heavy metals, the situation can be expressed in the chart below (Figure 5).

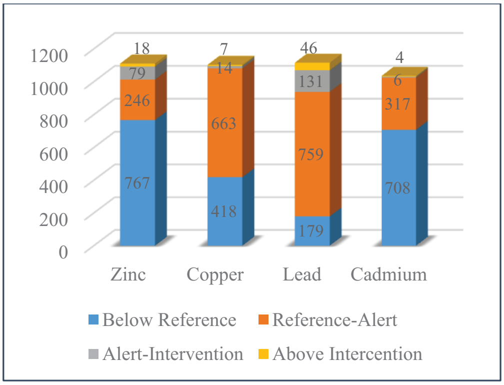

Figure 5. Distribution of examined sites by the pollution level

CONCLUSIONS

There are a lot of areas in Romania that are poorly investigated, investigated a long time ago or which have not been studied at all.

Even that the most part of Romania had safe lands for agriculture, there are some areas with very high pollution which exceeds Romanian regulations, according to the reviewed papers.

Copper and cadmium exceed intervention threshold in less than 1% of the examined points.

Lead is "leader", regarding the points who exceeds reference value, with 83.95% of sites in this category. At the opposite pole, zinc has 26.47% of sites in this category.

REFERENCES

Albulescu, M., Popovici, H., Turuga, L., Masu, S., Chiriac, A. (2012). Analysis of Heavy Metals Content of Soil and *Vitis vinifera* in Two Vineyard Areas of the Caras-Severin County, Romania. *Journal of Environmental Protection and Ecology, 13*(1), 48–55.

Ballabio, C., Panagos, P., Lugato, E., Huang, J. H., Orgiazzi, A., Jones, A., Fernández-Ugalde, O., Borrelli, P., Montanarella, L. (2018). Copper distribution in European topsoils: An assessment based on LUCAS soil survey. *Science of the Total Environment*, *636*, 282–298.

Berbecea, A., Radulov, I., Sala, F., Crista, F., Lato, A. (2011). Interrelation between metal availability, soil pH and mineral fertilization. *Research Journal of Agricultural Science, 43*(3), 19–22

Big, C.L., Lăcătuşu, R., Damian, F. (2012). Heavy metals in soil-plant system around Baia Mare city, Romania. *Carpathian Journal of Earth and Environmental Sciences, 7*(3), 219–230.

Boros, M.N., Smical, I., Micle, V., Lichtscheidl-Schultz, I. (2015). Heavy metal pollution of soils from Baia Mare - case study: Cuprom industrial area. *Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering, IV*, 99–106.

Buruiana, D.L., Balta, S., Iticescu, C., Georgescu, L.P., Humeniuc, I.I. (2016). Determining the concentration of heavy metals in the soils near slag landfills. *Revista Romana de Materiale, 46*(1), 108–114.

Buzatu, G.D., Dodocioiu, A.M. (2018). Study of heavy metals in the soils of two localities in Dolj County. *Agriculture for Life, Life for Agriculture, 1*(1), 352–357.

Chira, I., Damian, G., Chira, R. (2014). Spatial distribution of heavy metals in the soils of Băiuţ area, Maramureş County, Romania. *Carpathian Journal of Earth and Environmental Sciences, 9*(1), 269–278.

Chirilă, M.E. (2013). Studiul distribuţiei metalelor grele în sol în perimetrul municipiului Bacău folosind sisteme informaţionale geografice. Universitatea „Vasile Alecsandri” din Bacău. http://www.ub.ro/files/scoala-doctorala/Rezumat_Teza_Faciu_Ema.pdf.

Cojoc, E. (2011). *Modelarea bioacumulării metalelor grele în plante de cultură din zona Copșa Mică*. Bucuresti, Universitatea din București. Retrieved February 20, 2019, from http://www.cesec.ro/pdf/CojocEmilia2011.pdf.

Curcă, G. (2011). Studiul geochimic al poluanților solizi din atmosfera zonelor industriale ale orașului Iași, cu privire special asupra repartiției metalelor grele. Universitatea din București. Retrieved February 20, 2019, from http://gg.unibuc.ro/wp-content/uploads/2018/05/CURCA-Geanina.pdf.

Cyraniak, E., Draszawka-Bołzan, B. (2014). Heavy metals in circulation biogeochemical. *World Scientific News*, *6*, 30–36.

Damian, F., Damian, G., Lăcătușu, R., Iepure, G. (2008). Heavy metals concentration of the soils around Zlatna and Copșa Mică smelters Romania. Carpth. *Journal of Earth and Environmental Sciences, 3*(2), 65–82.

Damian, F., Damian, G., Lăcătușu, R., Postolache, C., Iepure, G., Jelea, M., Năsui, D. (2013). The heavy metals immobilization in polluted soils from Romania by the natural zeolites use. *Carpathian Journal of Earth and Environmental Sciences, 8*(4), 231–250.

Dimitriu, D. (2014). Restoration of heavy metals polluted soils case study - camelina. *AgroLife Scientific Journal, 3*(2), 29–38.

Dinu, C., Ungureanu, E.M., Vasile, G.G., Kim, L., Ionescu, I., Ene, C., Simion, M. (2018). Soil and Vegetation Pollution from an Abandoned Mining Area Situated in Hunedoara County, Romania. *Rev. Chim., 69*(1), 14–20.

Dumitrel, G.A., Popa, M., Glevitzky, M., Vica, M., Todoran, A. (2013). Evaluation of Soil Heavy Metal Pollution in the Zlatna Region. *Journal of Environmental Protection and Ecology, 14*(4), 1569–1576.

Dumitru, M., Manea, A., Ciobanu, C., Dumitru, S., Vrînceanu, N., Calciu, I., Tănase, V., Preda V., Rîşnoveanu, I., Mocanu, V., Eftene, M. (2011). *Monitoringul stării de calitate a solurilor din România, Institutul Naţional De Cercetare-Dezvoltare Pentru Pedologie Agrochimie Şi Protecţia Mediului, ICPA Bucureşti.* Craoiva, RO: Sitech.

Elekes, D.D. (2014). Assessment of Historical Heavy Metal Pollution of Land in the Proximity of Industrial Area of Targoviste, Romania. Environmental Risk Assessment of Soil Contamination, Intech Open. Retrieved February 20, 2019, from https://www.intechopen.com/books/environmental-risk-assessment-of-soil-contamination/assessment-of-historical-heavy-metal-pollution-of-land-in-the-proximity-of-industrial-area-of-targov.

Ene, A., Stihi, C., Popescu, I.V., Bosneaga, A., Radulescu, C., Gheboianu, A. (2008). XRF-AAS analysis of heavy metals in soils around of a ferrous metallurgical plant in eastern part of Romania. ISINN-18 proceedings, Retrieved February 20 from http://isinn.jinr.ru/proceedings/isinn-18/pdf/Ene.pdf

Fâciu, M.E., Lazăr, G., Nedeff, V. (2012). The assessment of heavy metals concentration in Bacau city soil. Necessity and working methods. *Journal of Engineering Studies and Research, 18*(1), 80–95.

Gămăneci, G., Căpăţînă, C. (2011). Study on soil pollution with heavy metals in Rovinari area. *Analele Universităţii „Constantin Brâncuşi” din Târgu Jiu, 3*. 309–318.

Hassan, Z., Ali, S., Rizwan, M., Hussain, A., Akbar, Z., Rasool, N., Abbas, F. (2017). Chapter 14. Role of Zinc in Alleviating Heavy Metal Stress. Essential Plant Nutrients, Springer International Publishing.

Hura, C., Munteanu, N., Stoleru, V. (2013). Heavy Metals Levels in Soil and Vegetables in Different Growing Systems. E3S Web of Conferences 1. 08007.

Iancu, O.G., Buzgar, N. (2008). *Atlasul geochimic al metalelor grele din solurile municipiului Iaşi şi împrejurimi/The geochemical atlas of heavy metals in the soils of the municipality of Iaşi and its surrounding areas.* Iasi, RO: Ed. Universităţii „Alexandru Ioan Cuza”.

Iordache, M., Popescu, L.R., Pascu, L.F., Iordache, I. (2015). Environmental Risk Assessment in Sediments from Jiu River, Romania. *Rev. Chim., 66*(8), 1247–1252.

Laţo, A., Radulov, I., Berbecea, A., Laţo, K., Crista, F. (2012). The transfer factor of metals in soil-plant system. *Research Journal of Agricultural Science, 44* (3), 67–72.

Lăcătușu, R., Lăcătușu, A.R. (2009). Heavy Metals Soil Pollution in Some Urban Location from Romania. *Alliance for Global Sustainability Bookseries, 17*, 347–355.

Mandoc, L.R. (2013). Metode şi metodologii în determinări analitice cu aplicaţii în evaluări de mediu şi sănătate. Universitatea Politehnica din Bucureşti

Retrieved February 20, 2019, from http://dspace.incdecoind.ro/bitstream/123456789/443/1/Rezumat_teza_luiza%20Roxana%20MANDOC.pdf.
Marin, C., Tudorache, A., Moldovan, O.T., Povară, I., Rajka, G. (2010). Assessing the contents of arsenic and of some heavy metals in surface flows and in the hyporheic zone of the Arieş stream catchment area, Romania. *Carpathian Journal of Earth and Environmental Sciences, 5*(1), 13–24.
Masindi, V., Muedi, K. (2018). Chapter 7. Environmental Contamination by Heavy Metals. Heavy Metals. Intechopen.
Mihali, C., Oprea, G., Michnea A., Jelea S.G., Jelea, M., Man, C., Senila, M., Grigor, L. (2013). Assessment of heavy metals content and pollution level in soil and plants in Baia Mare area, NW Romania. *Carpath J. Earth Env., 8*(2), 143–152.
Moldoveanu, A.M. (2014). Chapter 9. Assessment of Soil Pollution with Heavy Metals in Romania. Assessment of Soil Pollution with Heavy Metals in Romania, 285–305.
Morar, F., Hasegan, I. (2010). The metal content of soils in the Mures County and phytoremediation prospects use as a remediation method. *Scientific Bulletin of the „Petru Maior" University of Tîrgu Mureş, 7*(2), 53–58.
Muntean, E., Muntean, N., Mihăiescu, T. (2010). Cadmium and lead soil pollution in Copsa Mica area in relation with the food chain. *Research Journal of Agricultural Science, 42*(3), 731–734.
Nimirciag, R. (2012). Heavy metals in the soils of Rodna mining area, Romania and zeolite efficiency for remediation. *Environmental Engineering and Management Journal, 11*(2), 421–426.
Oprea, G., Mihali, C., Michnea, A., Senila, M., Gogoasa, I., Vosgan, Z. (2011). Assessment of Lead and Cadmium Content in the Soils and Plants in Industrial Area. *American Journal of Environmental Sciences, 7* (5), 402–408.
Popa, R.G., Pecingina, I.R. (2016). Study on chemical analysis of soil in the county of Gorj, in terms of the content of heavy metals and chlorides. *Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering, V*. 105–108.
Popescu, E., Nimirciag R., Vina, G., Ajmone-Marsan, F. (2013). Spatial Distribution of Potentially Toxic Elements in Soils Polluted by Mining Activities. *Rev. Chim., 64*(5), 477–481.
Roba, C., Roşu, C. (2015). Ingestion of lead due to the consumption of drinking water from private wells in four rural areas from Baia Mare - Romania metropolitan area. *Scientific Papers Series Management, Economic Engineering in Agriculture and Rural Development, 15*(3), 265–268.
Roşu, C., Naghiu, R., Onofrei, C. (2011). Impact of urban landfill on soil quality. Case study towns: Aiud and Gura-Humorului. *AES Bioflux, 3*(2), 69–82.
Stefu, N., Neculae, A., Turcu, L., Balint, A. (2013). Analysis of metal content in soil in Timis County. *Analele Universităţii de Vest din Timişoara, LVII*, 44–51.
Suciu, I., Cosma, C., Todică, M., Bolboacă, S. D., Jäntschi, L. (2008). Analysis of Soil Heavy Metal Pollution and Pattern in Central Transylvania. *International Journal of Molecular Sciences, 9*(4), 434–453.
Sur, I.M., Micle, V., Plopeanu, G. (2012). Cercetări asupra stării de calitate a solurilor din zona Romplumb Baia Mare. *Ecoterra-Journal of Environmental Research and Protection, 31*, 105–109.
Trîmbiţasu, E., Neagoe, S., Mateescu, C. (2006). Investigation of soil pollution with Pb and Cd. *Analele Universităţii din Bucuresti – Chimie, XV*(II), 15–19.
Ungureanu, T., Iancu, G.O., Pintilei, M., Chicoș M.M. (2017). Spatial distribution and geochemistry of heavy metals in soils: A case study from the NE area of Vaslui County, Romania. *Journal of Geochemical Exploration, 176*, 20–32.
Zgripcea, M. (2014). Evaluarea poluării cu substanțe prioritar periculoase a ecosistemelor apă și sediment în zona unei platform industrial complexe și metode de distrugere a poluanților. Universitatea Politehnica din Bucureşti. Retrieved February 20, 2019, from http://dspace.incdecoind.ro/bitstream/123456789/452/1/Rezumat_teza_Mihaela%20IORDACHE.pdf.
***ICPA Bucuresti (2000). Heavy metal distribution in soils in Romania. Monitoringul stării de calitate a solurilor din România.
***Ordin nr. 756 din 3 noiembrie 1997 pentru aprobarea Reglementarii privind evaluarea poluarii mediului.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

A CALCARIC ALLUVIAL SOIL CHARACTERISTICS AT SUBMICROSCOPIC SCALE USING SEM

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Abstract

Soil characteristics play an important role on agricultural crop production, especially in the organic farming system. Increasing the productive potential of organic agroecosystems in the context of climate change is nowadays possible also using soil treatment with microbial bioinoculants. Microbial bioinoculants have the capacity to enhance nutrient availability, nutrients uptake, support the soil and plant health, by controlling the pest and diseases or enhance the plant defence responses. In 2018, at the Vegetable Research and Development Station Buzau, a soil micromorphology study was performed on an Aluviosol Molic Calcaric, in an organic testing field cultivated with vegetables. Soil samples taken at six different depths were analysed using SEM images. The soil micro and macromorphological characteristics were correlated with the analytical data. The results showed the $CaCO_3$ abundance in soil, while at sub-microscopic level it appears as acicular crystals into the efflorescences coatings. The soil skeleton is well represented, the sand-sized mineral grains being abundant. Many skeleton grains (as feldspars and mica flakes) had altered surfaces, being partially covered with secondary decomposed products, whose presence favours the installation of microorganisms.

***Key words**: soil micromorphology, SEM image analysis, Buzau, Romania, organic soil.*

INTRODUCTION

At the level of 2017 the total organic area (areas under conversion plus the certified areas) in the EU-28 was 12.6 million hectares (ha) and the trend is still ascendant. The increase in organic area between 2012 and 2017 was with 25%, Bulgaria and Croatia reflecting a growth of over 100%. However, four EU Member States reported reductions in the organic area: Romania (-10.3%), Greece (-11.3%), the United Kingdom (-15.6%) and Poland (-24.5%) (EUROSTAT, 2019). From 2012 to 2017, the share of total organic area in the total used agricultural area (UAA) within the EU rose from 5.6% to 7% (EUROSTAT, 2019). Romania is on the 26 place, of the 28 EU countries, but our organic land under conversion represented in 2017, 42.3%, one of the biggest in Europe. To be able to sustain Romanian farmers in their attempt to produce at high quality standards into organic, in the recent climate change constraints, high support is required from the scientists.

Soil is the most important resource of organic farmers. The attempt of performing in organic crops on calcaric alluvial soils, which are almost like sandy soils, requires high knowledge of crop technologies (Stefan et al., 2018), as usually these soils have less nutrients and low water holding capacity leading to frequent irrigation and fertilizations to meet crop requirements (Alshankiti & Gill, 2016). In studies on soil microbial activity of sandy soils it was found that overall activity was low, due to low organic matter and low rainfall (Unkovich, 2014). Due to lower organic fertility and fewer microsites to protect soil biota, sandy soils have also a lower capacity to suppress pathogens and other pests (Coventry, 1998). Eftene et al. (2014) showed that physical and chemical properties of a soil with sandy texture and low soil organic matter content (due to intense mineralization of organic residues) create conditions for a reduced activity of microflora. Carminati et al. (2008) exploring the soil-water interfaces between isolated heterogeneous aggregates within a soil (controlled by matric tensions) reveal the intimate associations between pore geometry and water.

The physical characteristics of the soils, mainly of the young soils (as Aluviosols) are important for the further evolution of the soils, for the structural and the adjacent poral space genesis that strongly influence the air and soil solution circulation and the biochemical processes. The role and transformations of agricultural inputs, as fertilisers, microbial inoculants, pesticides, etc. are highly dependent on soil characteristics. For a deeper understanding of these characteristics and its interrelations with biotic factors, natural or applied during crop technologies, analysis using Scanning Electron Microscopy (SEM) proved their utility over time. Early SEM observations have been conducted by Barden in 1973 and Grabowska-Olszewska in 1975 on European loess from Belgium and Poland (Delage et al., 2005) and become currently widely used for micromorphological studies. Răducu (2018) showed that soil biota initiated and controlled the important pedogenetic processes, thus soil fauna activity had a major influence on the illuvial process of pedobioplasma, while the activity of microorganisms had a major influence on coatings evolution.

In the soil environment, the microbial phase development (on the mineral and organic constituents), usually represent the constituents transformation and weathering/decomposition (according to their mineral or organic status). Mica destruction was detected by SEM, due to the growth and metabolic activities of bacteria (*Bacillus cereus)*, which cause the extraction of iron atoms from the octahedral position in mica from the kaolin sample (Štyriakov et al., 2003). Also, Balogh-Brunstad et al. (2008) found, by the aim of SEM images, a biotite surface covered with biofilms containing bacteria and fungal hypha. Often the concept of soil macro-aggregates (>200 mm) and micro-aggregates (50-200 mm) is invoked in an attempt to understand the importance of physical protection of organic matter isolated from microbial and chemical degradation (Young et al., 2008).

Many Alluvial soils have calcium carbonate accumulations, either inherited from the parent material or accumulate under water table influence.

Based on the results obtained following the investigation of a pedochronosequence, Kuznetsova et Khokhlova (2012) conclude that the sub-micromorphology (by the aim of SEM) of the carbonate accumulations may be useful for reconstruction of recent climatic conditions and allow to estimate the general trend of soil evolution, and even the rate and dynamics of soil formation processes.

Heberling et al. (2016) showed that calcite based environmental remediation strategies rely on the fast recrystallization of calcite and the concurrent uptake and immobilization of pollutants. Young et al. (2008) showed that it is need to bring together a new discipline that combines the biology and physics of the soil ecosystem. Thus, the biophysical approach, combined, where required, with important mineral-microbe knowledge is needed to help us understand the mechanisms by which soils remain productive, and to identify the tipping-points at which there may be no return to sustainability. Manea et al. (2016) studying a Fluvisols with neutral - slightly alkaline reaction, concluded that the correlations between soil fungal number and soil chemical properties are generally positive and significant, especially for soil reaction, the degree of soil base saturation, the sum of cation exchange capacity, as well as the calcium contents.

It is proven that beneficial microorganisms have the ability to improve yield quality and soil fertility. For organic crops, the use of plant resistance enhancers started to become very popular and appreciated by the market.

In a climate change perspective, a different distribution of microbial organisms is expected and that the progressive increase of air temperature and reduced cold shocks on microbial cells may lead to spring water pollution also during winter, differently from the actual observations Naclerio et al. (2012). Therefore, a throughout research of soil - microorganisms - plants interactions is needed for each soil type.

The aim of the present study was to emphasize the main characteristics of an Aluviosol Molic Calcaric from an organic agroecosystem, before the treatment with microbial bioinoculants, using scanning electron microscope (SEM), in order to better understand the changes that occur in the soil (open field) after application of such microbial inoculants.

MATERIALS AND METHODS

The study was carried out in the Vegetable Research and Development Station Buzău (V.R.D.S. Buzău), Romania (lat.: 45.16108714 N and long: 26.82423914 E, alt: 92 m) in 2018, in the organic farming research plot, having an area of 1.8 ha. The studied area it is situated inside the Buzău city, in Buzău county, at around 1 km distance of the river. V.R.D.S. Buzau is well known for tomato production and breeding, being obtained here, over time, valuable varieties appreciated by both consumers and growers (Zamfir et al., 2018).

The soil is Aluviosol Molic Calcaric (according to SRTS-2012 and Mollic Fluvisol Calcaric - according to WRB-SR-2006) formed in calcaric fluvial deposits.

For the detailed study of the soil characteristics, it was used the scanning electron microscopy (SEM) technique, to obtain details of soil samples at very small scale. The SEM analysis of the Aluviosol Molic Calcaric was carried out with FEI Inspect S50 model, in the Laboratory of Microscopy and Plant Anatomy, of the Research Center for Study of Food and Agricultural Products Quality, inside the University of Agronomic Sciences and Veterinary Medicine of Bucharest.

The terminology used for description at microscopic level was according to Bullock et al. (1985).

RESULTS AND DISCUSSIONS

Physical and chemical characteristics

On the general background of a soil with specific characteristics for an Aluviosol Molic Calcaric (Table 1), the granulometric data showed a medium sandy-loam texture into the top Am horizon, and a fine to coarse sandy-loam texture into the deeper horizons.

The colloidal clay (<0.002 mm) is very low, ranging between 18.5% in the surface Am horizon and 10.3% in the deeper analysed horizon (Ck_2).

Table 1. Morphological characteristics of the Aluviosol Molic Calcaric

Am (0-37 cm)	**Ck_1 (37-96 cm)**	**Ck_2 (96-124 cm)**
Medium sandy-loam; crumb structure weak developed; moderate biological activity; frequent fine pores; weak-moderate effervescence (locally); very frequent thin roots from cultivated vegetation.	Fine sandy-loam; massive; frequent thick pores; frequent fine roots; moderate-strong effervescence.	Coarse sandy-loam; massive; very friable; coarse mineral grains (visible by the necked eye); frequent $CaCO_3$ pseudomycelium; strong effervescence.

The physical characteristics data shows that total porosity is very high (52-56% v/v), while the bulk density is very low (1.13-1.22 g/cm^3).

The chemical characteristics data showed that studied soil is moderate alkaline, the pH values ranging between 8.06 in the surface horizon and 8.3 in the deeper analyzed horizon.

The base saturation degree is very high (96-100%) as a result of the calcium carbonate presence into the soil profile.

The organic matter content is medium. However, being highly humified and predominating the calcium humates, the colour of the surface horizon is very dark (under 10YR 5/3 on dry), which classified the studied soil to the mollic subtype.

The nitrogen index (calculated according to the humus content and the saturation degree at hydrolytic acidity) is medium (3.45-2.00) in the most part of the soil profile, except the deeper horizon, where the value decrease drastically (to 1.4).

The potassium content decreased with the increasing depth, ranging between medium (123-162 ppm) in the upper horizons and low (13 ppm) in deeper ones.

Very significant exponential relationship was established, by Manea et al. (2011) researches, between mobile phosphorus and soil reaction.

In the studied Aluviosol Molic Calcaric, the mobile phosphorus decreased with the soil profile depth, from the higher value (46 ppm) in the surface to the lowest value (13 ppm) in the deeper analyzed horizon.

Soil characteristics at submicroscopic scale

The microscopic observations by the aim of SEM were performed on the freshly broken surfaces of the soil sampled from each pedological horizon.

In the top soil, developed an A mollic horizon (Am 0-37 cm) having crumb structure (Figure 1). The spatial organization of the structural elements generated a relatively high porosity composed predominantly by the interconnected pores.

The spatial organization of the soil structural elements is also emphasized by the analytical data of total porosity.

Detailing to a higher scale of observation (Figure 2) it became obvious that although the clay content is low (18.5%), the soil constituents are relatively well structured, the skeleton grains and the colloidal constituents are binding together relatively tight.

The content of the organic matter (an important binding agent of the soil) is ranging from 3.6% to 1.4% (decreasing from the surface to the deeper horizons). At sub-microscopic scale, the organic matter appears only as humified substances, embedded into the soil matrix together with the clay particles.

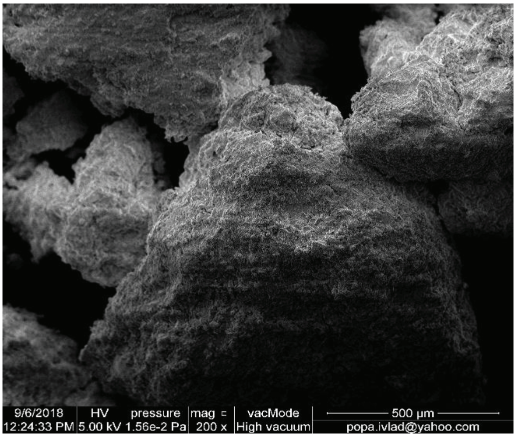

Figure 1. SEM image of Am (0-37 cm) horizon: crumby structure with interconnected pores (200 X)

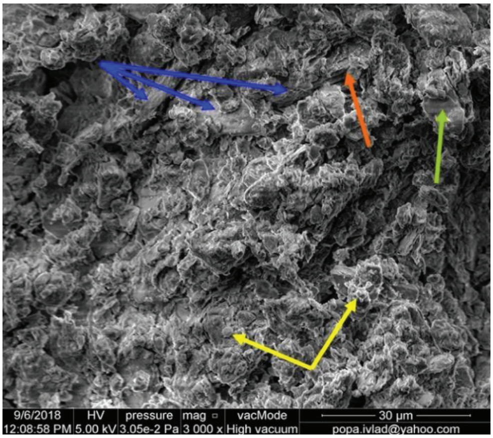

Figure 2. SEM image of Am (0-37 cm) horizon: mineral skeleton grains (→); microorganisms (→); plagioclase feldspars (→); mica flakes (→) (3000 X)

The soil skeleton is well represented mainly by the fine sand-size and silt-size mineral grains. Quartz dominates the coarse fraction of the horizon.

Many fine sand-size plagioclase feldspars were observed among the skeleton grains (with polysynthetic twinning - Figure 2) together with the silt-size mica flakes (emphasised by their foliated texture - Figure 2).

These mineral grains being poorly weathered, their surfaces are partially covered with secondary alteration products (secondary clays and/or Fe oxy-hydroxides etc.).

The presence of the weathered products, favour the microorganism development on the mineral grains surfaces (Figure 4).

Another important aspect, related to the structuring process and consequently to the building of a favourable environment for plant development, is the presence of microorganisms (Figure 2) which, by generating "organic cement" (their metabolic products dominated by the presence of mucilaginous polysaccharides) increase the particle cementation and consequently the soil structuring process.

The high interconnected porosity (Figure 3) reveals excessive drainage, and therefore low water retention capacity of the soil, which negatively affects the plant water supply.

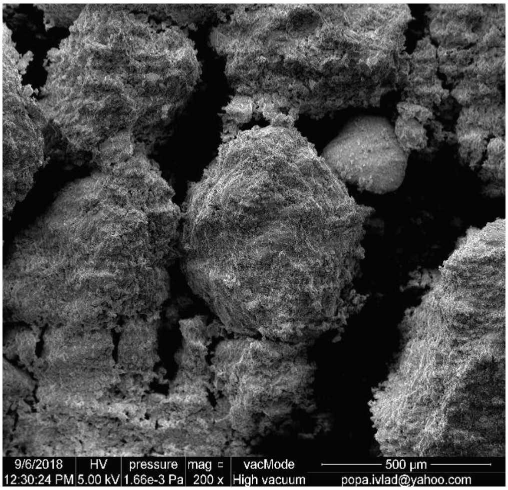

Figure 3. SEM image of Am (20-37 cm) horizon: crumby structure with interconnected pores (200 X)

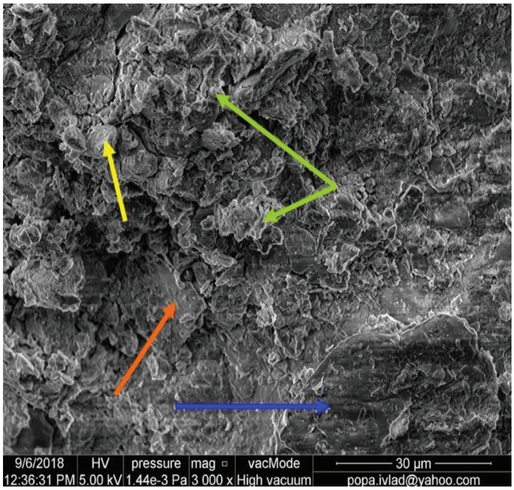

Figure 4. SEM image of Am (20-37 cm) horizon: mineral skeleton grains (→); calcite (→); clay particles binding together (→); mica flakes (→) (3000 X)

The SEM emphasised the organization of the soil constituents at micro-scale, their aggregation into the peds with the formation of the adjacent micro-pores, represent the bricks of the next levels of organization (until the macro-scale) and induce the physical and chemical characteristics of the soil, and further the health of the organic agroecosystem environment.

The **Ck_1 (60-96 cm)** horizon, being dominated by the characteristics inherited from the parent material, is low structured, with subangular blocky structure.

The friability of the horizon (Figures 5 and 6) is the result of the granulometry dominated by the fine sand-size and loam-size particles (Table 1 and Figure 6).

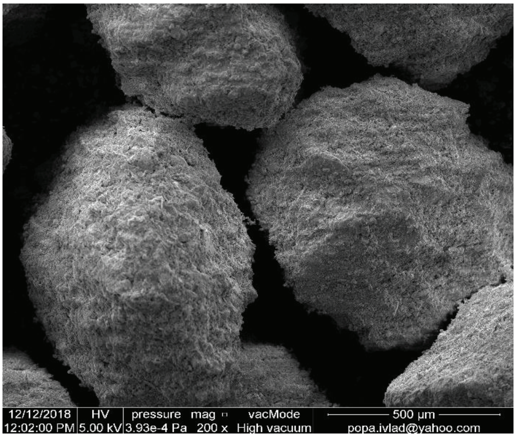

Figure 5. SEM image of Ck_1 (60-96 cm) horizon: crumby structure with interconnected pores (200 X)

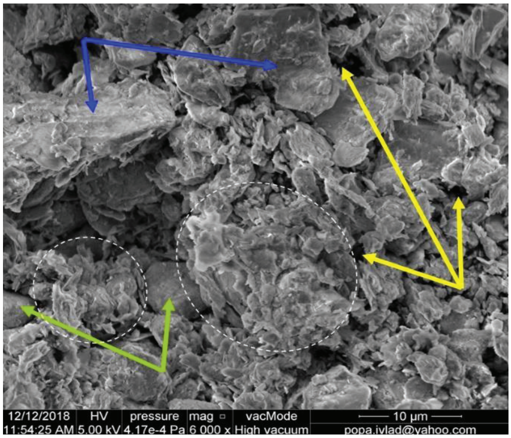

Figure 6. SEM image of Ck_1 (60-96 cm) horizon: mineral skeleton grains (→); silt-size micro-aggregates (◌); intrapedal packed pores (→); calcit crystals (→) (6000 X)

The clay particles formed micro-aggregates of silt-size by binding together face-face and/or edge-edge, and also embedding calcite crystals (Figure 6). The small intrapedal packed pores also formed by the spatial organization of the micro-aggregates.

The **Ck_2 (96-124 cm)** horizon is very friable (Figures 7 and 8), showed also by the granulometric data that highlighted a coarse sandy texture.

The main part of the horizon material is un-aggregated. The aggregates are small and poorly formed (Figure 7).

The high structural friability that characterizes this horizon is also given by the abundance of calcium carbonate which, on the one hand, impregnates the horizon matrix, and on the other hand it appears as efflorescences (coatings) on the walls of the poral space.

The presence of calcium carbonate was also reported by the macroscopic description in the field, where frequent $CaCO_3$ pseudomycelium has been observed.

At the sub-microscopic level (Figure 8) it can be noted that these efflorescences coatings are made up of acicular crystals (needle from calcite), also called *lublinite*. This type of acicular calcite grows up from the saturated solutions, when the soil moisture rapidly changes.

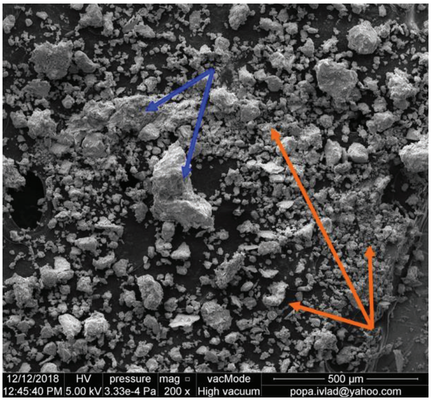

Figure 7. SEM image of Ck_2 (96-124 cm) horizon: poorly developed and non-durable structural elements (→) by the side of un-aggregated coarse material (→) (200 X)

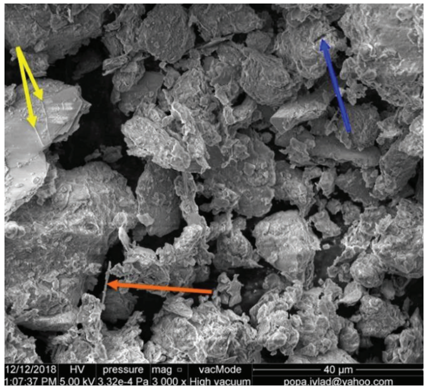

Figure 8. SEM image of Ck_2 (96-124 cm) horizon: acicular calcite crystal (→); fungi mycelium on a platy mica grain (→); fissured calcite and with local recrystallizations (→) (3000 X)

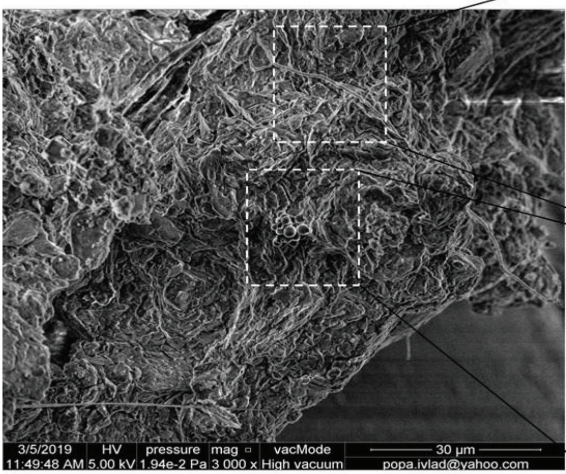

Figure 9. SEM image of Am (0-37 cm) horizon: a highly decomposed vegetal residue (3000 X)

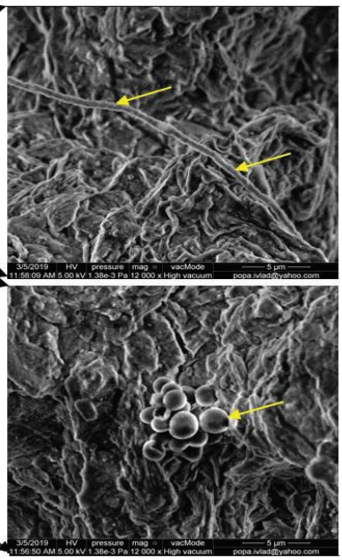

Figure 10. Details of figure 9: microorganisms (→) on the decomposed vegetal residue (12000 X)

Into the soil profile the microorganism activity was also detected, mainly on highly decomposed vegetal remains (Figures 9 and 10).

The presence of different species of microorganisms, showed the biological diversity of the soil and their high activity, which leads the decomposition processes, as well as the release of the nutrients into the soil and the humic substances accumulation.

The data of the physical and chemical characteristics, together with the detail observations of the soil constituent fabric at sub-microscopic scale reveal that Aluviosol Molic Calcaric environment is favorable to the development of the microbial bioinoculants.

CONCLUSIONS

The results obtained with the help of the scanning electron microscope (SEM) following the study of an Aluviosol Molic Calcaric from an organic agroecosystems before the treatment with microbial bioinoculants revealed detailed aspects related to the spatial organization of the soil at sub-microscale.

The high magnification (until 6000 X) allowed the visualization of the soil elementary fabric into the peds.

Even if the SEM images are in the greyscale, the skeleton grains were detected according their specific characteristics (type of twinning, foliated texture etc.).

Many skeleton grains are weathered and partially covered with secondary alteration products (clay, Fe etc.), whose presence favours the development of microorganisms on the faces of the mineral grains.

The study of the behaviour of $CaCO_3$ in the soil pedological horizons showed the presence of the different types of calcite crystals: crypto-crystals (embedded into the soil matrix), acicular crystals (on the pore walls) etc.

ACKNOWLEDGEMENTS

The article was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

Many thanks to res Dr. Daniela Răducu, from the National Research and Development Institute for Soil Science, Agro-Chemistry and Environment (ICPA-Bucharest), for her continuous and devoted support and patience.

REFERENCES

Alshankiti, A., Gill, S. (2016). Integrated Plant Nutrient Management for Sandy Soil Using Chemical Fertilizers, Compost, Biochar and Biofertilizers - Case Study in UAE. *Journal of Arid Land Studies, 26*(3), 101–106.

Balogh-Brunstad, Z., Keller, C.K., Gill, R.A., Bormann, B.T., Li, C.Y. (2008). The effect of bacteria and fungi on chemical weathering and chemical denudation fluxes in pine growth experiments. *Biogeochemistry, 88*, 153–167.

Bullock, P., Fedoroff, N., Jongerius, A., Stoops, G., Tursina, T., Babel, U. (1985). *Hanbook for soil thin section description*, Wine Research Public, 152.

Carminati, A., Kaestner, A., Lehmann, P., Fluhler, H. (2008). Unsaturated water flow across soil aggregate contacts. *Advances in Water Resources, 31*(9), 1221–1232.

Coventry, D.C., Holloway, R.E., Cummins, J.A. (1998) Farming fragile environments: low rainfall and difficult soils in South Australia. *Proc. 9th Australian Agronomy Conference.* Wagga Wagga, 12.

Delage, P., Cui, Y.J., Antoine, P. (2005). Geotechnical problems related with loess deposits in Northern France. *Proceedings of International Conference on Problematic Soils,* 25-27 May, Eastern Mediterranean Univ., Famagusta, N. Cyprus, 1–24.

Eftene, A., Matei, S., Matei, G.M., Raducu, D. (2014). Influence of land use on microbiological activity of sandy soils. *Proceedings of ICAMS 2014 - 5th International Conference on Advanced Materials and Systems*, 427–432.

Florea, N., Munteanu, I. (2012). *Sistemul român de taxonomia solurilor (SRTS-2012)*. Ed. Estafia, Bucureşti, 206.

Heberling, F., Pauling, L., Nie, Z., Schild, D., Finck, N. (2016). Morphology Controls on Calcite Recrystallization. *Environmental Science Technol, 50*(21), 11735–11741.

Kuznetsova, A.M., Khokhlova, O.S. (2012). Submicromorphology of pedogenic carbonate accumulations as a proxy of modern and paleoenvironmental conditions. *Boletin de la Sociedad Geologica Mexicana, 64*(2).

Manea, A., Dumitru, M., Matei, M., Lăcătuşu, A., Matei, S., Dumitru, S. (2016). Effects of acidification on the soil microbiological activity in the Zlatna area. *Journal of Environmental Protection and Ecology, 17*(2), 503–511.

Manea, A., Dumitru, M., Rizea, N., Tănase, V., Preda, M. (2011). Influence of soil acidification on some soil chemical properties (exchangeable aluminium and mobile phosphorus). *Scientific Papers, Series A., Agronomy, LIV*, 160–165.

Naclerio, G., Allocca, V., Bucci, A., Capobianco, G., Fiorillo, F., Celico, F. (2012). Winter Survival of

Microbial Contaminants in Soil. *AgroLife Scientific Journal, 1*, 165‒168.
Panagos, P., Van Liedekerke, M., Jones, A., Montanarella, L. (2012). European Soil Data Centre: Response to European policy support and public data requirements. *Land Use Policy, 29*(2), 329‒338.
Răducu, D. (2018). Pedobioplasma illuviation - a new theory. *18th International Multidisciplinary Scientific Geo Conference SGEM, Conference proceedings, Water Resources. Forest, Marine and Ocean Ecosystems; Soils, Forest Ecosystems, Marine and Ocean Ecosystems, 18*(3.2), 357‒363.
Roper, M. (2006). Potential for remediation of water repellent soils by inoculation with wax-degrading bacteria in south-western Australia. *Biologia 61*, S358‒S362.
Stefan, M., Gheorghe, D., Ion, V. (2018). Results regarding the impact of crop rotation and fertilisation on the grain yield and some plant traits at maize cultivated on sandy soils in south Romania. *AgroLife Scientific Journal, 7*(2), 124‒131.
Štyriakov, I., Štyriak, I., Nandakumar, M.P., Mattiasson, B. (2003). Bacterial destruction of mica during bioleaching of kaolin and quartz sands by *Bacillus cereus*. World *Journal of Microbiology and Biotechnology, 19*(6), 583‒590.
Unkovich, M.J. (2014). *A review of the potential constraints to crop production on sandy soils in low rainfall south-eastern Australia and priorities for research*. A technical report for the Grains Research and Development Corporation. Mallee Sustainable Farming, Mildura NSW, 96.
Young, I.M., Crawford, J.W., Nunan, N., Otten, W., Spiers, A. (2008). Chapter 4. Microbial Distribution in Soils: Physics and Scaling. *Advances in Agronomy*, *100*, 81‒121.
Zamfir, B., Hoza, D., Vînatoru, C., Bratu, C., Barcanu, E. (2018). Assessing Tomato Germplasm from V.R.D.S. Buzau to Identify Genotypes with Distinct Features. *Scientific Papers. Series B, Horticulture*, *LXII*, 395‒398.
***EUROSTAT (2019). Organic farming statistics, https://ec.europa.eu/eurostat/statistics-explained/index.php/Organic_farming_statistics#Key_messages.
***WRB-SR-2006 (2006). *World reference base for soil resources*. World Soil Resources Reports No. 103, FAO, Rome: 116.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOIL DEGRADATION - A PRIMARY FACTOR IN THE DECLINE OF FERTILITY AND WAYS TO RECOVER IT

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Abstract

The paper presents the results obtained from an agrochemical study carried out in a site affected by soil degradation phenomena, being known the fact that the dramatic decrease of the agricultural production is due primarily to the deterioration of the soil fertility status, even though the rest of the technological chains of production are respected. The study was carried out, determining and analyzing the main soil agrochemicals, and the results obtained are a starting point for their improvement. Based on these results, it is possible to launch and carry out various actions to restore soil fertility, which will lead within a reasonable time, to obtain the expected, constant qualitative and quantitative productions.

***Key words**: soil degradation, abasement fertility.*

INTRODUCTION

Soil fertility is a decisive factor in the sustainable development and performance of agriculture. Being a complex soil, fertility provides the physico-chemical and biological conditions in the optimum, necessary for the growth and development of the crop plants, permanently supplying water and nutrients simultaneously to the proper development of vegetative phenophases. (Borlan & Hera, 1984)
The depreciation of soil fertility has as causes, a multitude of factors, recalling here the export of nutrients with the crops, which is not compensated by fertilization, but also the accentuation of the acidification or saltiness phenomena - which attract a whole range of disturbances in soil chemistry, erosion phenomena etc. (Cârstea, 2000)
The acidification of soils is mainly due to the unilateral application of nitrogen fertilizers on the same surface in large quantities and scientifically unfounded. These soils degraded by acidification are characterized by processes of alteration, leaching, debasification and migration of colloids and can often present deficiency or excess of moisture, but also of nutrients, implicitly (Davidescu, 1981).
As agro-pedo-ameliorative measures of these soils, some are important in achieving higher crop yields, such as calcareous refining, organic and / or mineral fertilization, as well as technological sequences such as scarification and the practice of suitable crop rotations.

MATERIALS AND METHODS

The research was carried out within a family association, Letcani village, in Iasi county, in 2016 and aimed at identifying the areas affected by acidification phenomena on the agricultural holding, by laboratory analysis of the soil samples taken and the issuance of possible good practices, to avoid accentuation of the deficiencies in the ionic composition of soils, with damaging repercussions on soil fertility and agricultural production.
The land stage consisted of the identification of the territory, marking the crates under analysis, the harvesting and labelling of soil samples.
The laboratory stage consisted in the preparation of soil samples for analysis.
There were determined: the pH in the aqueous extract, the mobile phosphorus content by the Egner-Riehn-Domingo method in acetate extract - ammonium lactate (P-AL), the available potassium content in the acetate-lactate extract ammonium, the determinations being made to the atomic absorption flame photometer (K-AL).

RESULTS AND DISCUSSIONS

Analysing the pH values for the selected areas, it was found that for all three sites, this indicator had values ranging from 5.4 to 6.8, including the soils analysed from the moderate acid to the weak acid to neutral.

For the Bedreag site, with a surface area of 20.7 ha, it was found that the pH ranges from 5.4 to 6.8, the distribution being 25% of soils with pH from 5.4 to 5.8, 50% soils with a pH between 5.8 and 6.4 and 25% of soils show a pH of 6.4 to 6.8 (Figure 1).

For the Vorontoia site, with a surface area of 30.45 ha (Figure 2), the pH distribution is 17% of surfaces with pH between 5.4 and 5.8 and 83% of the surface with a pH of between 5.8-6.4.

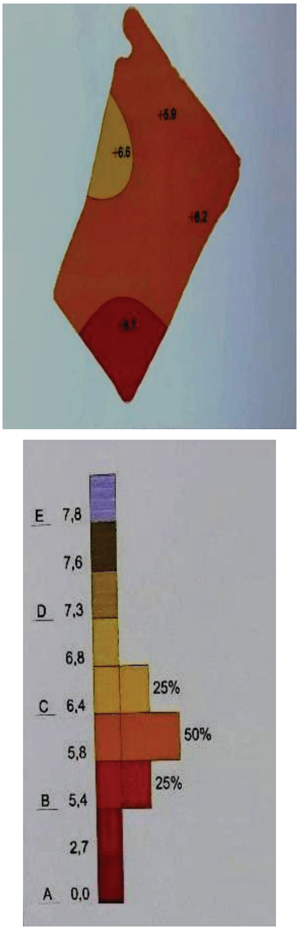

Figure 1. PH values, for Bedreag site

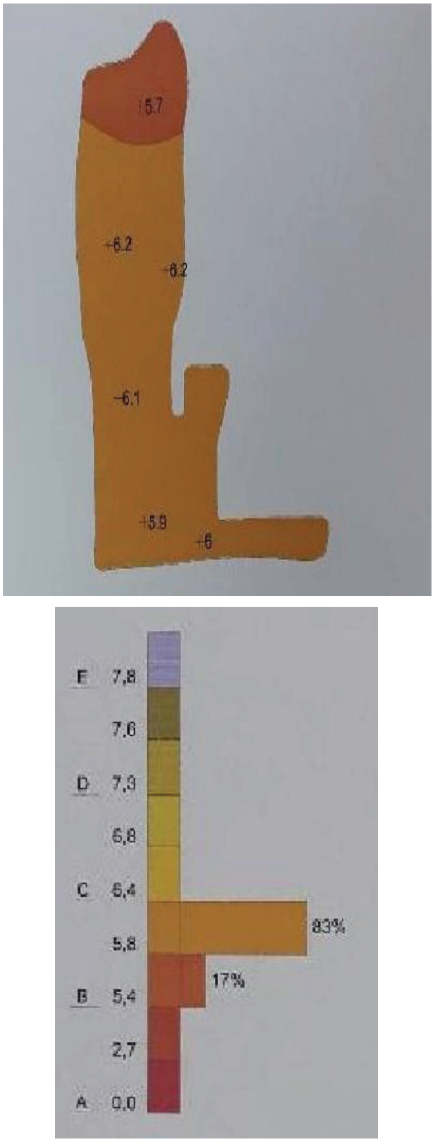

Figure 2. PH values, for Vorontoia site

For the site located in Toloaca, with an area of 11.81 ha, the pH ranges from 5.4 to 5.6 per 33% of the surface and from 5.6 to 6.8 on 87% of the surface.

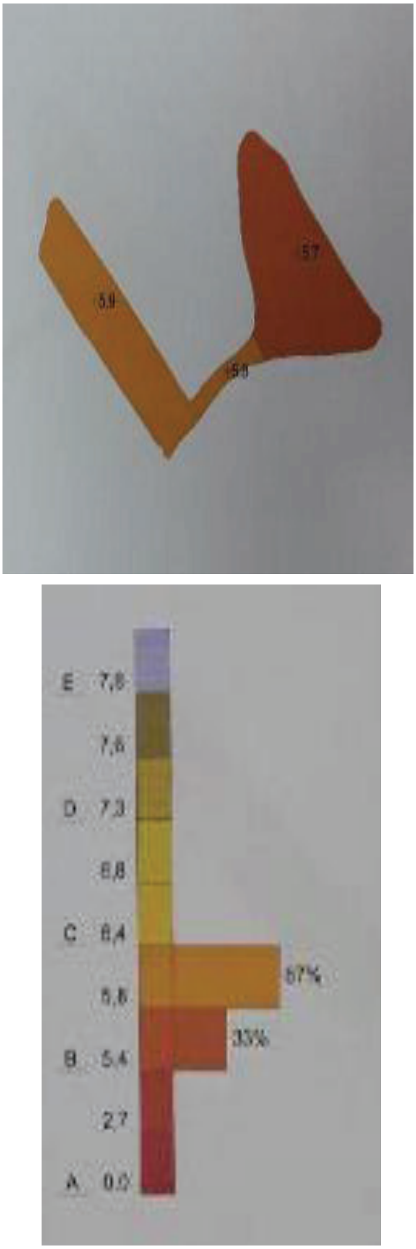

Figure 3. PH values, for Toloaca site

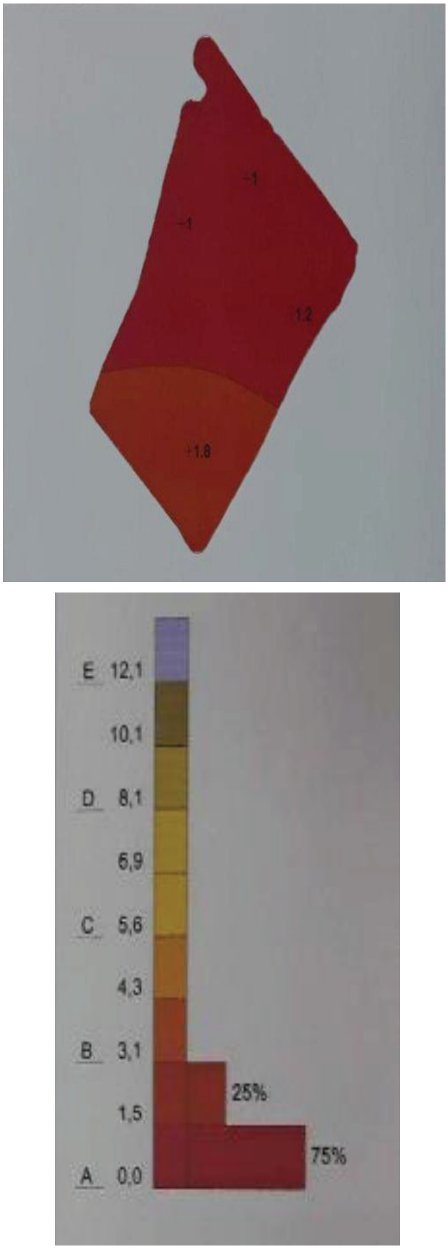

Figure 4. P-AL values (mg/100g sol), for Bedreag site

The soil response influences the accessibility of plant nutrients. In this sense, the acidic reaction of soils, when the pH falls below 5.8, causes the phosphorus to be demodulated, basic phosphates of iron and aluminium, of which phosphorus is difficult or not at all accessible to plants.

This can be highlighted in all locations, with mobile phosphorus contents ranging between 0 and 1.5 mg P-AL/100 g soil - poor supply - on 75% of the surface for the Bedreag site, 50% for the Vorontoia site and, respectively 33% for Toloaca (Figures 4, 5, 6)

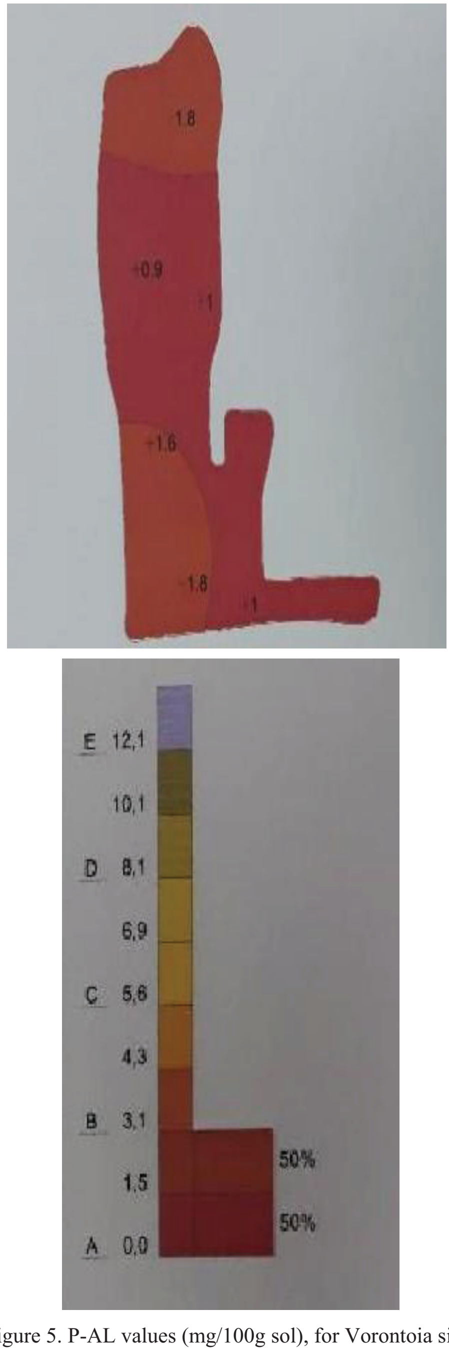

Figure 5. P-AL values (mg/100g sol), for Vorontoia site

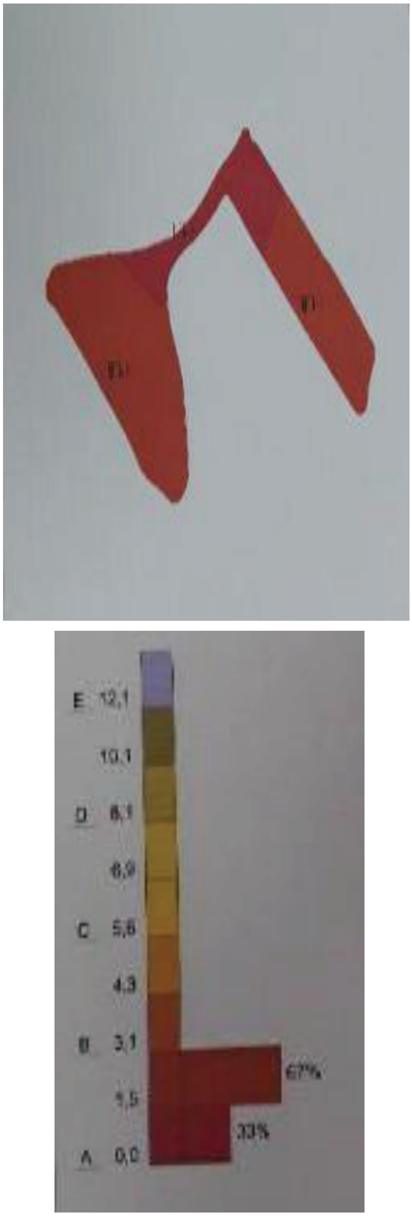

Figure 6. P-AL values (mg/100g sol), for Toloaca site

Another nutritive element that is in direct relationship to soil pH is the available potassium. Its undesirable capacity, in case of acidification of the soil, to pass from the soil solution into exchangeable forms, then in unchangeable forms and finally in fixed forms, represents a shortcoming for the agricultural crops, and thus it cannot be assimilated.

The analysis of the laboratory results on potassium revealed that on all the studied surfaces the available potassium has suffered, the resulting amounts being insufficient for a balanced nutrition of the plants.

In the case of the Bedreag site, the values of the mobile forms of potassium are between 5 and 10 mg K-AL, mg/100 g soil - 50% of the surface and between 10 and 13 mg K-AL K mg/100 g soil, which the specialised literature considers to be a low and medium supply with this element (Figure 7).

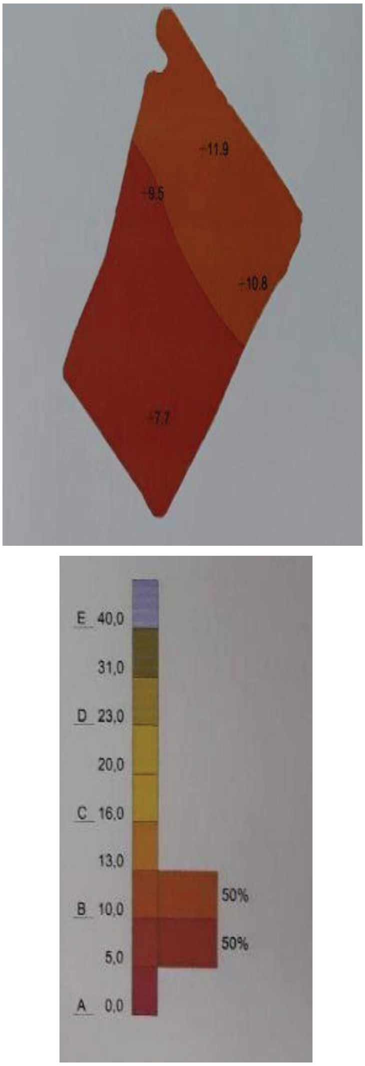

Figure 7. K-AL values (mg/100 g sol), for Bedreag site

For the Vorontoia site, the accessible potassium is approximately the same, occupying 67% of the surface with values of 10-13 mg/100 g soil, 17% of the surface with values ranging from 13 to 16 mg/100 g soil and another 17% of the surface with significantly higher values, respectively 16-20 mg K-AL/100 g soil (Figure 8)

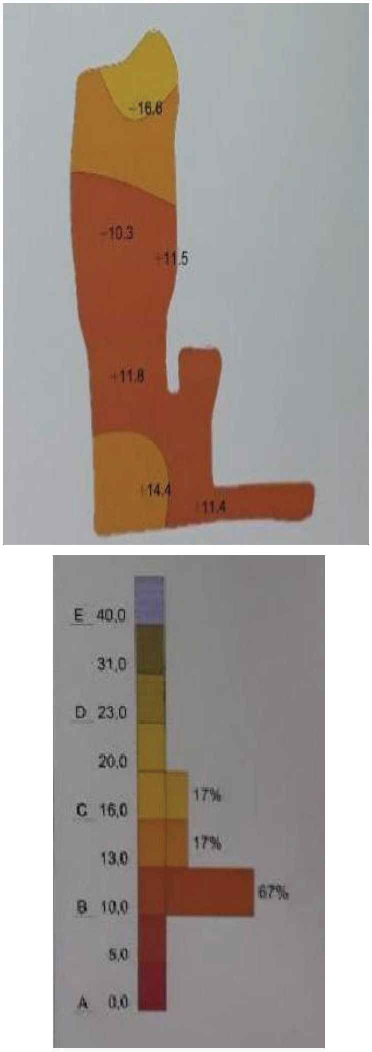

Figure 8. K-AL values (mg/100 g sol), for Vorontoia site

Figure 9 shows that in the Toloaca site the content of the mobile potassic forms is between 10 and 13 mg K-AL mg/100 g of soil per 100% of the surface, a totally inadequate quantity for assuring quantitative and qualitative production.

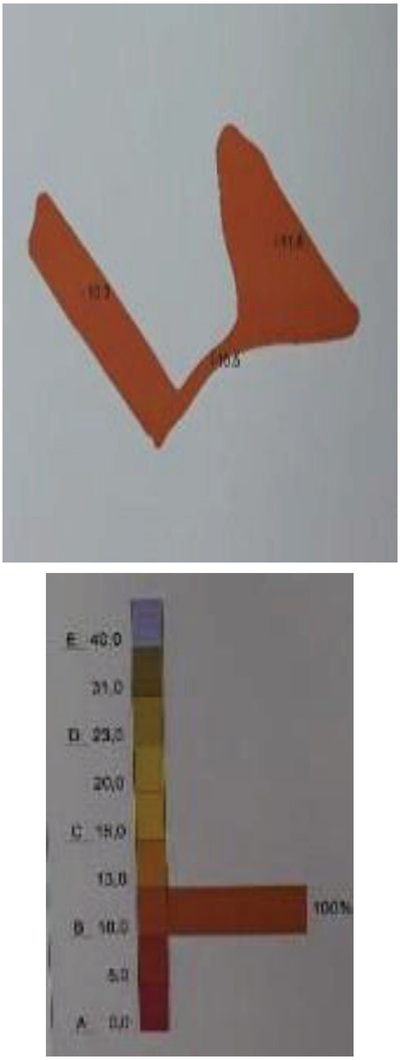

Figure 9. K-AL value (mg/100g sol), for Toloaca site

CONCLUSIONS

The obtained results highlight the necessity of periodic determination of the soil reaction state, in order to prevent degradation occurring in soil chemistry, respectively for the preservation of phosphorus and potassium, in accessible forms and in optimum quantities.

The pH values below 5.8 determined the dramatic decrease in the accessible phosphorus content, which also attracts the retardation of potassium, their synergistic relationship being known.

Above the pH values of 6.4, the assurance status with phosphorus and potassium is significantly improved, tending towards optimal concentrations.

As measures to counteract the harmful effect of acid pH, the use of calcareous amendments is indicated and for the increase of soil fertility, the administration of organic and chemical fertilizers in optimal economic doses and judiciously chosen assortments.

REFERENCES

Borlan, Z., Hera, Cr. (1984). *Metode de apreciere a starii de fertilitate a solului in vederea folosirii rationale a ingrasamintelor*, Bucharest, RO: Ceres Publishing House.

Cârstea, St. (2000). Unele aspecte ale strategiei privind protectia, ameliorarea si utilizarea durabila a solurilor din Romania, *Stiinta solului*, *XXXIV*(1).

Davidescu, D., et al. (1981). *Agrochimia*, RO: Didactical and Pedagogical Publishing House.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

PHYSICO-CHEMICAL PARAMETERS AND ENZYMATIC ACTIVITIES ON TILLED SOILS FROM JIBOU AREA, ROMANIA

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Abstract

Different soil samples from five tilled areas of Jibou (Salaj County, Romania) have been physico-chemically and enzymologically studied. Several physico-chemical parameters of tilled soil from these sulphurous areas were determined: pH, conductivity, humus and different mineral ion contents. Also, in the soil samples, the following enzymatic activities have been quantitatively determined: actual and potential dehydrogenase, phosphatase and catalase activities. In all soil samples all enzymatic activities have a uniform behavior, without big differences between the five sampling zones. Also, the physico-chemical parameters were very uniform, as compared to the other sampling sites. The enzymatic potential was appreciate by the enzymatic indicator of soil quality (EISQ), calculated on the base of these enzymatic activities. The EISQ values were good, between EISQ = 0.574 and 0.635. The granulometric analysis of the five soil samples classified them as loamy-clayey soils.

***Key words**: tilled soil, enzymatic activity, Enzymatic Indicators of Soil Quality, Jibou.*

INTRODUCTION

Most of the processes which are essential to maintaining the terrestrial ecosystems occur at soil level (Roger-Estrade et al., 2010). The soil represents the internal lab of the ecosystems, the deposit of elements needed for their optimal functioning. The organic matter of soil accomplishes more functions, such as: contributing to the structure and porosity of soil, adjustment of cations absorption and water retention, solubility control, mobility and availability of macro and micro-elements, carbon source for microorganisms, nutrients supply for vascular plants and many other (Bieganowski et al., 2013; Paul, 2014; Bojko & Kabala, 2017). It maintains a high biodiversity, by its physico-chemical properties provides important functions and services to the ecosystems, as decomposition, nutrients cycle, sustainability of productivity, as well as resistance and flexibility to abiotic disturbances (Brussaard et al., 2007).
Microbial communities at soil level are the most susceptible indicators of the disturbances and the changes in land use (García-Orenes et al., 2013). The soil quality represents a more and more essential assessment tool, important by quantitatively describing the structure of the microbial communities, along with the enzyme activities (Zelles, 1999; Zornoza et al., 2009). Due to the existence of a functionality bond between the environment and soil, elements regarding the influence of the microorganisms in soil, as microbial activity, were proposed as indicators for assessing the soil response to different management practices (García-Orenes et al., 2013). It is well known that the biological activities in soil (microbial and enzyme) are largely correlated with the physico-chemical properties of soil and participate at the metabolism of organic matter, thus deriving the availability of nutrients in soil, as well as accumulation and mineralization (Zhang et al., 2018).
The soil use practices of the human activities can intervene by changing the physico-chemical and biological properties of soil and can also influence the functions of soil (Jangid et al., 2008). For instance, the sources of nutrients in the ecosystems can modify the ecological processes and can influence the global changes (Oro et al., 2013). These sources can modify the characteristics of soil and influence the biological, chemical and physical processes occurring at this level

(Macci et al., 2013). Yuan and Yue (2012) showed that the enzyme activities of soil are correlated with the microbial biomass and the microbial activity, because these catalyze the biochemical reactions and the cycling of elements in soil. For instance, the urease activity from soil is closely connected to the soil mineralization with nitrogen (N) and the nitrogen cycle, while the enzyme activity of soil phosphatase is involved in the metabolism and transformation of phosphorus (P) (Adamczyk et al., 2014). Different factors as humidity, light and temperature stimulate microbial and enzyme activities in soil, resulting an increase of nutrient content in soil. Previous studies also showed that the relation between C: N: P stoichiometry and enzyme activities of different functions responsible for mineralization and C, N and P equilibrium are very close (Sinsabaugh et al., 2009; Xu et al., 2017). In conclusion, this interaction between the physico-chemical properties of soil and their biological characteristics can be an important key for revealing the processes and mechanisms of planting ecosystems restoration (Adamczyk et al., 2014). Thus, by understanding the different physico-chemical properties and the biological activities of soil while restoring it, the essential variations of the status of nutrients in soil could provide a proper environmental result (Zhang et al., 2018). However, the relation between the microbiological properties, the enzyme activities and the nutrients content of soil is not fully understood yet.

The present study consisted in correlating some physico-chemical parameters with some enzyme activities of agricultural soils, from Jibou area, where multiple sulphurous springs are present.

MATERIALS AND METHODS

Soil samples. Five soil samples were collected in autumn of 2018 from Jibou area. The collection sites were: sample 1: N 47°15’04 and E 23°14’16; sample 2: N 47°15’06 and E 23°14’15; sample 3: N 47°15’07 and E 23°14’14; sample 4: N 47°15’08 and E 23°14’14 sample 5: N 47°15’09 and E 23°14’14. The soil samples were passed in a portable storage box and transported on ice into the laboratory for different analyses. The soil samples used for measuring enzymatic activity and physico-chemical properties were air-dried and then temporarily stored at 4°C, for further analyses.

Physical and chemical properties of soils. Soil samples were air-dried, ground and sieved (2 mm) prior to determination of available nutrients and soil characteristics. The pH and electrical conductivity (EC) of the soils were potentiometrically measured in aqueous fraction (1: 5) (ISO 10390, 2005). The humus quantity in the soil samples was determined by Walkley-Black method, while total nitrogen by Kjeldahl method. The availability of nutrients in soil was also tested. Mobile phosphorus was determined by colorimetric method (with ammonium molybdate) and with Metertech 830 Plus spectrophotometer, while mobile potassium was calculated by flame photometer method, by extraction with ammonium lactate-acetate using Sherwood flame photometer. Carbonates were determined by Scheibler method with Scheibler calcimeter. SO_4 anion was determined by precipitation with $BaCl_2$ and titration with sodium thiosulfate from aqueous extract aqueous extract. At all the soil samples a granulometric analysis by Kacinski method was done. All these analyses ware done in the lab of the Office of Pedology and Agrochemical Studies from Cluj and in the pedology lab of the University of Agricultural Sciences and Veterinary Medicine from Cluj-Napoca.

Enzymatic activities in soil samples. Activities of the following four enzymes in soil were measured in triplicate: phosphatase, catalase, actual and potential dehydrogenase (Alef & Nannipieri, 1995; Carpa et al., 2014). These analyses were done in the microbiology lab of Babes-Bolyai University Cluj-Napoca.

Dehydrogenase activity (actual and potential) was determined after 24 h incubation of the soil samples at 37ºC, with TTC solution, and expressed by the amount of the formed 2, 3, 5-triphenylformazan (mg formazan/g soil).

Enzymatic activity of dehydrogenases was determined at λ=485 nm by Able Jasco V530 spectrophotometer.

Phosphatase activity was determined after 24 h incubation of the soil samples at 37ºC, with phenyl phosphate disodium solution, and it is expressed in mg phenol/g soil. Phosphatase activity was determined at $\lambda = 600$ nm by using an Able Jasco V530 spectrophotometer.

Catalase activity was determined after 1 h incubation of the soil samples at room temperature. The residual H_2O_2 is determined by titration with $KMnO_4$ in the presence of H_2SO_4. Catalase activity was expressed in mg splitting H_2O_2/g soil.

The analytical data serves as the base for calculating the enzymatic indicator of soil quality (EISQ) (Muntean et al., 1996).

RESULTS AND DISCUSSIONS

The earth of each soil type is made of clay, dust and sand. Dependent on the share of the three granulometric fractions in the soil formation, each pedogenetic horizon can be classified in different texture groups, classes and subclasses. (Blaga et al., 2005). Because sampling was done at 5-15 cm depth, all the soil samples belong to the pedogenetic horizon A. The texture (granulometric composition) of these soil samples was determined in the pedology lab of the University of Agricultural Sciences and Veterinary Medicine from Cluj-Napoca, by pipette method and was concluded that all the samples have loamy-clayey texture.

According to the criteria set by the National Institute of Research-Development for Pedology, Agrochemistry and Environmental Protection (Blaga et al., 2005), it was valued that the reaction of the analyzed soils (pH) is weak alkaline (Table 1).

Assessing the soil pH is important not only for the pedology studies but also for the microbiology and enzymology ones, because the pH influences the physiological groups of bacteria, which reach an optimum at specific pH values.

Table 1. Physico-chemical parameters in tilled soils from Jibou area

Soil samples	pH	Electrical Conductivity (mS)	Humus (%)	SO_4 mg/mc/100 g soil	N_2 (%)	P (ppm)	K (ppm)	Carbonates (%)
1	8.02	0.24	1.6	19.20/0.40	0.093	129	612	6.70
2	7.98	0.21	1.4	19.02/0.36	0.091	122	600	6.51
3	8.05	0.26	1.5	19.15/0.39	0.099	127	610	6.66
4	8.03	0.27	1.7	19.21/0.40	0.098	126	611	6.69
5	8.04	0.28	1.6	19.23/0.41	0.095	128	612	6.72

The humus content in the soil samples from Jibou zone was assessed by wet oxidation and by titration method (Walkley-Black) in the lab of the Office of Pedology and Agrochemical Studies from Cluj-Napoca.

According to the methodology for pedological studies (Florea et al., 1987), the data obtained show a low humus content, no higher than 1.7% (Table 1).

The nitrogen uptake level was also assessed in the lab of the Office of Pedology and Agrochemical Studies, by direct Kjeldahl method and the obtained data were interpreted according to the next intervals: very low <0.100; low between 0.100-0.140; medium between 0.141-0.270; big between 0.271-0.600; very big >0.600 (Paulette, 2007).

We can say that the nitrogen uptake is very small in all the soil samples analyzed.

Also, the nutrients from the tested soil, as phosphorus and potassium, did not surpass the value of 129 ppm and 612 ppm, respectively, and the carbonate level reach a maximum of 6.72, in sample 5.

The soil can be viewed as a biological entity in which a complex of biochemical reactions occur.

The action of microorganisms on the substrates from the environment is carried out in an enzymatic way, thereby assessing the enzyme activities offers suggestive data and in a shorter time than the microbiological analyses, regarding the soil processes.

The enzyme activities were quantitatively determined: phosphatase activity, actual and potential dehydrogenases activities and catalase activity (Table 2).

The enzyme activities were assessed using UV-VIS Jasco-V530 spectrophotometer.

Table 2. Enzymatic activities in tilled soils from Jibou area

Soil samples	Enzymatic activities				EISQ
	Dehydrogenase activities (mg formazan/g soil)		Catalase activity (mg splitting H_2O_2/ g soil)	Phosphatase activity (mg phenol/g soil)	
	Actual	Potential			
1	0.282	0.989	48.62	9.354	0.625
2	0.278	2.144	46.58	6.275	0.609
3	0.499	1.110	43.52	10.796	0.574
4	0.303	1.782	47.94	9.128	0.633
5	0.363	1.671	48.28	9.456	0.635

Actual dehydrogenase activity (ADHA) (reduction of 2, 3, 5-triphenyltetrazolium chloride in samples without added glucose) and potential dehydrogenase activity (ADHP) (with added glucose) of soil samples was expressed in mg formazan/g soil by measuring the absorbance at 485 nm (Carpa et al., 2014).
As illustrated by Figure 1, ADHA was more intense in soil samples 3 and 5, followed by samples 4, 1 and 2. The least actual dehydrogenase activity was displayed by the soil from sample 2, where pH was also lower and probably influenced the development of microorganisms. The values are low, but they fall within the general tendency for enzyme activities of agricultural soils at which the actual dehydrogenase activity presents highest values in spring and lowest in autumn and summer (Drăgan-Bularda & Kiss, 1986).

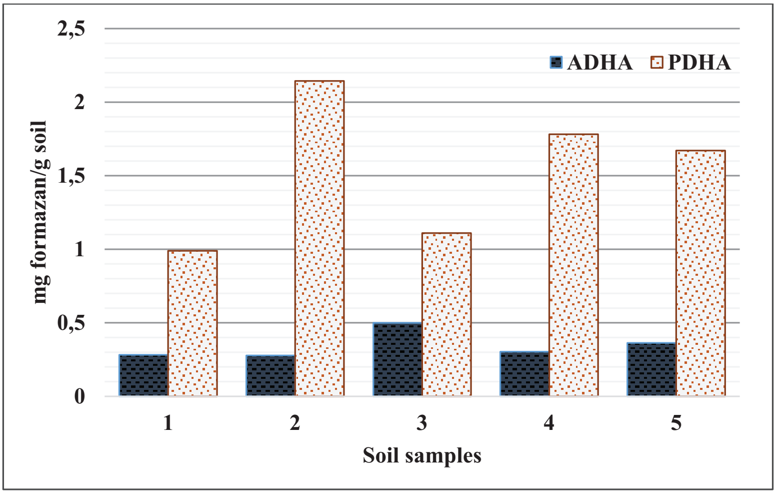

Figure 1. Actual and potential dehydrogenases activities (ADHA and PDHA) in tilled soil from Jibou

It can be noted that PDHA presents higher values in all samples, as compared to ADHA. The glucose addition has a stimulative effect on dehydrogenase activity. These enzymes which catalyze the oxidation of many organic compounds by exchange of electrons and protons are localized only in live, intact cells. Phosphatase activity of the soil samples was expressed in mg phenol/g soil by measuring absorbance at 600 nm (Carpa et al., 2014).
Phosphatase activity was detected in all soil samples. Phosphatase activity was more intense in sample 3, and weakest activity was recorded in sample 2 (Figure 2). Phosphatase activity was fairly good in the analyzed soils and this is supported also by the samples being taken in the autumn, when this activity grows due to the increase of organic matter in soil (Maheshwari, 2011). However, phosphatase activity is limited by availability of organic carbon in soil.

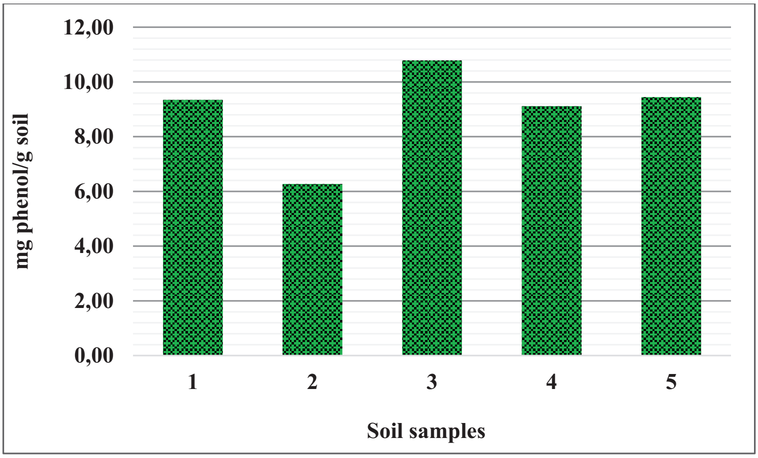

Figure 2. Phosphatase activity in tilled soil from Jibou area

Catalase activity is expressed by mg H_2O_2/g soil. Catalase activity is determined by expressing the breakdown intensity of peroxide (Carpa et al., 2014). Catalase activity was detected in all the soil samples analyzed (Figure 3). Especially in samples 1 and 5 catalase activity was more intense.

Catalase is an enzyme which accumulates in soil and thus keeps its activity for a long time. Also, catalase is correlated with the humus quantity in soil and pH. This explains the fact that in samples 1, 4 and 5 catalase activity was more intense, these spots being characterized by a higher humus content and a more alkaline pH (Table 1).

The quality of the soil enzymologically studied is characterized by the intensity of enzyme activities, defined by the values of the enzymatic indicator of soil quality (EISQ).

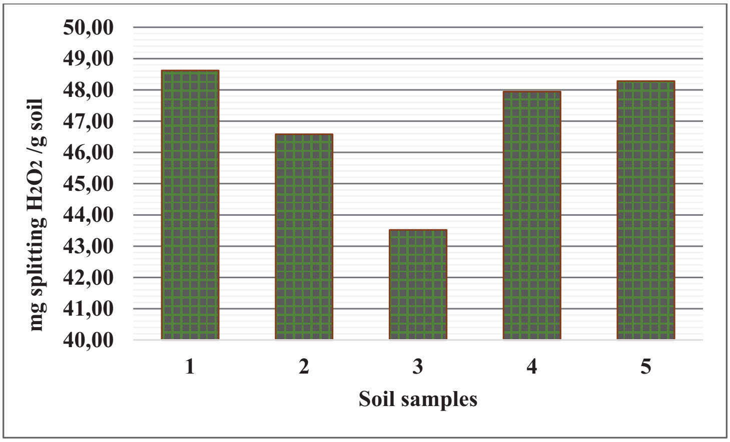

Figure 3. Catalase activity in tilled soil from Jibou area

The greater the enzymatic indicator is the higher the enzymatic potential of that soil. Enzymatic indicator of soil quality (EISQ) offers a general image on the enzyme potential of it, being calculated based on computing formula developed by Muntean et al. (1996).

Theoretically, the enzymatic indicator can reach values in the range 0 (when there is no activity in the studied samples) to 1 (when all the individual real values are equal to theoretical individual maxima of all activities). The enzymatic indicator of soil quality values are presented in Table 2.

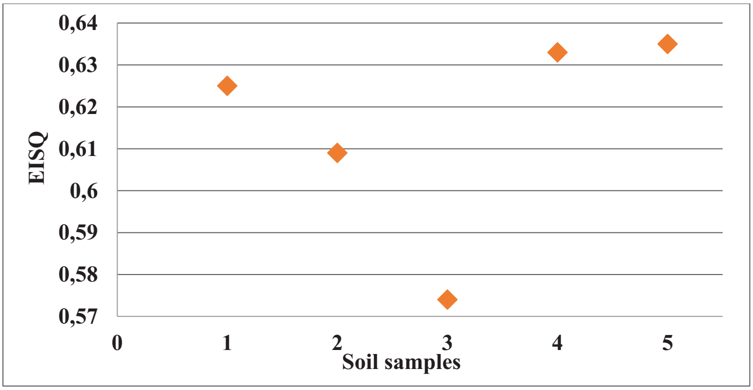

Figure 4. Enzymatic indicators of soil quality (EISQ) in tilled soil from Jibou area

The studied soils have a fairly high enzymatic activity. EISQ values have ranged between 0.574 and 0.633 (Figure 4). The quality of soil is better as the EISQ is higher (Muntean et al., 2001).

The enzymatic potential of a soil directly or indirectly reflects the microbiota activity, the influence of different physical, chemical, anthropogenic and even of the intensity of the different enzymatic activities in soil. Thus, the functioning of an ecosystem can not be understood without the active participation of enzymatic processes (Drăgan-Bularda et al., 2004). Based on the results obtained and compared with the data in published studies (Pasca et al., 1993; Carpa, 2007) we can consider that the analyzed soils have a wide biological potential.

CONCLUSIONS

Physico-chemical and enzymological properties of soils from Jibou area, where there are sulphurous springs, were the subject of this study.

Physico-chemical analyses showed that all the five analyzed soil types have a loamy-clayey texture and are alkaline. The weakest alkaline character was displayed by the soil in sample 2 (pH = 7.98). The humus quantity does not pass 1.7% and the nitrogen supply is small in all the analyzed soil samples. Also, the nutrients from the tested soil, as phosphorus and potassium, did not surpass the 129 ppm and 612 ppm, respectively. The carbonates value was maximum 6.72% in sample 5.

The enzymological research on tilled soils from Jibou area encompassed assessing 4 quantitative enzymatic activities, all of these displaying intensity fluctuations, according to the sampling place and the substrates needed for enzyme synthesis by microorganisms.

PDHA was greater than ADHA in all the studied soils, reflecting the stimulating effect of the easy available carbon source on synthesis of enzymes by microorganisms.

The values of dehydrogenase activity prove the existence of a microbial potential and a medium respiratory activity in the analyzed soils.

Phosphatase activity showed medium values which can be caused by the accumulation of plant debris in soil at the end of the growing season.

Catalase activity is correlated with the level of the humus quantity, being very intense in soil samples where the humus is more abundant. Based on the EISQ values, the soil in Jibou area has a fairly good enzymatic activity (values above 0.57). Based on these values it can be stated that the analyzed soils have a wide biological potential.

ACKNOWLEDGEMENTS

This work was supported by a grant of the Ministery of Research an Innovation, CCCDI-UEFISCDI, project number PN-III-P1-1.2-PCCDI-2017-0056: Functional collaboration model between public research organizations and the economic environment for the provision of high-level scientific and

technological services in the field of bio-economy, within PNCDI III.

REFERENCES

Adamczyk, B., Kilpelainen, P., Kitunen, V., Smolander, A. (2014). Potential activities of enzymes involved in N, C, P and S cycling in boreal forest soil under different tree species. *Pedobiologia, 57*(2), 97–102.

Alef, K., Nannipieri, P. (1995). *Methods in applied soil microbiology and biochemistry*. Academic Press, London.

Bojko, O., Kabala, C. (2017). Organic carbon pools in mountain soils-Sources of variability and predicted changes in relation to climate and land use changes. Catena, 149, 209–220.

Bieganowski, A., Witkowska-Walczak, B., Glinski, J., Sokolowska, Z., Stawinski, C., Brzezinska, M., Wlodarczyk, T. (2013). Database of Polish arable mineral soils: a review. Int. Agrophysics, 27, 335-350. http://dx.doi.org./10.2478/intag-2013-0003.

Blaga, Gh., Filipov, F., Paulette, L., Rusu, I., Udrescu, S., Vasile, D. (2008). *Pedologie*. Ed. Mega, Cluj-Napoca.

Blaga, Gh., Filipov, F., Rusu, I., Udrescu, S., Vasile, D. (2005). *Pedologie*. Ed. AcademicPress, Cluj-Napoca.

Brussaard, L., de Ruiter, P.C., Brown, G.C. (2007). Soil biodiversity for agricultural sustainability. *Agric. Ecosyst. Environ., 121*, 233–244.

Carpa, R. (2007). Microbial communities and enzymatic activities in the mountainous soils in Parâng. *Studia Universitatis Babeş-Bolyai, Biologia, LII*(2), 129–136.

Carpa, R., Drăgan-Bularda, M., Muntean, V. (2014). *Microbiologie Generală*. Lucrări Practice, Ed. Presa Univ. Clujeană, Cluj-Napoca (in Romanian).

Drăgan-Bularda, M., Kiss, S. (1986). *Microbiologia Solului*. Univ. Babeş-Bolyai, Cluj-Napoca, Uz intern.

Drăgan-Bularda, M., Grigore, C.E., Tura, D. (2004). The use of enzymatic indicator of soil quality for saline lakes capitalization and protection. *Studia Universitatis Babes-Bolyai, Biologia, 49*(1), 129–140.

Florea, N., Bălăceanu, V., Rauță, C., Canarache, A. (1987). *Metodologia de elaborare a studiilor pedologice.* Partea I, II, III, Institutul de Cercetari pentru Pedologie și Agrochimie București.

García-Orenes, F., Morugan-Coronado, A., Zarnoza, R., Scow, K., Cerda, A. (2013). Changes in soil microbial community structure influenced by agricultural management practices in a Mediterranean agroecosystem. *PloS One 8*. e80522.

Jangid, K., Williams, M.A., Franzluebbrs, A.J., Sanderlin, J.S., Reeves, J.H., Jenkins M., Endale, D.M., Coleman, D.C., Whitman, W.B. (2008). Relative impacts of land-use, management intensity and fertilization upon soil microbial community structure in agricultural systems. *Soil Biol. Biochem*., *40*, 315–323.

Macci, C., Doni, S., Peruzzi, E., Bardella, S., Filippis, G., Ceccanti, B., Masciandaro, G. (2013). A real-scale soil phytoremediation. *Biodegradation, 24*, 521–538.

Maheshwari, D.K. (Ed). (2011). Bacteria in Agrobiology: Crop Ecosystems. Springer-Verlag, Heidelberg, Dordrecht, London, New York.

Muntean, V., Crişan, R., Paşca, D., Kiss, S., Drăgan-Bularda, M. (1996). Enzymological classification of salt lakes in Romania. *Int. J. Salt Lake Res., 5*(1), 35–44.

Oro, D., Genovart, M., Tavecchia, G., Fowler, M.S., Martinez-Abrain, A. (2013). Ecological and evolutionary implications of food subsidies from humans. *Eco. Lett., 16*, 1501–1514.

Pasca, D., Crisan, R., Muntean, V., Dragan-Bularda, M., Kiss, S. (1993). *Enzymatic activity in soil of the Retezat National Park*. The Retezat National Park - Ecological Studies - Popovici, I. (ed.), Ed. West Side, Braşov, 117–129.

Paul, E.A. (2014). *Soil Microbiology, Ecology and Biochemistry*. 4th Ed. Academic Press, Cluj- Napoca.

Paulette, L. (2007). *Pedologie-Studiul solului în teren şi laborator.* Ed. Todesco, Cluj-Napoca.

Pascual-Rico, R., Morugán-Coronado, A., Botella, F., García-Orenesb, F., Sánchez-Zapata, J.A. (2018). Soil properties in relation to diversionary feeding stations for ungulates on a Mediterranean mountain. Applied Soil Ecology, https://doi.org/10.1016/j.apsoil.2018.03.017

Roger-Estrade, J., Anger, C., Bertrand, M., Richard, G. (2010). Tillage and soil ecology: partners for sustainable agriculture. Soil Tillage Res, 111. 33–40.

Sinsabaugh, R.L., Hill, B.H., Shah, J.J.F. (2009). Ecoenzymatic stoichiometry of microbial organic nutrient acquisition in soil and sediment. Nature 462 (7274) (795-U117).

Xu, Z.W., Yu, G.R., Zhang, X.Y., He, N.P., Wang, Q.F., Wang, S.Z., Wang, R.L., Zhao, N., Jia, Y.L., Wang, C.Y. (2017). Soil enzyme activity and stoichiometry in forest eco-systems along the North-South Transect in eastern China (NSTEC). *Soil Biol. Biochem.*, *104*, 152–163.

Zhang, W., Qiao, W., Gao, D., Dai, Y., Deng, J., Yang, G., Han, X., Ren, G. (2018). Relationship between soil nutrient properties and biological activities along a restoration chronosequence of *Pinus tabulae* formis plantation forests in the Ziwuling Mountains, China. Catena, 161, 85–95.

Zelles, L. (1999). Fatty acid patterns of phospholipids and lipopolysaccharides in the characterization of microbial communities in soil: a review. *Biol. Fertil. Soils, 29*, 111.

Zornoza, R., Guerrero, C., Mataix-Solera, J., Scow, K. M., Arcenegui, V., Mataix-Beneyto, J. (2009). Changes in soil microbial community structure following the abandonment of agricultural terraces in mountainous areas of Eastern Spain. *Appl. Soil Ecol. 42*. 315–323.

Yuan, B.C., Yue, D.X. (2012). Soil microbial and enzymatic activities across a chronose-quence of Chinese pine plantation development on the Loess Plateau of China. *Pedosphere, 22*(1), 1–12.

***ISO 10390:2005, Soil quality - Determination of pH.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EVALUATION OF BULK DENSITY AND SOIL WATER DYNAMICS AFTER BIOCHAR APPLICATION

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Abstract

The aim of the present study is to trace the dynamics of soil moisture and bulk density in field experiment with adding carbonated plant residues (biochar) to the soil of broad bean.The experiment was carried out in 2018 on the experimental field of the University of Forestry - Sofia. The experiment was set with two meliorants - biochar (tree different levels) and manure (used as a background). Adding carbonated plant residues to the soil is a good solution for improving some soil properties, reducing fertilizer levels and increasing yields. Biochar has been extensively studied in terms of its impact on the water-physical properties of the soil, but only a small part of the results is obtained from field experiments.The soil dampness is crucial for cultivation of agricultural crops. The most accurate assessment of humidification conditions can be made on the content of productive moisture in the soil, because it is the moisture that plants actually use. Measurements carried out with a high-frequency moisture meter show that the dynamics of soil moisture is a closely related to irrigation regime and climatic conditions.

Key words: *soil moisture, biochar, physical properties.*

INTRODUCTION

Over the last decades, the humanity has witnessed various extreme weather events. The modern agriculture is facing intense rainfall, prolonged periods of drought and extreme temperatures during the winter and summer months.

In recent years, in the context of climate change, there is a great interest in studying biochar (BC) use in agriculture (Lehmann, 2007). The concept of biochar is increasingly falling under political and academic attention, and in several countries (e.g. the United Kingdom, New Zealand, the United States) "biochar exploration centers" have been set up.

Biochar is a porous, carbon rich material produced by heating organic matter to temperatures between 300°C and 1000°C in an environment with limited or no oxygen (Verheijen et al., 2010).

Biochar has been extensively studied in terms of its impact on the water-physical properties of the soil, but only a small part of the results is obtained from field experiments. The soil dampness is crucial for cultivation of agricultural crops. The most accurate assessment of humidification conditions can be made on the content of productive moisture in the soil, because it is the moisture that plants actually use.

The regularities of the formation of productive moisture in the soil and their quantification in different climatic areas and in different soil types and climatic zones most accurately determine the crop yield.

Adding BC results in a reduction in the bulk density, increasing the total pore volume, and increasing the water content in the root zone. Soil bulk density (BD) is one of the most important physical characteristics affecting water infiltration and water holding capacity (WHC). Biochar, being highly porous in nature (Downie et al., 2009; Lehmann & Joseph, 2009; Sohi et al., 2010; Verheijen et al., 2010) and having a high surface area can decrease soil BD after soil amendment (Oguntunde et al., 2008). Decrease in soil BD due to biochar amendment may enhance soil aeration and porosity thereby improving soil WHC (Haider G., 2016.). Tammeorg et al. (2014) found improved plant available water content in the topsoil (20 cm) and decreased soil BD during the first and second years, respectively, following biochar amendment. Jien and Wang (2013) found a significant decrease in soil BD from 1.4 to 1.1 Mg m^{-3}, saturated hydraulic conductivity and soil aggregation by using (*Leucaena leucocephala* (Lam.) de Wit) biochar at 0 and 5% in acidic Ultisol.

The assessment of soil humidity is important for studying water movement, crops water stress, evapotranspiration, irrigation schedule, etc. Methods used to determine soil moisture (gravimetric, tensiometricaly, electro-resistive, electrically absorptive, neutron, capacitive, TDR, lysimetric, etc.) require pre-installation time at the measuring site. With the exception of weighing lysimeters, they are point methods and do not give representative data for large areas.

The aim of the present study is to trace the dynamics of soil moisture and bulk density in field experience with broad beans.

MATERIALS AND METHODS

The experiment was carried out in 2018 on the experimental field of the University of Forestry - Sofia. The soil is fluvisol, slightly stony, slightly acidic. This area came under a continental climatic sub region, in a mountain climatic region.

To assess the effect of BC on soil moisture dynamics and bulk density, a field experiment with broad bean variety - Supre Gualdaluche originating in Italy was set up. The sowing was carried out on 05.04.2018, by scheme 60+50+50/25 cm.

The experiment was set with two meliorants - biochar and manure (used as a background). During the spring cultivation, the two meliorants were incorporated into the soil and were developed six variants: 1) control - no biochar and manure; 2) only with manure - 4 t/ha^{-1}; 3) biochar - 500 kg/ha^{-1}; 4) manure + reduced amount of biochar (250 kg/ha^{-1}); 5) manure + optimal amount of biochar; 6) manure + increased amount of biochar (750 kg/ha^{-1}). The experiment was carried out by randomized complete block design with four replications and protection zones.

Plants are irrigated by a drip irrigation system, the tape drip hose used has the following characteristics: I-Tape 8 mil/distance between drippers 20 cm/ 5.3 lh. The irrigation rate is 40 mm.

In determining the dynamics of soil moisture, a high frequency moisture meter DM400 (HIGH FREQUENCY MOISTURE METER) was used. For device calibration are taken parallel soil samples and was evaluated by weight thermostatic method in parallel. To determine the impact of BC on the soil water-physical properties as well as to further calibration precision, the soil density was determined to 40 cm depth in every 10 cm using the Kachynski method with cutting rings, with a volume of 100 cm^3.

RESULTS AND DISCUSSIONS

For the purpose of the experiment, information on climatic conditions and changes has been collected. An analysis of the meteorological conditions for the last 31 years was carried out in order to track the annual rainfall in the Sofia region (Figure 1).

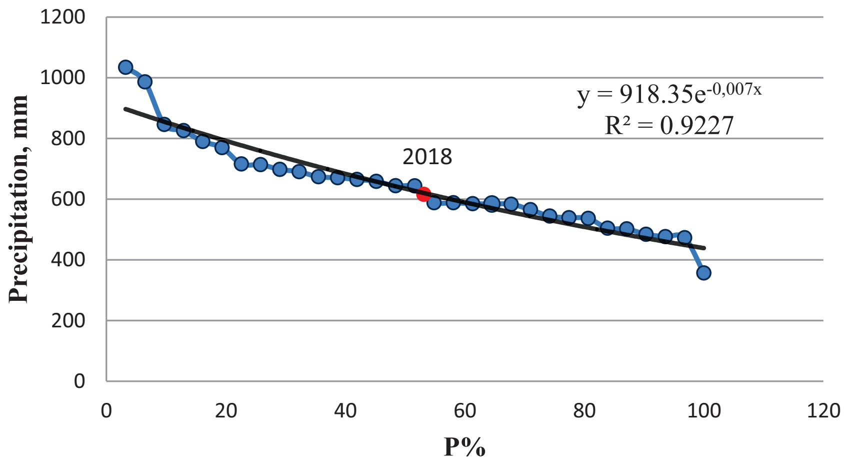

Figure 1. The precipitation provision curve for the period 1987-2018

Information on annual rainfall in the Sofia region is also collected. On the basis of this information curve of probability of precipitation per year has been prepared and according to the results obtained in 2018 it is characterized by secure close to 53%, which determines it as a mean dry year (Figure 2).

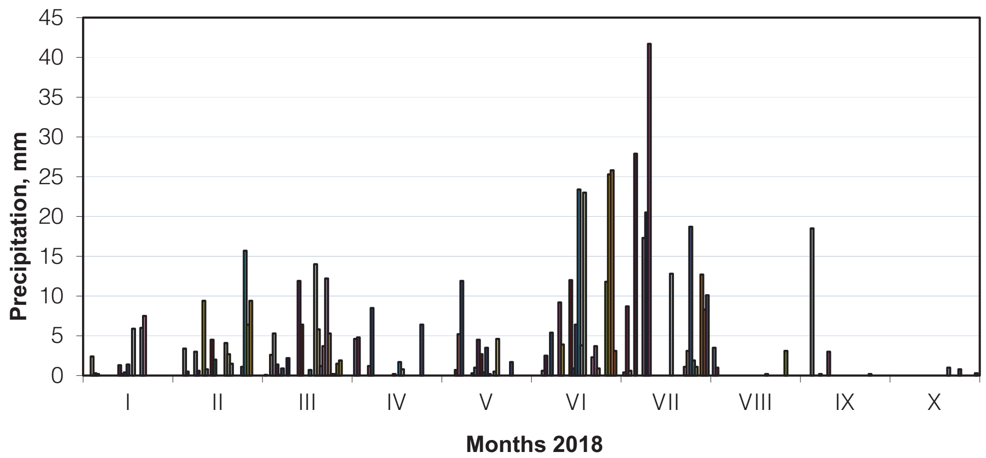

Figure 2. The precipitation for 2018, filed experiment - Vrajdebna

The reported precipitation from the beginning of vegetation period for faba bean (05.04.2018) is about 65.4 mm. In April, they range from 0.2 mm to 6.4 mm, mainly in the first two decades. These rainfall is insufficient to satisfy the culture's needs of water, which requires weekly irrigation. In May there is a uniform distribution, but the total rainfall for the month is 37.2 mm. The most intense rainfall was recorded on 07.05.2018 - 11.9 mm. Rainfall in April and May is below the average for a long time in the region of Sofia - V-54 mm and 72 mm for VI.

After the incorporation of soil ameliorants, in 2018, samples for evaluation of bulk density were taken from all variants, in the beginning of April The data are shown on Figure 3.

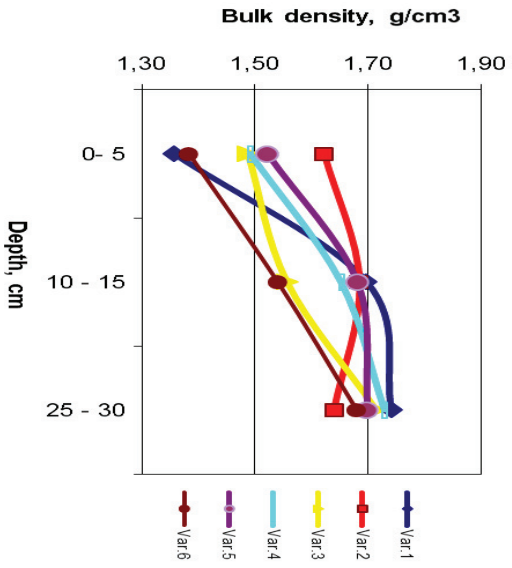

Figure 3. Bulk density of the soil - Vrajdebna, 2018

Data show that the introduction of BC and manure does not have the significant impact on bulk density. The bulk density increases in depth by moving from 1.36 to 1.74 g/cm^3. The lowest bulk density is in variant 6 with the highest content of BC.
This result probably due to the great amount of stones in soil, which that affect bulk density and humidity and cause greater variation. The compaction of surface layer is most likely due to irrigation. Similar data has also been obtained by Githinji L (2013) which concludes that with the increase of the imported BC the density volume decreases.
Application of BC improves the physical properties of the soil, but the results depend on the type (pyrolysis conditions and type of biomass) from the type of soil on which is applied and the rate of application.
Parallel to the direct soil sampling was measured with a high-frequency hygrometer, the data obtained are shown in Figure 4.

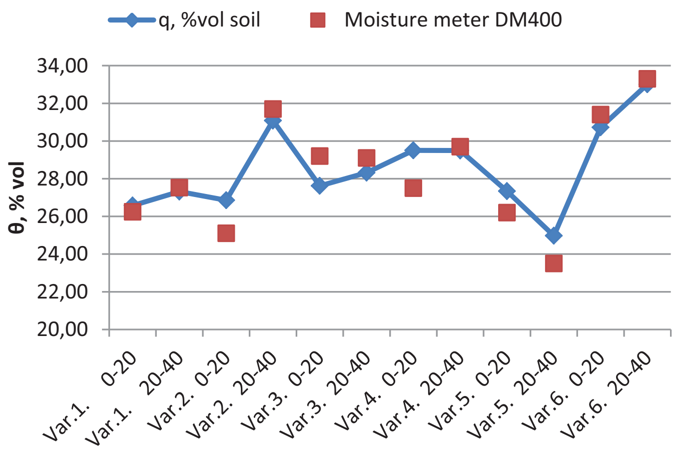

Figure 4. Data from soil moisture measured by two methods -2018

Because the device measures humidity by volume, it is necessary the result obtained by thermostatic method to be multiply by BD. In the data processing and the conversion of the weight moisture to volume, the average density (1.61 g/cm^3) for the layer of 0-30 cm was used. It is clear from the graph that the measurements in the different variants in the two selected methods almost completely coincide. In some places, deviations of ± 0.5% are observed, which is within the standard error of the appliance specified by the manufacturer.

Based on the sampling, the dynamics of soil moisture (Wt%) in the main root zone is monitored. The data are reported by variants to track the influence of BC on the soil (Figures 5, 6, 7).

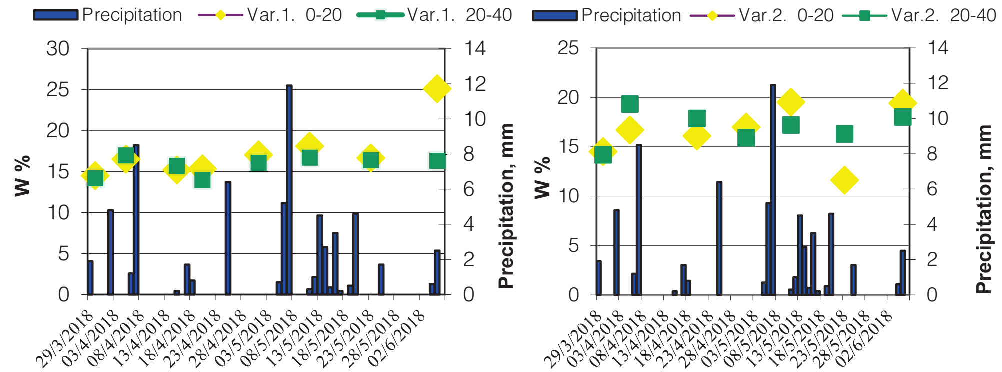

Figure 5. Dynamics of soil moisture in variants 1 and 2 - Vrtajdebna, 2018

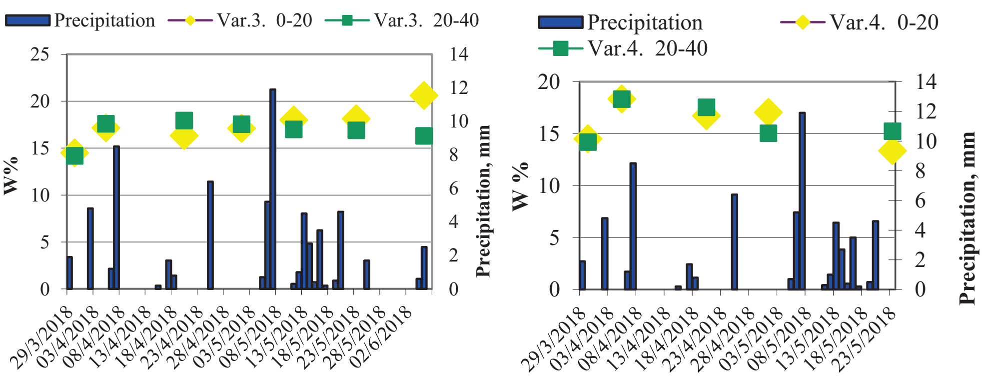

Figure 6. Dynamics of soil moisture in variants 3 and 4 - Vrtajdebna, 2018

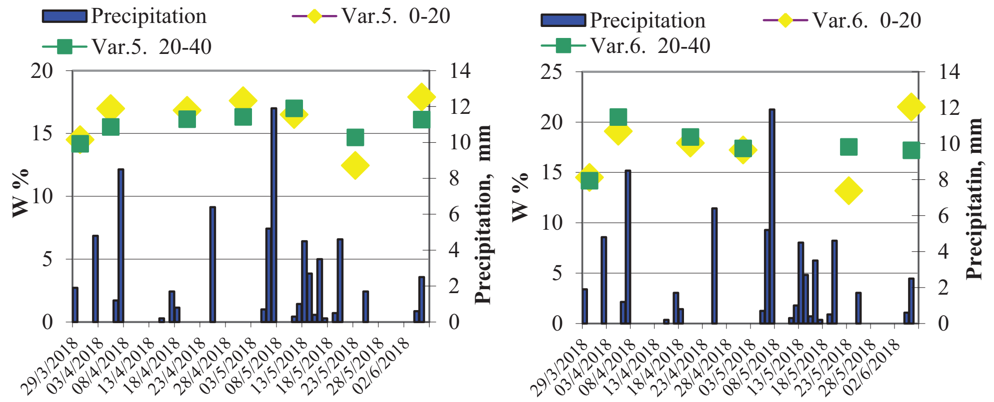

Figure 7. Dynamics of soil moisture in variants 5 and 6 - Vrtajdebna, 2018

At the beginning of the reporting period, moisture ranges between 16.5 and 20.5%, the lowest value being recorded in the control version of the two depths. Higher values of moisture are observed in depth, which has been shown for a good moisture load during the autumn-winter period. Highest values at both depths are reported for option 6 with an elevated rate of BC. This is due to the high porosity of the carbon, which increases the water holding capacity of the soil.

In early May, soil moisture ranges between 11.6% and 18.27%, with a decrease in moisture in the deep layer, due to the water derange in the underlying layers. Measured data at the beginning of June is one day after the irrigation, which also affects soil moisture regime. The moisture in the surface soil layer ranges between 16.1% and 25.1%. It has been noticed that the dynamics of soil humidity follow that of precipitation and applied irrigation. The available soil moisture in the soil is sufficient for the development of the faba bean, moving in the range close to the field capacity.

CONCLUSIONS

According to the obtained result biochar can be a good solution for improving some soil properties, reducing fertilizer levels and increasing yields.

The rainfall curves characterize the area of Vrajdebna-Sofia, as moderately humid and warm. Irregular distribution of rainfall required often irrigation of broad bean during the vegetation season. Accurate determination of the exact time of water application, as well as the correct calculation of the irrigation rate is of utmost importance. The need for a fast, accurate and non-destructive method required the use of the high frequency moisture meter. It was found that after accurate calibration for certain soil type, the method is accurate and fast.

The BD is determined by variant, the application of BG at the rate of 750 kg/da (var. 6) leads to a decrease in the soil density. In the other variants, no significant differences were found in compared to control variant.

It has been found that the dynamics of soil moisture follows that of the irrigation regime and climatic conditions. When saturation of soil with BC (var. 6) the highest and evenly distributed values are recorded. The available soil moisture in the soil is sufficient for the development of the broad bean, moving in the range close to the FC%.

ACKNOWLEDGMENTS

The data for this report was obtained during the implementation of the Project N158

Scientific Resurch Sector of University of Forestry, taitle: "Investigation of the Influence of Biochar on Soil fertility and Agricultural Crops Development". The articale is published with the financial support of Project No BG05M2OP001-2.009-0034-C01 "Supporting Scientific Capacity at University of Forestry" (2014-2020), co-funded by the European Union through European Structures and Investment Funds.

REFERENCES

Downie, A, Crosky, A, Munroe, P. (2009) Characteristics of biochar – physical and structural properties. *Biochar Environ Manag - Sci Technol*, 89–108. doi: 10.4324/9780203762264.

Ghulam, H. 2016. Biochar as a Beneficial Soil Amendment in Sandy Soils. Dissertation submitted for the award of the degree “Doctor of Natural Sciences”.Justus-Liebig-University Giessen, Germany.

Githinji, L. (2013). Effect of biochar application rate on soil physical properties of a sandy loam. *J. Agriculture and Environmental Sciences*.

Jien, SH, Wang, C.S. (2013). Effects of biochar on soil properties and erosion potential in a highly weathered soil. Catena 110:225–233. doi: 10.1016/j.catena.2013.06.021.

Lehmann, J. (2007). Bio-energy in the black. *Frontiers in Ecology and the Environment, 5*, 381–387

Lehmann, J., and Joseph, S. (2015). Biochar for environmental management: An introduction. In *Biochar for environmental management: Science, technology and implementation*. 2nd Ed. J Lehmann and S Joseph, 1-14. Routledge, New York.

Oguntunde, P.G., Abiodun, B.J., Ajayi, A.E., Van De Giesen, N. (2008). Effects of charcoal production on soil physical properties in Ghana. *J. Plant. Nutr. Soil Sci., 171*, 591–596. doi: 10.1002/jpln.200625185.

Sohi, S.P., Krull, E., Lopez-Capel, E., Bol, R. (2010). A review of biochar and its use and function in soil, 1st Ed. Elsevier Inc.

Tammeorg, P., Simojoki, A, Mäkelä, P., et al (2014b). Short-term effects of biochar on soil properties and wheat yield formation with meat bone meal and inorganic fertiliser on a boreal loamy sand. *Agric Ecosyst Environ.*, *191*, 108–116. doi: 10.1016/j.agee.2014.01.007

Verheijen, F., Jeffery, S., Bastos, C. et al (2010). Biochar Application to Soils.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF THE TILLAGE SYSTEM AND FOLIAR FERTILIZATIONS ON YIELD AND *Fusarium* EAR ROT MANIFESTATION IN MAIZE CROP IN THE TRANSILVANIA PLAIN

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Abstract

In the concept of sustainable agriculture, there is no valid universal soil tillage system, and systems are applied in a differentiated way due to the ecological features and the characteristics of the cultivated plants. The purpose of this paper is to study the behavior of a maize hybrid under the influence of tillage systems and foliar fertilizers in terms of tolerance to specific diseases and pests as part of an integrated sustainable agriculture management. The experimental factors studied: soil tillage: a_1- classical system with turning the furrow, a_2- minimum tillage, chisel variant, a_3- minimum tillage, disc harrow variant, a_4- no tillage, sowing directly and foliar fertilizers: b_1- control variant unfertilized; b_2- Haifa 19:19:19 + Mg + ME (5 kg/ha); b_3- Folimax Oleo 12-04-24 + 2.0% MgO + 36.5% SO_3 + ME (1.5 kg/ha), b_4- Folimax Gold 27% N + 1.5% MgO + 0.02% B + 0.2% Cu + 0.02% Fe + 1.0% Mn + 0.02% Mo + 0.02% Zn (3 l/ha). With vegetal remains left at the surface of the soil, conservative soil tillage systems that preserve at least 30% of the vegetal remains of the pre-plant increase the degree of attack of diseases and pests on maize crops having a negative effect on production and quality harvest. The highest maize yield was recorded in the classical tillage system (9566 kg/ha), with a very significant difference of 2192 kg/ha compared to the direct sowing system and 94 kg/ha, respectively 93 kg/ha in minimum tillage systems, and these yield differences are not statistically assured.

Key words: *maize, yield, tillage system, Fusarium ear rot, fertilization.*

INTRODUCTION

Corn, a new plant of culture for the Old World, is native to America, where it is cultivated by natives from ancient times. In Mexico and Peru, traces of old irrigation canals have been discovered, showing the particular attention that native maize culture enjoys (Muntean et al., 2008). Genetic and archaeological evidence suggests that maize domestication began about 9,000 years ago, most studies lead to the idea that the wild ancestor of maize was a herbivorous form of Teosinte (*Zea mays* ssp. *parviglumis*) (Matsuoka et al., 2002; Piperno et al., 2009; Flint-Garcia, 2017).

In the concept of sustainable agriculture development it is widely accepted that there is no valid universal soil tillage system due to local differences, especially climate and soil, but also because of the technical level of endowment. Soil conservation tillage systems in different areas must have certain specific characteristics in relation to ecological features and characteristics of cultivated plants, so they must be applied in a differentiated way (Canarache, 1999; Guş et al., 2004; Moraru & Rusu, 2013; Moraru et al., 2015).

The influence of the soil tillage system on the soil properties is important indicators for soil fertility conservation and the assessment of the sustainability of the agricultural system (Guş, 1997; Rusu, 2001; Rusu et al., 2017).

In the maize crop, by treating the seeds (before sowing) with fungicides, some or almost all of the pathogens present on the maize grain are eliminated, this being an effective measure of protecting plants against pathogens existing in the soil, preventing the transmission of one year to another in cultures (Cheţan, 2017).

Fusarium rot of cobs is one of the most common fungal diseases on corn cobs. It is caused by *Fusarium verticillioides* and by its symptoms it reduces the quantity and quality of production to 25%. In disease-favorable years, the massive accumulation of *Fusarium* mycelium biomass is recorded on cereals and cobs that lead to mycotoxin contamination, such as deoxynivalenol (DON), zearalenone (ZEA) and fumonisin (FUM) (Horia et al., 2018). Maize is susceptible to a large number of pathogens that invade the ears and seeds causing the rotting of the ear. The disease is prevalent in all regions where maize is grown. Generally, the disease rarely causes severe production losses (Şopterean, 2018).

An important role in *Fusarium* ear rot infections have, together with the genetic factor, the climatic conditions, but also the attack of pests *Ostrinia nubilalis* and *Helicoverpa zeae*, which increase their frequency and intensity (Horia et al., 2018).

The purpose of this paper is to study the behavior of a maize hybrid under the influence of tillage systems and foliar fertilizers in terms of tolerance to specific diseases and pests as part of an integrated sustainable agriculture management.

MATERIALS AND METHODS

The research was carried out in 2018, at Agricultural Research and Development Station Turda (ARDS Turda) located in the Transylvanian Plain, on a faeoziom vertic soil with neutral pH, clay-loam texture, good and very good supply with mobile phosphorus and potassium, soil content in humus medium. The experience is bifactorial, and the area of an experimental parcel is 48 m^2. In the experience, maize sowing was done with the MT 6 - Maschio Gaspardo machine. Sowing density was 65,000 plants/ha and the depth of seed incorporation was 5 cm. The rotation of crops is achieved in a 3 years soybean-wheat-corn system, the previous plant was winter wheat. The biological material was Turda 332 corn hybrid, created at ARDS Turda.

The experimental factors studied: Factor A-soil tillage: a_1- classical system with turning the furrow (CS), a_2- minimum tillage, chisel variant (MTC), a_3- minimum tillage, disk harrow variant (MTD), a_4- no tillage, sowing directly (NT); Factor B - foliar fertilizers: b_1- control variant unfertilized; b_2- Haifa 19:19:19 + Mg + ME (5 kg/ha); b_3- Folimax Oleo 12-04-24 + 2.0% MgO + 36.5% SO_3 + ME (1.5 kg/ha), b_4- Folimax Gold 27% N + 1.5% MgO + 0.02% B + 0.2% Cu + 0.02% Fe + 1% Mn + 0.02% Mo + 0.02% Zn (3 l/ha), doing two treatments. The first treatment was performed in the 8-10 leaves phenophase, and the second treatment was done in the 12-14 leaf phenophase. With the sowing, NPK 27: 13.5: 0, 250 kg/ha was fertilized, and a second fertilization was done in the 6-8 leaf phenophase, a nitrogen fertilizer of 120 kg/ha (a.s. 33 kg/ha N) in all variants.

For weed control, treatments were carried out comprising combinations of herbicides Tender 1.2 l/ha and Merlin Flex 0.4 l/ha at 260 l/ha of water applied pre-emergence and in the vegetation used Starane 1 l/ha at 260 l/ha of water.

The obtained results were statistically processed by the variance analysis method and the lowest significant difference was determined - LSD - (5%, 1% and 0.1%) (ANOVA, 2015).

The degree attack of fusarium ear rot was calculated according to the frequency and severity of the attack on 25 cobs per variant, and the frequency of attack of the pest *Ostrinia nubilalis* was determined by analyzing 25 cobs per variant.

Year 2018 was characterized as a warm year but normal in terms of rainfall recorded at the weather station, but the data analyzed monthly and decadal shows that all the months of crop growing were warm or hot months, except month July when we report average temperatures, as can be seen from the data presented in Figure 1.

The climatic conditions of the first crop period of corn crops were beneficial to optimal development at temperatures slightly higher than normal for this period, correlated with normal rainfall. The precipitations in 2018 and shown in Figure 2 indicate that the amount of rainfall varied monthly, the absence of precipitation during important periods of maize crop being recorded only during the production period. June, which was characterized as a slightly rainy month, recorded a higher amount of rainfall, 13.5 mm above the 60-year average we are reporting, with higher atmospheric humidity in this period being one of the most important factors favoring the attack on *Ostrinia nubilalis*.

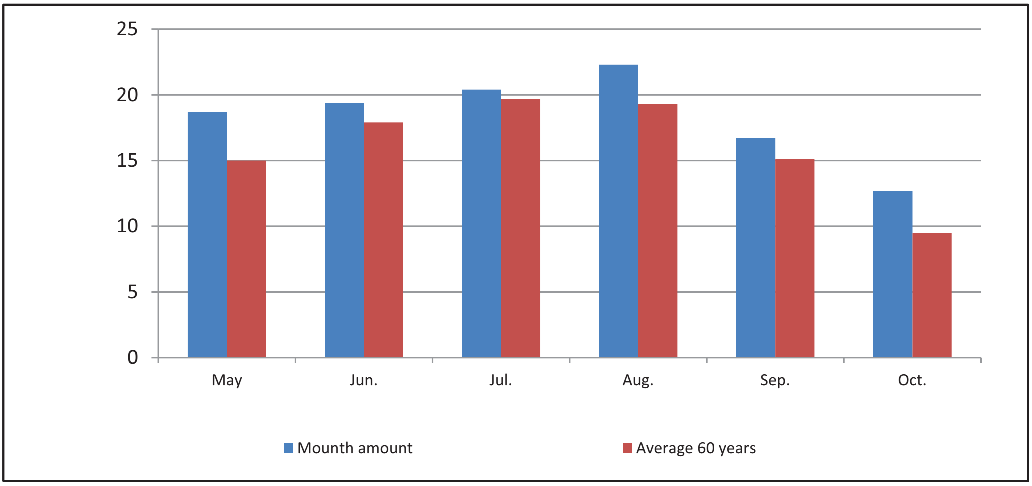

Figure 1. The thermal regime at Turda during 1 May 2018 - 31 October 2018

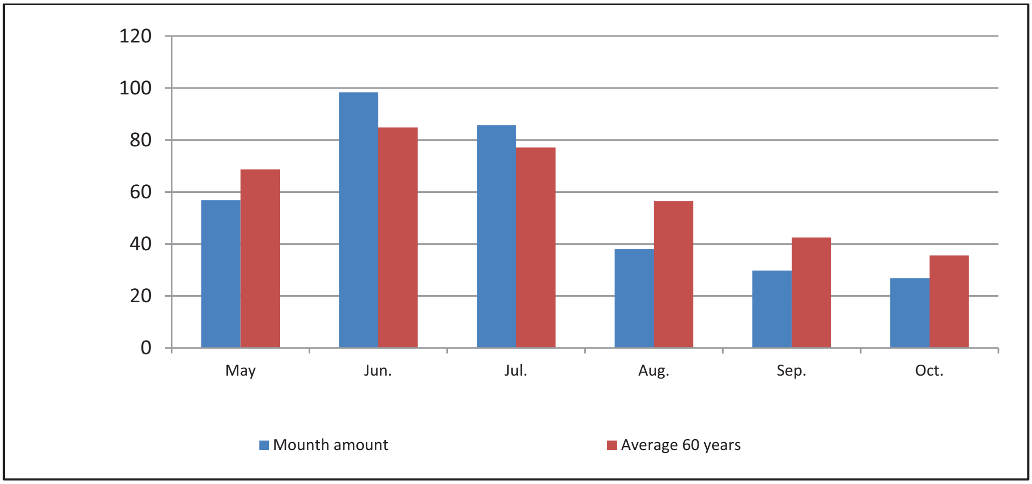

Figure 2. The pluviometric regime at Turda during 1 May 2018 - 31 October 2018

The precipitations that were dropped in early July are in addition to those recorded in June, prolonging the period in which the adults of the corn borer grow and plow.

The lack of rainfall in July and August puts its mark on corn crops by reducing the amount of water available to plants in the production of corn.

In maize culture, the water reserve was above the minimum ceiling in the early stages of vegetation, but due to the low rainfall this reserve decreased at a time when maize has significant requirements for water demand (the period between flowering and baking in the wax because the migration of the substances to the grain is diminished), the lack thereof resulting in a slight decrease in production due to the grain deflection.

RESULTS AND DISCUSSIONS

Maize responds best to the conventional soil cultivation system where the highest yields of 9566 kg/ha (Table 1) were obtained, but also to the minimum of tillage systems, where the soil was processed with the help of the chisel or the disc harrow, where the yields of the obtained are smaller, the differences of 94 kg/ha and 93 kg/ha are not statistically assured, in no tillage system, obtained the smallest yield, values with very significant differences of 2191 kg/ha compared to the control variant.

Similar data was also obtained from the experiments of Șimon et al. (2009-2011), from which it follows that the difference between the classical tillage system and the minimum tillage system was only 18 kg/ha.

Table 1. Influence of the soil tillage system factor on maize yield, Turda, 2018

Tillage variant	Yield (kg/ha)	Difference (kg/ha)	Signification
Classical system (CS) (control variant)	9566	0	Cv.
Minimum tillage (MTC)	9472	-94	-
Minimum tillage (MTD)	9473	-93	-
No tillage (NT)	7374	-2192	000
LSD (p 5%) 221 LSD (p 1%) 405 LSD (p 0.1%) 898			

The use of foliar fertilizers can positively influence production as shown in Table 2, where the use of fertilizers with a variable content of microelements in variants b_2 and b_4 showed very positive yields of 731 kg/ha and 761 kg/ha compared to the unfertilized control variant, the fertilizer used in variant b_3 recorded a significant positive yield of 331 kg/ha compared to the non-fertilized control fertilizer.

From the data presented, it appears that corn has benefited from the intake of microelements contained in the products used, contributing to the increase of yield by using them in vegetative phases where the maize plants have high water requirements, which has decreased with a decrease the amount of rainfall and relatively high temperatures.

Table 2. Influence of foliar fertilization factor on maize yield, Turda, 2018

Fertilization variant	Yield (kg/ha)	Difference (kg/ha)	Signification
b_1- Unfertilized (control variant)	8515	0	Cv.
b_2- Haifa	9246	731	***
b_3- Folimax Oleo	8847	332	*
b_4- Folimax Gold	9276	761	***
LSD (p 5%) 300 LSD (p 1%) 421 LSD (p 0.1%) 594			

From the results obtained from the analysis of the influence of the interaction of the experimental factors on the maize yield and presented in Table 3, it can be noticed that by applying the foliar fertilizers in the minimum tillage system, chisel variant, obtained a yield increase with distinctly significant differences, respectively very significant compared to the control variant to which no foliar fertilizer was applied. The lowest yield differences are registered in the soil tillage system with a disk harrow, which has a significant decrease in yield of 607 kg/ha after application of foliar fertilization, while the other two fertilization fertilizers have gains of non-assured yield. Significant production springs (641 kg/ha and 730 kg/ha respectively) are also obtained in the direct sowing system in the second and fourth treatment variants, the production increase obtained in the case of the application of the fertilizer from the third fertilization variant being not statistically assured.

By applying conservative tillage systems, the risk of specific diseases and pests increases significantly due to soil debris in which the pathogens and pests survive, which are often factors that are important in determining the production and quality of the crop, and as Șopterean and his collaborators say in 2017, in the climatic conditions in Romania, in general and in Transilvania in particular, the most damaging disease of maize crops that is compulsory to be considered in the improvement process is fusarium, produced mainly by two species of the *Fusarium genus (graminearum and moniliformes),* which may appear alone or associated with environmental conditions (soil, climate, technology etc.).

Table 3. Influence of the interaction of experimental factors on maize yield, Turda, 2018

The variant	Yield (kg/ha)	Difference (kg/ha)	Signification
Unfertilized x CS (control variant)	9047	-	Cv.
Haifa x CS	9837	790	*
Folimax Oleo x CS	9428	381	-
Folimax Gold x CS	9952	906	**
Unfertilized x MTC (control variant)	8610	-	Cv.
Haifa x MTC	9893	1283	***
Folima Oleo x MTC	9796	1186	**
Folimax Gold x MTC	9588	978	**
Unfertilized x MTD (control variant)	9465	-	Cv.
Haifa x MTD	9675	210	-
Folimax Oleo x MTD	8858	-607	0
Folimax Gold x MTD	9896	431	-
Unfertilized x NT (control variant)	6940	-	Cv.
Haifa x NT	7581	641	*
Folimax Oleo x NT	7306	366	-
Folimax Gold x NT	7670	730	*
LSD (p 5%) 598 LSD (p 1%) 840 LSD (p 0.1%) 1186			

From the data presented in Figure 3, following the interaction of the two experimental factors, compared to the classical soil management system considered as a control variant, one can notice an increase in the degree of *Fusarium* attack on the cobs in all three soil conservation conservative systems, with significant differences in the unfertilized variant with foliar fertilizer, In the Haifa fertilizer version there are no statistically ensured differences between the four soil cultivation systems; for the Folimax Oleo product, there is a significant increase in the degree of *Fusarium* attack in the application of the MTD system and statistically uninsured increases from the classical system to application of MTC and NT systems respectively. The greatest differences in the degree of *Fusarium* attack on the ear are recorded between the classical soil cultivation system and the conservative soil cultivation systems in Folimax Gold foliar fertilizer application.

Research by Chetan, 2015 on the occurrence of maize *Fusarium* indicates an increase in the degree of attack *Fusarium* on the ear in the minimal tillage system compared to the classic system. The lower attack rate determined in the classical soil cultivation system is considered to be due to the efficiency of the breeding in the prevention and control of this disease, there is a positive correlation between the degree of attack of *Fusarium* and the pests present in the maize culture, the plants attacked by the pests being more prone to disease.

Ostrinia nubilalis is found in all maize cultivation areas and is considered to be one of the most important pests of this crop in Transylvania, especially plants grown in conservative tillage systems.

The conservation of vegetal debris on soil surface in conservative tillage systems leads to a significant increase in the frequency of *Ostrinia nubilalis* attack since these vegetal remains increase the chances of increasing the biological reserve of diseases and pests in maize crop.

Although June was considered a warmer month than normal for this period, the fallen rainfall favored the emergence and development of adults of the species *Ostrinia nubilalis*, also increasing the life of the female, which manages to deposit more potes, thus the climatic conditions of the summer period being crucial in increasing the number of pests, the current climate change being an important factor in the multiplication of phytopathogenic agents and pests specific to maize crops.

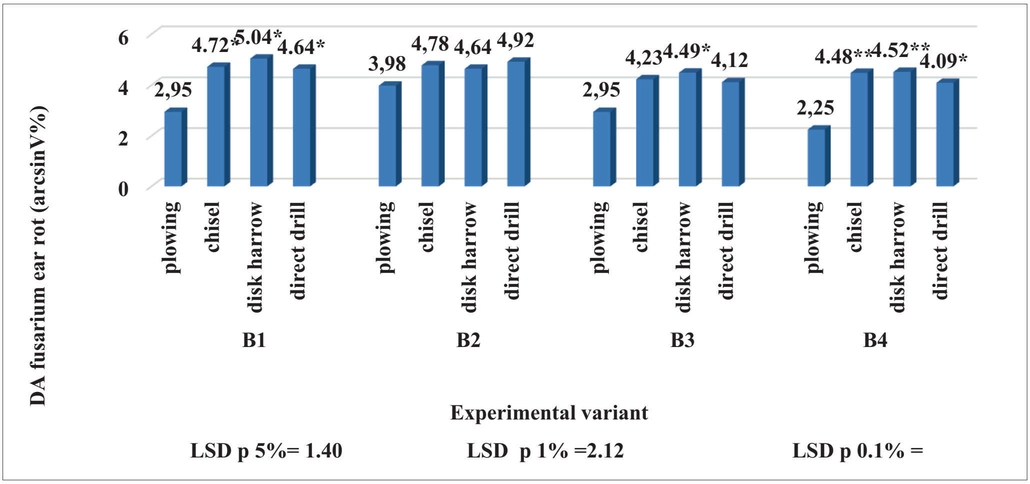

Figure 3. Influence of experimental factors on degree attack of *Fusarium* ear rot, Turda, 2018

The data presented in Figure 4 confirms that plant debris in conservative tillage systems leads to an increase in the frequency of *Ostrinia nubilalis* attack on maize cobs, but after application of foliar fertilizers, the frequency remains between 50,8-56,8% arcsin√% Haifa fertilizer and varies in variants where Folimax Oleo and Folimax Gold have been applied.

From research on maize crop the damage species produce to maize crops is direct, by qualitatively and quantitatively reducing the plant production potential and indirectly by breaking plants and cobs, making the process more difficult harvesting, as well as the fact that the larvae are a vector for blight, *Fusarium* and other diseases.

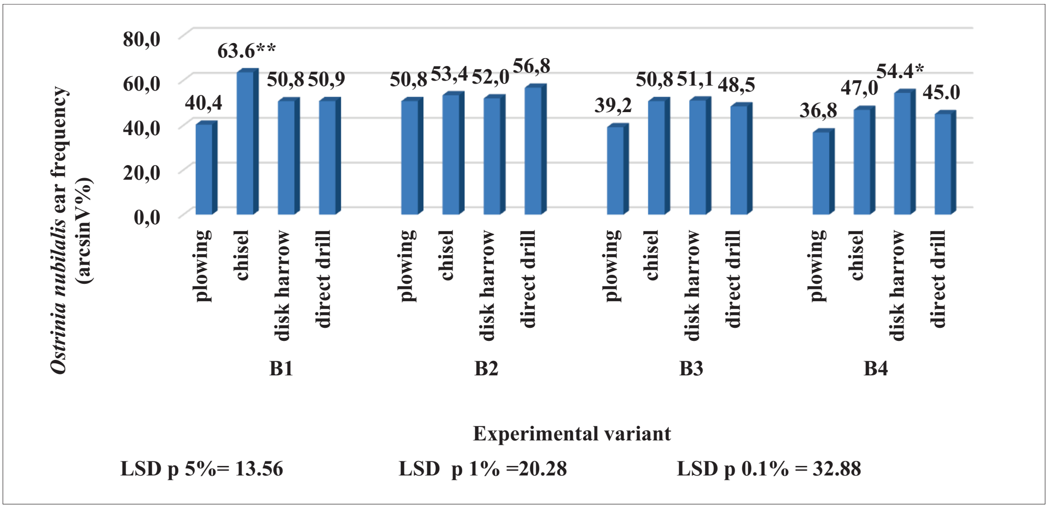

Figure 4. Influence of experimental factors on the ear frequency attack by *Ostrinia nubilalis*, Turda, 2018

CONCLUSIONS

By introducing the minimum soil tillage systems, maize crop yield decreases with statistically significant differences but following the direct sowing system, the yield decreases with a very significant difference of 2192 kg/ha compared to the conventional tillage system.

The application of foliar fertilizers to maize crop bring significant or very significant yield increases in the climatic conditions of 2018.

Soil tillage is of major importance in the emergence and development of phytopathogens of the species *Fusarium* and pest *Ostrinia nubilalis*.

ACKNOWLEDGEMENTS

This work was supported by a grant of the Ministery of Research an Innovation, CCCDI-UEFISCDI, project number PN-III-P1-1.2-PCCDI-2017-0056: Functional collaboration model between public research organizations and the economic environment for the provision of high-level scientific and technological services in the field of bio-economy, within PNCDI III.

REFERENCES

Carnache, A. (1999). Differentiation of soil tillage systems according to soil and land conditions in Romania. *Alternatives to soil tillage, Cluj-Napoca, II,* 215–223.

Chețan, F. (2017). Contributions of scientific research to the development of agriculture. Vol. VII, Homogeneous volume - 60 years SCDA Turda.

Chețan, F. (2015). Suitability of rotation soybean - wheat - corn cultivation system for the conservation properties of soil but aggressive fluid medium hilly. PhD thesis USAMV Cluj-Napoca.

Flint-Garcia, S.A., 2017. Kernel evolution: from teosinte to maize. In: *Maize kernel development*., CABI Publishing: 1–15.

Guş, P. (1997). The influence of soil tillage on production and soil characteristics. *Alternatives to soil tillage, Cluj-Napoca, II*. 151–155.

Guș, P., Rusu, T., Bogdan, I. (2004). *Agrotechnics*. Editura Risoprint, Cluj-Napoca, p. 524.

Iordan, H.L., Cană, L., Ciocăzanu, I. (2018). Behaviour of some maize hybrids to the attack of *Fusarium* spp. pathogen through artificial inoculation, at Fundulea, during 2015-2017. Analele *INCDA Fundulea, LXXXVI*, 61–83.

Matsuoka, Y., Vigouroux, Y., Goodman, M.M, Sanchez, G.J., Buckler, E. (2002). A single domestication for maize shown by multilocus microsatellite genotyping. *Proceedings of the National Academy of Sciences of the United States of America, 99*, 6080–6084.

Moraru, P.I., and Rusu, T. (2013). Effect of different tillage systems on soil properties and production on wheat, maize and soybean crop. *World Academy of Science, Engineering and Technology, Paris, France, 83*, 162–165.

Moraru, P.I., Rusu, T., Bogdan, I., Pop, A.I. (2015). Limitative factors for expanding soil tillage systems in the hilly areas of the Transylvanian Depression. *Agriculture - Science and Practice, 3-4*(94-95), 5–10.

Muntean, L.S., Cernea, S., Morar, G., Duda, M.M., Vârban, D.I., Muntean, S. (2008). *Fitotehnie*. Editura AcademicPres, Cluj-Napoca.

Piperno, D.R., Ranere, A.J., Holst, I., Iriarte, J., Dickau, R. (2009). Starch grain and phytolith evidence for early ninth millennium B.P. maize from the Central Balsas River Valley, Mexico. *Proceedings of the National Academy of Sciences of the United States of America, 106*, 5019–5024.

Rusu, T. (2001), The influence of minimum soil tillage upon the soil, yeld and efficiency. PhD Thesis, University OF Agricultural Sciences and Veterinary Medicine of Cluj- Napoca.

Rusu, T., Coste, C.L., Moraru, P.I., Szajdak, L.W., Pop, A.I., Duda, B.M. (2017). Impact of climate change on agro-climatic indicators and agricultural lands in the Transylvanian Plain between 2008-2014. *Carpathian Journal of Earth and Environmental Sciences, 12*(1), 23–34.

Șimon, A., Șopterean, L., Vălean, A.M., Chețan, F., Chețan, C., Ignea, M., Mureșanu, F. (2015). Influence of tehnological and biotic factors on some elements of the maize productions grown in the classical system and with minimum tillage at ARDS Turda. *Proenvironment, 8*(22), 142–149.

Șoptrean, L. (2018). Research on the behavior of maize hybrids and isonuclear inbred lines in pathogen attack of *Fusarium* spp. PhD thesis USAMV Cluj-Napoca.

***ANOVA, 2015, PC Program for variant analyses made for completely randomized polifactorial experiences.

***Stația Meteo Turda.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

ENGINEERING MEASURES FOR LANDSLIDE PREVENTING AND MITIGATION IN REDIU-ALDEI AREA, IASI COUNTY

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Abstract

The annual rainfall, relief and inappropriate anthropogenic interventions, in the absence of specific landscaping works in the slopes, have resulted in severe degradation processes in the Rediu-Aldei area. In the perimeter, some physico-geological processes were identified and localized, such as active, semi-active and stabilized landslides, ravines, collapsing areas, areas with excess humidity, the main weight being nonetheless landslides. The slope landscaping is opportune because the sliding process over time intensifies and has a major negative economic and social impact and even unpredictable environmental conditions. In this way the protection and the avoidance of particularly serious damages on the agricultural land in the perimeter, as well as other social-economic objectives in the area, consisting of numerous individual households, roads, electrical networks, can be ensured. At the same time, the degradation of environmental conditions and the improvement of existing ones, which are severely affected due to the landslides produced, are avoided. In the paper will be proposed and analyzed specific works on the existing field problems.

Key words*: anti-erosion works, soil erosion, landslides, Iasi county.*

INTRODUCTION

From an administrative point of view, the area where to propose solutions and works to prevent and combat landslides is located in Iasi County and is part of the Aroneanu village.

The area under study is delimited as follows: to the north - Valea Leta, to the east - Valea Leta and Valea Satului, to the south - the limit of the territory of the commune of Holboca and to the west Chirita brook. The access to the area is made on the communal road DC 17 Iaşi and DC 19 Iaşi - Golaeşti.

Rediu Aldei perimeter is located at the contact between the major geomorphological units - Moldavia Plain to the north and Moldavian Central Plateau to the south.

From the morphological point of view, the studied area meets the characteristics of both units, specific being the presence of the cue relief of cuestas.

The main hills that appear in the area on the left bank of the Chirita stream are oriented from north to south as follows: Leta hill, Coman hill and Rediu Aldei hill.

The physico-geological phenomena affect the entire morphology of the studied area, they are represented by active, semi-stabilized and stabilized landslides.

Due to the apperance loessoid soils and high relief of the area, there have been numerous active ravines and slopes on the slopes with depths of 2-10 m, the only stable areas being hillocks occurring in the confluence areas.

From a geological point of view, the researched area represents a small part of the large unit of the Moldovan Platform, which develops Sarmatian (Bessarabian and Kersonian) and quaternary deposits.

The underground waters in the perimeter are cantonated at the base of the quaternary deposits in the sandy formations of the latter. Their flow and chemistry varies greatly depending on the geological formations in which they are cantoned (on the slope or the meadow). In areas of landslides, several springs are reported, which are cantonated in the sandy intersections of the loessoid complex, some of which flow constantly, sometimes forming muddy portions. The number of springs can be higher, especially during precipitation periods.

From a climatic point of view, the area has a prominent continental character, falling into the sub-wet steppe. According to data from Iasi, the average annual air temperature is 9.4°C. The average for January is -3.7°C, and June for 20.4°C. The main feature of the area is the uniform distribution of rainfall and their torentiality. Thus, the average annual precipitation value is 537.5 mm, with maximum rainfall values in 24 hours during the vegetation period between 50 and 110 mm.

The water regime from precipitation indicates a low rate in the cold season with a minimum in February. In the warm season, rainfall sometimes has a pronounced torrential character, especially in the summer, when the showers are recorded with a high intensity. Hailstones are very rare and have a local character.

The wind regime shows that 27% of the time is calm, the rest of the time manifested in winter, winds from the northwest and north, and in the spring from the south-east and east.

From a hydrographic point of view, the area falls into the lower basin of the Bahlui River, whose valley flows from west to east and the Jijia River, which has the same flow direction.

The surface waters concentrated on the valleys separating the hills in the area represent the tributaries on the left side of the Bahlui River, of small importance and with low flows, oriented to NNV-SSE, the most important of which constituted the western limit of the studied area: the Chiriţa stream, which springs from the north of Icuşeni, crosses the villages of Rediu Aldei, the Lungă Valley and flows southwards into the accumulation of Chiriţa (Aroneanu).

In this brook a series of torrential runoff from the existing ravines on the adjacent slopes, and plugging existing beds and ponds downstream, causing excess humidity in the low meadow areas.

By regressive erosion and implicitly the deepening of the depth erosion formation, there has been a submergence of the sloping foot, the emergence of new springs, these being one of the main causes of the stability of the slopes and, implicitly, of the landslides in the area of Rediu Aldei.

For the pedological characterization of the analyzed area, the studies elaborated by the County Office of Pedological and Agrochemical Studies Iaşi.

From an economic point of view, agricultural lands represent 87.8% of the proposed surface area to be arranged with a capacity to works, of which the degraded grasslands have a significant share due to erosion and landslides, and non-agricultural land represents 12.2% of the total area proposed for landscaping, generally made of non-productive land due to active landslides and deep erosion.

MATERIALS AND METHODS

The detailed mapping carried out in the Rediu Aldei perimeter led to the identification and localization of physico-geological processes, such as active, semi-active and stabilized landslides, ravines, collapsing areas, humid excess areas, the main weight being the landslides.

As a working methodology, was used the inventtory of agricultural lands in the studied area, as they were recorded in the cadastral registers of Iasi County, on administrative units, on the most recent topographic maps and even with the help of the Google Earth site.

In order to know the complex problems of the quality of the ground-land units in terms of the sustainable use of land resources, a study was carried out on the current state of the land and land improvement works in the Rediu-Aldei area; at OSPA Iasi, APIA Iasi, North-East Region ANIF, Agricultural Chamber in Aroneanu. Soil maps were also used at stairs 1: 200 000 and 1: 100 000.

RESULTS AND DISCUSSIONS

To prevent and combat landslides, a series of works are proposed, namely:

- land planning with landslides on an area of about 420 ha through drainage channels of interception - capture - evacuation of groundwater which feeds landslides and eliminates excess moisture from the soil profile, collecting springs, interception and control channels for surface leakage and forest protection plantations.
- planning of valleys and ravines on approximately 8 km through transversal works which consists of reinforced soil dams, concrete thresholds and forced slopes, storm drains, including culverts on agricultural technology roads and forest protection plantations.

I Improvement works proposed and variants analyzed

Starting from the new existing conditions, regarding the land ownership structure in the perimeter, with a share of 59% of the private property, it is planned to design and carry out the works for prevention and control of the landslides, including the other works to combat soil erosion associated with a new design concept, with viable solutions that can be applied in stages.

On arable surfaces affected by surface erosion, specific works such as terraces and buffer strips can be done by landowners after increasing their private property or creation of associations, which will lead to an even more substantial reduction of soil loss on these lands.

Taking into consideration the specificity of the necessary fitting works and their purpose, it is important to perform a comparative technical-economical analysis of two or more variants. Several variants will be analyzed from the point of view of the planning scheme, as well as several constructive solutions for the realization of the works. So, the general scheme of landscaping proposed, results from the composition of the optimal technical and economical constructive solutions for each capacity, respectively type of works.

1. **Arrangament of slips** - They make up the main improvements works proposed by the present study, in terms of effect on improving the stability of sliding slopes and are proposed to be carried out on an area of 400 ha and comprises:

a) ***Works with interceptive, uptake and evacuation of groundwate -*** Interceptions, capture and rapid evacuation of groundwater, which feeds the landslides from the fronts groundwater, coming from the high plateau or rain fallen directly on the surface slopes and which is currently the main cause of landslides produced, complemented by the collection and discharge of stagnant water at the surface of the land, which produce significant beneficial effects, leading to a substantial increase in the stability factor of the slopes by:

- reducing the heap of the sliding mass, by maintaining a low moisture content of downstream land;
- reduction or disappearance of the erosion-reducing process and the internal friction angle of the soils forming the slipping mass;
- reducing the hydrodynamic force acting on downstream land by essentially modifying the underground current spectrum;
- avoiding infiltration of water at the level of the slip surface;
- avoiding the softening of the slope at the base of the detachment fronts;
- the delay of the physico-mechanical alteration process of the rock forming the massive at the top, by the catalyst factor that represents the water in the process of alteration;

b) ***Biological consolidation works through forest protection plantations -*** Are planned works both for the consolidation of the lands through the roots that carry out their biological reinforcement, and for the provision of forestry use on degraded surfaces due to landslides.

Forest protection plantations are proposed to be built on an area of about 35 hectares, in the areas of cornices, detachment steps, land with excessive erosion and active landslides, which cannot be used in the agricultural circuit. Plantations may be of the following species: willow (*Eleganus angustifolia*) - 20%, acacia (*Robinia pseudoacacia*) - 60%, wicker (*Salix purpurea*) - 20%.

2. **Planning of valleys and ravines.** It is proposed to be improvement about 10 km of valleys and ravines to prevent the development of landslides by undermining their base and defending neighboring agricultural lands through transversal works, escape channels and forest protection plantings.

a) ***Cross-sectional work.*** A possible scheme landscaping with works could be designed for production a compensation slope in the longitudinal profile on ravines, optimal technically-economical, comprising 1 earth dam, 7 concrete thresholds and 3 forced slopes.

In order to establish an optimal technical and economical solution development for the formation of deep erosion with transversal works to stabilize them and to retain the alluvial material, can be analyze several constructive solutions for the accomplishment of the works, with the provision of technologies and performance materials at the level those currently used worldwide.

Earthfill dams and forced slopes with heights between 3 and 5 m. The earthfill dam is analyzed in four constructive solutions, namely:

❖ Earthfill dam, with the spillway, rapid channel and disipator basin consolidated with reinforced concrete casting on the spot;
❖ Earthfill dam, with the spillway, rapid channel and disipator basin, consolidated with modulated prefabricated of reinforced concrete;
❖ Earthfill dam, reinforced with geogrids, with the spillway and the dissipator basin consolidated with modulated prefabricated of reinforced concrete;
❖ Filterable dams, from prefabricated elements.

Thresholds, with heights between 1.0 m and 1.5 m, were analyzed in three constructive solutions:
❖ Concrete thresholds;
❖ Thresholds made of modulated prefabricated of reinforced concrete;
❖ Gabion thresholds with galvanized wire mesh.

b) ***Drainage channels*** of water from valleys and ravines in the emissary. It is proposed for a length of 5 km to ensure the ability to transport of natural river bank and to avoid flooding in the locality of Rediu-Aldei and on the neighboring agricultural lands, in part to ensure the collector drainage rates.

In the built-up area, in small spaces or the dejection cones of the ravines, a consolidation solution that could be analyzed is the one with 100 mm geo-cell, filled with earth and covered with a layer of biodegradable geotextile, fixed by means of concrete steel anchors, a solution that would be very expensive.

c) ***Biological consolidation works*** through forest protection installations. Are proposed for a surface area of 7.0 hectares, for the consolidation of their embankment and thalweg with a planting scheme consisting of species wich are agreed by the animals: the willow (*Eleagnus angustifolia*), 2-3 rows on the banks of the ravine - 20%, sea-buckthorn (*Hippophae rhamnoides*), on embankment - 70% and wicker (*Salix purpurea*) on thalweg - 10%.

3. **Works to track evolution over time on landslide.** For the time tracking of the evolution of the landslides and the efficiency, respectively the behavior of the designed works, the following are proposed:

a) **A topometric landmarks network, consisting of:**
• The topometers that will highlight the support points that will be at the same time and the points of the station where the measurements will be made and which will be materialized by 8 reinforced concrete beams in the shape of a pyramid of great dimensions, planted in the lands stable;
• Topometric tracking landmarks, materialized by 20 reinforced concrete bollards, planted on lands with active landslides.

The final positioning of the topometric landmarks in the field and their installation will be done after the basic work has been done.

b) **A network of stationary hydrogeological,** composed of 12 observation wellss with an average depth of 12 m, arranged in 3 hydrogeological profiles through main slides, to track the evolution of groundwater levels and chemistry.

II. Works and agro-technical measures

In addition to the forest protection plantations, it is necessary to complement in addition a complex of works and measures of agropedoameliorative character, which ensure:

- Achieving a continuous slope of the land, including a gutters system, with the elimination of microdepressors where water stagnates and the sealing of cracks, allowing the regulation of superficial runoff and avoiding rapid infiltration of water to the sliding bed;
- Biological consolidation by grassed, by creating a well-worn herbaceous carpet on the lands covered with meadows;
- Transforming the non-agricultural use categories provided as a result of carrying out investment works for agricultural purposes;
- Increasing the productive capacity of the arable land and of the agricultural productions at the foreseen levels, mainly due to the elimination of the excess moisture in the soil profile in the areas where it is manifested on extended surfaces but also the reduction of erosion soil losses.

The necessary agropodoameliorative works and measures are:

1) Correction of the slope of the land by its easy modeling and the creation of drainage gutters on active landslides on a surface of 46 ha with a specific landfill volume of about 200 m3 / ha;

2) Works of creations and improvements of meadows on a total area of 218 ha, of which 46 ha of sowing (active landslides and non-agricultural lands which are transformed into pastures and meadows and on where land-modeling works are carried out) and 172 hectares of over-sowing (on semi-active and stabilized landslides), including necessary fertilization work. The following mixture of perennial herbs is recommended for sowing: *Bromnus inaermis* - 70% and *Onobrichis viciaepholia* - 30%. In case of oversowing, the following mixture will be used: *Bromus inaermis* - 50%; *Agropyrum cristatum* - 10%; *Onobrichis viciapholia* - 20%; *Lothus corniculatus* - 20%.

For fertilization of meadows for optimal germination conditions of herbaceous seeds and the creation of a rich herbaceous carpet, 200 kg/ha of active substance and calcium superphosphate 100 kg/ha of active substance will be used. These chemical fertilizers can be replaced by 20 t/ha of manure.

3) Works of deep loosening (scarification) 50-60 cm, an area of about 100 hectares to improve soil permeability for water and air, on land where excess moisture in the soil profile is manifested on extended surfaces and on which horizontal drainage works are planned.

4) Measures on the application on arable lands of specific agrotechnics on lands in the slope by performing all the cultural works only on the level curve and the crops in strips.

CONLUSIONS

By realizing the entire complex of proposed works for preventing and fighting the landslides in the Rediu Aldei area, it will be ensured first of all avoiding the reactivation and intensification of the landslides and of the deep erosion formations respectively, eliminating the possibility of extension of the out-of-area areas. At the same time, about 2 ha of land is transformed into productive agricultural land and turns into forest plantations about 42 ha, currently non-agricultural land, while removing excess surface moisture and soil profile and reducing soil loss through erosion, resulting in a significant increase of the agricultural output obtained on the other lands.

There are also beneficial effects on other socio-economic objectives, namely: avoiding to the damage to individual households in the vicinity of the slopes, avoiding damage to municipal roads in the area, reducing the intensity of clogging of Chiriţa accumulation.

Active landslides have profound consequences both on the environment and especially on the water regime, the floods being generated and aggravated by erosion as well as on social economic objectives.

Unlike the above synthetically presented, it results that landslides landscaped, simpler or more complex, can only have positive ecological consequences (favorable impact). Through the landscaping work, ecological changes are expected in wider spaces, including climate, flora and fauna.

Drainage works proposed in areas affected by landslides will allow the removal of excess moisture from the soil profile, preventing the infiltration of water to the sliding bed, thus reducing the risk of landslides triggering.

Designed transverse work will help to achieve the compensation slope so that the volume of the entrained materials is equal to the volume of deposited alluvial materials, leading to a deformation of the depth erosion process that could have negative influences on the stability of the slippery slopes. Thus, alluvial spills will be reduced and their deposition in watercourses, agricultural lands, crops will be diminished. At the same time, in the area of each transversal work, it will be possible to cover the soil with vegetation, which will lead to stabilization of the base of the slopes that will develop over time and due to the forest protection plantations.

Forest plantations in the area of transverse works, from torrential formations, cornice and with landslide areas, will have multiple functionality, besides the role of soil fixing in areas with high potential for landslide activation and environmental protection and can be used later as natural resources. These plantations also contribute and to brake leakage, maintenance of fertile soil in the perimeter, and cessation of landslide.

By carrying out the proposed works, the soil erosion process will be reduced, it will limit and reduce the areas affected by landslides, and the zone microclimate will also be modified.

REFERENCES

Barbu, N., Băcăuanu, V. (1977). *Evolution of the relief in the Moldavian plateau under the action of slope processes*. Proceedings of the French-Romanian.

Băcăuanu, V. (1968). *Câmpia Moldovei - studiu geomorfologic*. Ed. Acad. R.S.R., Bucureşti.

Băcăuanu, V. (1973). Evoluţia văilor din Podişul Moldovenesc. *Realizări în geografia României,* Ed. Ştiinţifică, Bucureşti.

Băcăuanu, V., Barbu, N., Pantazică, M., Ungureanu, Al., Chiriac, D. (1980). *Moldavian Plateau*. Nature, Man, Economy. Bucharest, Scientific and Encyclopedic.

Florea, N. (1997). *Degradarea terenurilor şi ameliorarea solurilor*, Universitatea Creştină Dimitrie Cantemir – Bucureşti, Facultatea de Geografie-Turism, Sibiu.

Heede, B.H. (1980). Gully Erosion - A Soil Failure: Possibilities and Limits of Control, *Int. Symposium, sept. 8-12, Bad Ischl, Austria, 1*, 317–330.

Hilborn, D., Stone, R.P. (1999). *Gully Erosion Control Agricultural Engineering Service*, Resources and Planning, Ontario Ministry of Agriculture, Food and rural Affairs (OMAFRA), Queen's Printer for Ontario.

Ioniță, I. (2000). *The relief of cuestas within the Moldavian Plateau.* Corson Publishing House, Iași, 109.

Marinescu, C. (1988). *Asigurarea stabilităţii terasamentelor şi versanţilor,* vol. I şi II, Ed. Tehnică, Bucureşti.

Mihai, Gh., Taloescu, I., Neguţ, N. (1979). Influenţa lucrărilor transversale asupra evoluţiei ravenelor formate pe alternanţe de orizonturi permeabile şi impermeabile. *Bul. Inf. ASAS, 8*, 103–105, Bucureşti.

Moţoc, M. (1963). *Eroziunea solului pe terenurile agricole şi combaterea ei*. Ed. Agrosilvică, Bucureşti.

Motoc, M., Munteanu, S., Baloiu, V., Stanescu, P., Mihai, Gh. (1975). *Eroziunea solului si metodele de combatere,* Ed. Ceres, Bucuresti.

Moţoc, M. (1975). *Combatera eroziunii solului*, Reeditare curs, Fac. Îmbunătăţiri funciare, I.A. "N. Bălcescu", Bucureşti.

Moţoc, M., Ouatu, O. (1977). *Rezultate preliminare privind încărcarea cu material solid a microcurenţilor de la suprafaţa versanţilor cu culturi agricole.* S.C.C.C.E.S. Perieni, vol. "Folosirea raţională a terenurilor erodate", 27–36.

Popa, N., Nistor, D., Nistor, D. (2005). *Amenajarea și Exploatarea terenurilor agricole degradate prin eroziune,* Ghid practic, Tipografia Moldova, Iaşi.

Pujină, D. (2008). *Landslides within the Moldavian Plateau.* Performantica Publishing House, Iași, 145 (In Romanian).

*** Soil surveys, at 1:10,000 and 1:200 000 scale, carried out by O.S.P.A. Iași.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EFFECT OF ORGANIC AND INORGANIC FERTILIZATION ON YIELD AND YIELD COMPONENTS OF TEFF [*Eragrostis tef* (Zucc.) Trotter] CULTIVATED UNDER MEDITERRANEAN SEMI-ARID CONDITIONS

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Abstract

Teff [Eragrostis tef (Zucc.) Trotter] has the potential to be one of those crops that have gained the interest of people of the Western world due to their beneficial health effects. The purpose of the present study was to evaluate the effect of organic and inorganic fertilization on yield and yield components of teff crop. A field experiment was laid out according to a randomized complete block design, with three replicates and three fertilization treatments [untreated, organic fertilizer (Bokashi) and inorganic fertilizer (34.5-0-0)]. The results showed that the higher plant height (85.9 cm), panicle length (33.4 cm), number of branches per panicle (9.3), number of grains per panicle (815.6), grain yield (1652 kg ha^{-1}) and straw yield (3793 kg ha^{-1}) were achieved through the application of inorganic fertilizer; however, the differences between the organic and inorganic fertilization were not statistically significant. The number of panicles per plant (9.6-10.4) and thousand grain weight (0.316-0.327 g) were not affected by fertilization. Finally, the results indicated that the organic fertilizer (Bokashi) should be considered as an alternative to chemical fertilizers for teff production.

***Key words**: Bokashi, grain yield, nitrogen fertilizer, teff, yield components.*

INTRODUCTION

Consumer's health consciousness has exhibited continued growth over the last two decades and the majority of population is trying to improve its health by consuming proper diet, containing foods which are not intended only to appease hunger and provide necessary nutrients for humans, but also to prevent nutrition-related diseases and improve mental and physical health of the consumers (Menrad, 2003). Cereals are getting appreciation from consumers and nutritionists, as they constitute the major source of carbohydrates and energy in diets worldwide. In addition, most of the cereals also contribute to the intake of a fair amount of dietary fiber, as well as, several vitamins, trace minerals, and phytochemicals (Poutanen et al., 2014). Wholegrain foods are recognized as an essential part of a healthy diet and many medical and epidemiological studies during the past 15 years have shown that higher consumptions of whole grains are strongly connected with reduced risk of acute and chronic diseases, including cardiovascular disease, type 2 diabetes, coronary heart disease, obesity, and certain cancers, including colorectal cancer (Aune et al., 2016).

Nowadays, the term "super-grain" has been become popular, and fewer cereals and pseudo-cereals are joined in this group. Some of these cereals like teff are endemic to limited to some parts of the globe but recent times due to their proven health beneficial effects, cultivation and consumption of them are spreading from origin to the different countries (Poutanen et al., 2014). In the case of the teff now it is gaining the interest of people particularly in the Western world and serious efforts are made to expand its cultivation in Europe and America (Belay et al., 2009). It has to be noted that a discussion has already been started in the scientific media as teff is the next super grain to replace quinoa (Satheess & Fanta, 2018).

Teff [*Eragrostis tef* (Zuccagni) Trotter] is a warm season C_4 annual plant that is a member of the Poaceae family, and it has been cultivated in the Horn of Africa for at least 3,000 years (Stallknecht et al., 1993). It is the major food crop in Ethiopia where it is annually cultivated on more than three million hectares of land (CSA, 2014). Compared to other cereals, teff can adapt to a wide range of agroecological conditions, such as marginal water-logged soils to drought conditions. Moreover, unlike other cereals, teff grain is less prone to attacks by storage pests and, thus, it can be safely stored under traditional storage conditions with no chemical protection and without losing its viability (Ketema, 1997).

Teff seeds are very minute (thousand grain weight = 0.18-0.38 g; thousand grain weight of *Arabidopsis* = 0.17-0.21 g) and, for marketing purposes, they are classified on the basis of outer caryopsis color: *netch* (white), *qey* (red/brown) and *sergegna* (mixed) (Tefera et al., 1995). The height of the plant ranges from 20 to 155 cm with the culm (11-72 cm) and the panicle (10-65 cm) accounting for about 47-65% and 35-53% of the total aboveground height, respectively (Assefa et al., 2001).

The importance of teff is mainly due to the fact that it has an exceptional nutritional profile and has additional health benefits including that its grains are free from the gluten and gluten-like proteins contained in other common cereals, such as wheat, rye, barley, and corn (Spaenij-Dekking et al., 2005). The increasing demand for gluten-free products is derived from the growing number of people who are diagnosed with celiac disease and other types of gluten sensitivity (Bultosa & Taylor, 2004; Spaenij-Dekking et al., 2005). Teff is a rich source of protein (9.4-13.3%) with a balanced essential amino-acid spectrum of leucine, valine, proline, alanine, and glutamic and aspartic acids. Studies showed that like other cereals teff is predominantly starchy (73%), as the starch content of teff is higher than that of most other cereals (Bultosa & Taylor, 2004). Teff starch granules consist of conglomerates of many polygonal simple granules (Helbing, 2009). Furthermore, it contains 2.6-3.0% ash and 2.0-3.1% lipid and constitutes a rich source of Fe, Ca, Zn, Mg than other cereal grains (Bultosa & Taylor, 2004). In a recent study, it was observed that the bio-available iron content was significantly higher in tef bread than in wheat bread (Alaunyte et al., 2012). Forsido et al. (2013) reported that the antioxidant properties of teff could be used for producing healthy food products.

Despite its versatility under various extreme environmental conditions, the productivity of teff is low with the yield standing at 1500 kg ha^{-1} in Ethiopia (CSA, 2014). The most important yield limiting factor is lodging. Under natural conditions, grain yield losses due to lodging are estimated at an average of 17%. (Assefa et al., 2011). In addition, lodging can reduce the quality of the seed in terms of germination capacity and energy, color and nutritional value (Assefa et al., 2015). However, farmers using improved cultivars and management practices can obtain yields up until 2500 kg ha^{-1} (Tefera & Belay, 2006).

Low soil fertility is intensified by soil fertility depletion through nutrient removal with harvest, tillage, weeding, and losses in runoff and soil erosion (Gebregziabher et al., 2006). Lower tef grain yield is mainly attributed to low soil fertility, and especially, nitrogen and phosphorus deficiencies (Kebede & Yamoah, 2009). According to several studies, grain yield exhibited a linear increase with the increasing rate of application of nitrogen fertilizer in teff (Giday et al., 2014; Assefa et al., 2016).

Since teff is a crop that has been systematically studied only during the last two decades, the available literature on plant growth and yield under organic fertilization is still quite limited. Substantial studies have shown the beneficial effects of organic fertilization on the yield and quality of several crops (Bilalis et al., 2012; 2018).

Bokashi is a soil fertilizer of Japanese origin made by fermenting organic wastes with a microbial inoculant. It is widespread to farming communities in Nicaragua, Mexico, and China. The use of Bokashi has even extended to developed countries, such as USA, where the microbial inoculant Effective Microorganisms or EM, a consortium of lactic acid bacteria, photosynthetic bacteria and yeasts, is used. A study conducted in China showed that application of EM bokashi significantly increased grain yields, nutrient content in straw and grain, and straw biomass in wheat (Hu & Qi, 2013).

Literature survey revealed that there was a lack of knowledge the performance of teff growth under Mediterranean semi-arid conditions and organic cropping system. Therefore, the aim of this study was to determine the effects of organic and inorganic fertilization on yield and yield components of teff crop.

MATERIALS AND METHODS

A teff crop was established in the organic experimental field of the Agricultural University of Athens (Latitude: 37°59′ 1.70″ N, Longitude: 23°42′ 7.04″ E, Altitude: 29 m above sea level) from April to August 2015. The soil was a Leptosol and soil texture was clay loam (29.8% clay, 34.3% silt and 35.9% sand) with pH (1:1 H_2O) 7.29, nitrate-nitrogen (NO_3-N) 12.4 mg kg^{-1} soil, available phosphorus (P) 13.2 mg kg^{-1} soil, available potassium (K) 201 mg kg^{-1} soil, 15.99% $CaCO_3$ and 1.47% organic matter.

The site was managed according to organic agricultural guidelines (EC 834/2007). Weather data (mean monthly air temperature and precipitation) pertaining to the experimental period were recorded by the weather station of Agricultural University of Athens and are presented in Table 1.

The mean temperature during the growing season (April-August) was higher (24.2°C) as compared to 30-year average (23.4°C).

The total rainfall of cultivation period 57.8 mm, which means it was the three-fourths of the 35 years average (75.9 mm).

Table 1. Monthly means of maximum (Max.), minimum (Min.) and average (Avg.) air temperature and precipitation for 2015 growing season and the 30 years average in Athens, Greece

Month	Max (°C)	Min (°C)	Avg. (°C)	Precipitation (mm)	Max (°C)	Min (°C)	Avg. (°C)	Precipitation (mm)
	2015				30 year average			
April	24.5	5.3	16.1	8.8	20.2	9.6	15.3	30.8
May	31.3	14.5	21.8	31.8	26.0	13.9	20.7	22.7
June	35.1	16.8	24.7	15.8	31.1	18.2	25.6	10.6
July	37.2	19.4	29.3	0.8	33.5	20.8	28.0	5.8
August	36.1	21.6	29.3	0.4	33.2	20.7	27.4	6.0
Total	-	-	-	57.6	-	-	-	75.9

The experiment was set up on an area of 238 m^2 according to a randomized complete block design (RCBD), with three fertilization treatments: control (untreated), organic fertilizer (EMIKO® Bokashi Organic NPK Fertilizer with EM, 1.06% N, EMIKO Handelsgesellschaft mbH) at a rate of 4200 kg ha^{-1} and inorganic fertilizer (Nutramon® 34.5-0-0, Hellagrolip S.A.) at a rate of 60 kg N ha^{-1}, and three replications for each treatment. The plot size was 20 m^2 (5 m x 4 m). There was a space of 1 m between plots and 1 m between replications. Soil was prepared by ploughing at a depth of about 0.25 m. Fertilizers were applied by hand on the soil surface and then harrowed in. Teff seeds were sown on 19th April by hand in rows 30 cm apart, at a rate of 5 kg ha^{-1} and a depth of 1 cm. Overhead sprinkler system was also set up on the field. The field area was irrigated 5 times. The total quantity of water applied during the experimental period was 328 mm. Throughout the experimental period, there was no incidence of pest or disease on teff crop. Weeds were controlled by hand hoeing when it was necessary.

Data were recorded on some growth and yield parameters including plant weight, number of panicles per plant, plant length and number of branches per panicle using five randomly selected plants in each plot at 100 days after sowing (DAS). Moreover, thousand grain weight, grain yield, and straw yield were assessed at 120 DAS. Thousand grain weight was counted on five randomly selected plants from each plot. For the determination of grain yield and straw yield, the middle sub-plot area of 4 m^2 (2 m x 2 m) was used. Biological yield was recorded by measuring the whole weight of plants from middle sub-plot areas after harvesting and drying for three days. Grain yield was determined by measuring the grain weight of plants used for straw yield.

The experimental data were checked for normality and subjected to statistical analysis according to the split-plot design. The statistical analysis was performed with the SigmaPlot 12 statistical software (Systat Software Inc., San Jose, CA, USA).

Differences between means were separated using Least Significance Difference (LSD) test. Correlation analyses were used to describe the relationships between yield and yield components using Pearson's correlation. All comparisons were made at the 5% level of significance.

RESULTS AND DISCUSSIONS

The effects of organic and inorganic fertilization on the plant height of teff are presented in Table 2. Regardless of the type of fertilizers, the teff plants in the plots received inorganic or organic fertilizer were significantly taller as compared to their respective plant heights in the untreated plots. In particular, the higher plant height (85.92 cm) was recorded in inorganic fertilization treatment followed by organic fertilization (82.44 cm); however, the differences among these values were not statistically significant. Plant height affects above-ground biomass yield of a crop. In earlier studies, nitrogen significance to teff's productivity has been described (Tesfahunegn, 2014; Assefa et al., 2016). Giday et al. (2014) reported that plant height of teff increased consistently with increasing rates of nitrogen and the maximum plant height of 122.33 cm was found in plants treated with 69 kg N ha^{-1} of slow release urea fertilizer. In contrast to this, Berhane et al. (2013) reported superior plant height of teff in plots received organic fertilizers as compared to conventional once. The values of our results are lower as compared to other studies, probably due to the different variations in environmental conditions and the genetic potential of populations (Roussis et al., 2017).

Table 2. Plant height, number of panicles per plant, panicle length, number of branches per panicle of teff crop as affected by organic and inorganic fertilization

Fertilization	Plant Height (cm)	Number of panicles per plant	Panicle length (cm)	Number of branches per panicle
Control	74.86 b	9.6 a	27.04 b	7.6 b
Organic	82.44 a	10.2 a	31.09 ab	8.7 a
Inorganic	85.92 a	10.4 a	33.52 a	9.3 a
$F_{fertilization}$	10.358*	3.087^{ns}	8.518*	20.265**
p-value	0.026	0.155	0.036	0.008

F-test ratios are from ANOVA. Significant at * and ** indicate significance at p ≤ 0.05 and 0.01, respectively and ns: not significant. Mean values within each column followed by different letters, differ significantly according the *LSD* test (p ≤ 0.05).

Concerning the number of panicles per plant, the effect of fertilization was found not to be statistically significant (Table 2). Despite the non-significant differences among the fertilization treatments, the highest value (10.4) was found in plants treated with inorganic fertilizer and the lowest (9.6) in the unfertilized plots. Giday et al. (2014) reported the positive and significant increase in the number of panicles per plant with the increasing rate of nitrogen fertilizer on teff. In the same way, Asefa et al. (2014) found that number of panicles was significantly increased with the increase of rate of NPK fertilizers. In contrast to above-mentioned studies, Assefa et al. (2016) observed no difference in the number of panicles per plant between the levels of compost and NP fertilizer, and the values did not present any increasing or decreasing trend. In a similar way, Tesfahunegn (2014) found that the unfertilized plot showed a significantly lower panicle number per plant as compared to others, but there were non-significant differences among the remaining which received different rates of fertilizers.

Panicle length constitutes one of the yield attributes of teff that could lead to a high increment of grain and straw yield. In our study, panicle length was significantly affected by the different fertilization treatments, with the values significantly varied between the inorganic fertilization and control (Table 2). The highest mean length (33.52 cm) was recorded in inorganic fertilization treatment, while, the lowest (27.04 cm) was obtained from the untreated plot. This may also be due to the same reason as that of plant height. The positive correlation between nitrogen fertilizer and panicle length of teff has also been reported by several authors (Giday et al., 2014; Assefa et al., 2016). The application of balanced fertilizer and efficient utilization of nutrients leads to high photosynthetic productivity and accumulation of high dry matter, resulting in increases of panicle length and grain yield (Assefa et al., 2016).

The results of the experiment indicated that the effect of organic and inorganic fertilization significantly affected the number of branches per panicle of teff (Table 2). This trait was significantly varied between the inorganic fertilization and control, and the organic fertilization and control. But, the number of branches per panicle did not significantly differ between the organic and inorganic fertilization, indicating that both treatments did show similar effect on the parameter. The highest mean branches number per panicle was observed in the inorganic treatment (9.3) and the lowest in the control (7.6). Studies have shown that nitrogen fertilizer application had a significant effect on the number of branches and spikelets in panicle (Matsui & Kagata, 2002; Kamiji et al., 2011). The panicle length and the number of panicle branches are important predictors of spikelets. Zhou et al. (2017) reported that the increase in panicle length and number of branches provided more space for spikelets development and increased spikelet density per panicle under optimized nitrogen fertilizer application.

Table 3. Thousand grain weight, grain yield and straw yield of teff crop as affected by organic and inorganic fertilization

Fertilization	Thousand grain weight (g)	Grain yield (kg ha^{-1})	Straw yield (kg ha^{-1})
Control	0.316 a	1287 b	3404 b
Organic	0.323 a	1535 a	3612 ab
Inorganic	0.327 a	1652 a	3793 a
$F_{fertilization}$	2.968^{ns}	27.431**	9.019*
p-value	0.162	0.004	0.033

F-test ratios are from ANOVA. Significant at * and ** indicate significance at $p \leq 0.05$ and 0.01, respectively and ns: not significant. Mean values within each column followed by different letters, differ significantly according the *LSD* test ($p \leq 0.05$).

Regarding the thousand grain weight, the analysis of variance revealed that the fertilization had no significant effect on this parameter (Table 3). However, the maximum thousand grain weight (0.327 g) was recorded after the application of inorganic fertilizer and followed by organic treatment (0.323 g). Gooding and Davies (1997) observed neither improvement nor reduced thousand kernel weight due to nitrogen fertilization even when yields increased. In general, a wide range of factors, including variety, growing conditions, climatic factors, and soil properties affect the thousand grain weight.

Grain yield was significantly responded to the fertilization (Table 3). The highest mean grain yield of teff (1652 kg ha^{-1}) was obtained in the inorganic fertilization treatment with 365 kg ha^{-1} yield advantage over the control. The next highest mean grain yield (1535 kg ha^{-1}) was obtained from the organic fertilization treatment with no statistically significant difference ($p>0.05$) compared to the yield obtained with the application of inorganic fertilizer, and the lowest grain yield (1287 kg ha^{-1}) was obtained from the untreated plot. Several studies indicated the positive and linear response of grain yield to increasing levels of nitrogen fertilizer (Geleto et al., 1995; Giday et al., 2014). Geleto et al. (1995) reported that application of 120 kg N ha^{-1} presented yield advantage ranging from 19 to 49% over the yield obtained with the application of 60 kg N ha^{-1} depending on the inherent nitrogen status of the soil as well as the amount and distribution of precipitation during the cultivation period of the respective locations. Assefa et al. (2016) also reported that the application of 2.5 tons ha^{-1} compost combined with the 50% of recommended NP (64/46 kg ha^{-1} N/P_2O_5) rate resulted in grain yield increments of 158% compared to the control (untreated) on teff which is comparable to the full NP dose. Compost could also improve the soil structure which leads to better root development that may result in more nutrient up take from the soil in addition to its slow and gradual release of macro and micro nutrients by itself (Al-Bataina et al., 2016). Grain yield had positive and significant correlation with plant height, number of panicles per plant, panicle length, number of branches per panicle and straw yield (*r* = 0.946, *p*<0.001; *r* = 0.702, *p* = 0.034; *r* = 0.799, *p* = 0.009; *r* = 0.856, *p* = 0.003 and *r* = 0.768, *p* = 0.016, respectively).

The application of different fertilization treatments had a significant effect on straw yield of teff. The highest mean straw yield (3793 kg ha^{-1}) was obtained in the inorganic fertilization treatment with 389 kg ha^{-1} yield advantage over the control plot. The next highest mean straw yield (3612 kg ha^{-1}) was obtained from the organic fertilization treatment with no statistically significant difference (p>0.05) compared to the yields obtained from the inorganic fertilization and untreated plots. The lowest straw yield (3404 kg ha^{-1}) was obtained from the untreated plot. The increase in nitrogen supplement can result in a more vigorous plant growth, producing a greater total plant biomass and a higher straw yield (Giday et al., 2014). Assefa et al. (2016) observed that application of compost beyond 5 tons ha^{-1} compost combined with NP beyond 32/23 kg ha^{-1} N/P_2O_5 rate had no significant effect on straw yield. Contrariwise, Berhane et al. (2013) found that the organic fertilizer increased straw yield of teff than the inorganic fertilizer and reasoned that could be due to higher plant height of organic teff than the conventional. Straw yield had positive and significant correlation with plant height, panicle length and number of branches per panicle (r = 0.716, p = 0.029; r = 0.801, p = 0.009 and r = 0.8831, p = 0.002, respectively).

CONCLUSIONS

The results showed that the higher plant height, panicle length, number of branches per panicle, number of grains per panicle, grain yield and straw yield were achieved through the application of inorganic fertilizer; however, the differences between the organic and inorganic fertilization were not statistically significant. The number of panicles per plant and thousand grain weight were not affected by fertilization. As a conclusion, the results of the present study indicated that the organic fertilizer (Bokashi) should be considered as an alternative to chemical fertilizers for teff production.

REFERENCES

Al-Bataina, B. B., Young, T. M., Ranieri, E. (2016). Effects of compost age on the release of nutrients. *International Soil and Water Conservation Research*, *4*, 230–236.

Alaunyte, I., Stojceska, V., Plunkett, A., Ainsworth, P., Derbyshire, E. (2012). Improving the quality of nutrient-rich teff (*Eragrostis tef*) breads by combination of enzymes in straight dough and sourdough breadmaking. *Journal of Cereal Science*, *55*, 22–30.

Asefa, F., Debela, A., Mohammed, M. (2014) Evaluation of teff [*Eragrostis tef* (Zuccagni) Trotter] responses to different rates of NPK along with Zn and B in Didessa district, southwestern Ehiopia. *World Applied Sciences Journal, 32*, 2245–2249.

Assefa, A., Tana, T., Abdulahi, J. (2016). Effects of compost and inorganic NP rates on growth, yield and yield Components of teff [*Eragrotis teff* (Zucc.) Trotter] in Girar Jarso District, Central Highland of Ethiopia. *Journal of Fertilizers and Pesticides*, *7*, 1000174.

Assefa, K., Cannarozzi, G., Girma, D., Kamies, R., Chanyalew, S., Plaza-Wüthrich, S., Blösch, R., Rindisbacher, A., Rafudeen, S., Tadele, Z. (2015). Genetic diversity in tef [*Eragrostis tef* (Zucc.) Trotter]. *Frontiers in Plant Science*, *6*, 177.

Assefa, K., Ketema, S., Tefera, H., Hundera, F., Kefyalew, T. (2001). Genetic diversity for agronomic traits in tef. In H. Tefera, G. Belay, & M. Sorrells (Eds), *Narrowing the Rift* (pp. 33-48). Proceedings of the International Workshop on Tef Genetics and Improvement, 16-19 October 2000. Debre Zeit, Ethiopia: Tef Research and Development.

Assefa, K., Yu, J. K., Zeid, M., Belay, G., Tefera, H., Sorrells, M. E. (2011). Breeding tef [*Eragrostis tef* (Zucc.) trotter]: conventional and molecular approaches. *Plant Breeding, 130*, 1–9.

Aune, D., Keum, N., Giovannucci, E., Fadnes, L. T., Boffetta, P., Greenwood, D. C., Tonstad, S., Vatten, L. J., Riboli, E., Norat, T. (2016). Whole grain consumption and risk of cardiovascular disease, cancer, and all cause and cause specific mortality: Systematic review and dose-response meta-analysis of prospective studies. *The Biomedical Journal*, *353*, 1–14.

Belay, G., Zemede, A., Assefa, K., Metaferia, G., Tefera, H. (2009). Seed size effect on grain weight and agronomic performance of tef [*Eragrostis tef* (Zucc.) Trotter]. *African Journal of Agricultural Research*, *4*, 836–839.

Berhane, M., Kebede, F., Fitiwy, I., Abreha, Z. (2013). Comparative productivity and profitability of organic and conventional teff [*Eragrostis tef* (Zucc.) Trotter] production under rainfed condition: Tigray, Northern Ethiopia. *World Journal of Agricultural Sciences*, *1*, 303–311.

Bilalis, D., Kakabouki, I., Karkanis, A., Travlos, I., Triantafyllidis, V., Hela, D. (2012). Seed and saponin production of organic quinoa (*Chenopodium quinoa* Willd.) for different tillage and fertilization. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, *40*, 42–46.

Bilalis, D., Krokida, M., Roussis, I., Papastylianou, P., Travlos, I., Cheimona, N., Dede, A. (2018). Effects of organic and inorganic fertilization on yield and quality of pro-cessing tomato (*Lycopersicon esculentum* Mill.). *Folia Horticulturae*, *30*, 321–333.

Bultosa, G., & Taylor, J.R.N. (2004a). Teff. In C. Wrigley, H. Corke, & C. Walker (Eds.), *Encyclopedia of Grain Science* (pp. 20–36). Oxford, England: Elsevier.

CSA. (2014). *Agricultural Sample Survey for 2013/14. Statistical Bulletin 532*. Addis Ababa, Ethiopia: Central Statistical Agency.

Forsido, S.F., Rupasinghe, H.P., Astatkie, T. (2013). Antioxidant capacity, total phenolics and nutritional content in selected Ethiopian staple food ingredients. *International Journal of Food Sciences and Nutrition*, *64*, 915–920.

Gebregziabher, S., Mouazen, A.M., Van Brussel, H., Ramon, H., Nyssen, J., Verplancke, H., Behailu, M., Deckers, J., De Baerdemaeker, J. (2006). Animal drawn tillage, the Ethiopian ard plough, *maresha*: A review. *Soil and Tillage Research*, *89*, 129–143.

Geleto, T., Tanner, D. G., Mamo, T., Gebeyehu, G. (1995). Response of rainfed bread and durum wheat to source, level, and timing of nitrogen fertilizer on two ethiopian vertisols. I. Yield and yield components. *Communications in Soil Science and Plant Analysis*, *26*, 1773–1794.

Giday, O., Gibrekidan, H., Berhe, T. (2014). Response of teff (*Eragrostis tef*) to different rates of slow release and conventional urea fertilizers in vertisols of southern Tigray, Ethiopia. *Advances in Plants and Agriculture Research, 1*, 190–197.

Gooding, M.J., & Davies, W.P. (1997). *Wheat production and utilization system, quality and environment*. USA: CAB International.

Helbing, J. (2009). *Konfokale laser scanning-mikroskopie und raster- elektronenmikroskopie zur beobachtung der mälzung verschiedener zerealien und pseudozerealien*. München, Germany: Technische Universität München.

Hu, C., & Qi, Y. (2013). Long-term effective microorganisms application promote growth and increase yields and nutrition of wheat in China. *European Journal of Agronomy*, *46*, 63–67.

Kamiji, Y., Yoshida, H., Palta, J.A., Sakuratani, T., Shiraiwa, T. (2011). N applications that increase plant N during panicle development arehighly effective in increasing spikelet number in rice. *Field Crops Research, 122*, 242–247.

Kebede, F., & Yamoah, C. (2009). Soil fertility status and NuMaSS fertilizer recommendation of typic hapluusterts in the northern highlands of Ethiopia. *World Applied Sciences Journal*, *6*, 1473–1480.

Ketema, S. (1997). *Tef, Eragrostis tef (Zucc.) Trotter*. Rome: Institute of Plant Genetics and Crop Plant Research, Gatersleben/ International Plant Genetic Resources Institute.

Matsui, T., & Kagata, H. (2002). Correlation of nitrogen concentration with dry-matter portioning to spikelets and total husk volume on the panicle in japonica rice. *Plant Production Science*, *5*, 198–202.

Menrad, K. (2003). Market and marketing of functional food in Europe. *Journal of Food Engineering*, *56*, 181-188.

Poutanen, K., Sozer, N., Della Valle, G. (2014). How can technology help to deliver more of grain in cereal foods for a healthy diet? *Journal of Cereal Science*, *59*, 327–336.

Roussis, I., Travlos, I., Bilalis, D., Kakabouki, I. (2017). Influence of seed rate and fertilization on yield and yield components of *Nigella sativa* L. cultivated under Mediterranean semi-arid conditions. *Agrolife Scientific Journal*, *6*(1), 218–223.

Satheesh, N., & Fanta, S.W. (2018). Review on structural, nutritional and anti-nutritional composition of Teff (*Eragrostis tef*) in comparison with Quinoa (*Chenopodium quinoa* Willd.). *Cogent Food and Agriculture*, *4*, 1546942.

Spaenij-Dekking, L., Kooy-Winkelaar, F., Koning, F. (2005). The Ethiopian cereal tef in celiac disease. *The New England Journal of Medicine*, *353*, 1748–1749.

Stallknecht, G.F., Gilbertson, K. M., Eckhoff, J.L. (1993). Teff: food crop for humans and animals. In: J. Janick, & J. E. Simon (Eds). *New crops* (pp. 231–234). New York, USA: Wiley.

Tefera, H., Belay, G. (2006). *Eragrostis tef* (Zuccagni) Trotter. [Internet] Record from Protabase. In: M. Brink, & G. Belay (Eds) *PROTA (Plant Resources of Tropical Africa/ Ressources végétales de l'Afrique tropicale*). Wageningen, Netherlands. https://www.prota4u.org (February, 2018).

Tefera, H., Ayele, M., Assefa, K. (1995). *Improved varieties of tef (Eragrostis tef) in Ethiopia, releases of 1970-1995. Researches bulletin, No. 1.* Debre Zeit, Ethiopia: Debre Zeit Agricultural Research Center, Alemaya University of Agriculture.

Tesfahunegn, G. B. (2014). Response of yield and yield components of tef [*Eragrostis tef* (Zucc.) Trotter] to tillage, nutrient, and weed management practices in Dura Area, Northern Ethiopia. Hindawi Publishing Corporation International Scholarly Research Notices. ID 439718.

Zhou, W., Lv, T., Yang, Z., Wang, T., Fu, Y., Chen, Y., Hu, B., Ren, W. (2017). Morphophysiological mechanism of rice yield increase in response to optimized nitrogen management. Scientific Reports, 7, 17226.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EVALUATION AND SPECIFICATION OF NATURAL FACTORS OF THE IALOVENI DISTRICT IN THE REPUBLIC OF MOLDOVA

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Abstract

The Republic of Moldova is characterized by temperate continental climate, mild and short winter, hot and long summer with a relatively small amount of atmospheric precipitation. The average annual rainfall volume in the North of Moldova is 550-600 mm. In the Center region, rainfall is predominantly low (500-550 mm), and in the South - very low (450-500 mm). The climatic zone determines the frequency of the drought phenomenon. In the North of the country, a severe drought is recorded every 10 years, in the Center area, 2-3 times, and in the South region, 3-4 times. With global climate change, the phenomenon of drought is more frequent. This creates a high moisture deficiency in soil and air, the consequences of which lead to a substantial decrease in crop productivity or even loss of crop. In the territory of Ialoveni district the pedoameliorative situation is very complex. As a result of the work carried out, new data were obtained regarding the current state of the natural factors influencing the eco-natural settlement within the studied territory. In order to appreciate the natural potential of Ialoveni district, protect and increase soil fertility, trace the ways of development of agricultural enterprises, and thus improve the socio-economic level of the rural population, it is necessary to study in detail the natural factors: climate, relief, surface waters and pedophrenatics, silvicultural arrangements, soil cover, and others.

Key words: *Digital Model - SIG, eco-natural settlement, Ialoveni district, natural factors, soil.*

INTRODUCTION

The rational use of soil resources is based on the detailed knowledge of the main natural and anthropogenic factors that influence effective fertility and their quality status. It is established that in recent decades such serious soil degradation has increased, such as water erosion, landslides, dehumidification and nutrient depletion, salinisation and secondary solonization as a result of irrigation, and so on.

In the last decades, the role of the anthropic factor has grown in erosion processes. An example of inappropriate human intervention and with extremely serious consequences on soil cover is agrarian reform carried out without scientific support. Thus, through the implementation of the "Earth" program, spraying (compacting) small-scale compact land, which excludes the possibility of organizing the territory and applying the anti-erosion technologies, occurred.

Concurrent assessment of land quality is necessary to justify a complex system of measures to prevent degradation and conservation of soil production capacity. For the development of complex measures to prevent soil degradation it is necessary to know the spatial spread and the intensity of the manifestation of these processes on a more detailed scale.

Achieving this goal in limited terms is possible with the use of the Geoinformatics System (GIS), which allows an operative and reasoned approach to inventory, analysis, planning and design issues. This system creates prerequisites for the development and implementation of measures to combat soil degradation at a new quality level aimed at maintaining and improving soil fertility.

The need to carry out these research results from the fragmentary and incomplete character of previous research. Concurrent assessment of land quality is necessary to justify a complex system of measures to prevent degradation and conservation of soil production capacity. The results of the works and the practical recommendations will be used for the elaboration of the irrigation, the erosion control, the planting of the fruit plantations, and the protection of the environment by afforestation and erosion of degraded lands (landslides, ravines etc.).

In the territory of Ialoveni district, the pedoameliorative situation is very complex. A special set of methodological procedures has been applied for its proper characterization. At their base are laid the principles of the physico-geophysical method (Gherasimov et al., 1960) and the conjugate analysis of the cartographic materials, the laboratory results and the data of the observations in nature with the application of the genetic methods of the system. As a result of the performed works, new data were obtained regarding the current state of the natural factors influencing the econo- mural situation within the studied territory.

The results of the works and the practical recommendations will be used for the elaboration of the projects for erosion control, irrigation systems development, establishment of fruit plantations, protection of the environment by afforestation and erosion of degraded lands (landslides, ravines, eroded etc.).

MATERIALS AND METHODS

The research methods were office, field, laboratory and office. Initial office work was carried out with the use of the informational geographic system, which allowed for the development of the digital model of the relief and the dimensioning of the land infrastructure. Fieldwork or focusing on updating land-based limestones and sampling of surface water samples. Determination of water quality properties and indices has been made by laboratory analysis with the application of classical methods and existing standards. The study objects were carried out within the Ialoveni district (25 communes) on an area of over 78 thousand ha.

The purpose of the works included in the stage - the evaluation of the geomorphological, infrastructure and hydrochemical factors for assessing the volume of the irrigation fund at the administrative level II. The evaluation of natural and anthropic parameters and the delimitation of boundaries between taxonomically subordinate soil units - is the most complex and important stage of research.

The mapping method used the comparative-geographic method of V.V. Dokuchaev. It helps to establish the natural relationships between soils and soil formation factors (Kashanskii, 1987). The most complete numerical form of factors determining soil properties at a certain point is reflected in the SCORPAN model (McBratney et al., 2003):

$$S = f * (S, C, O, R, P, A, N) \qquad (1)$$

where:

S - soil; C - climate; O - organisme; R - relief; P - parental rocks; A - age and N - territorial neighborhood.

It is assumed that the same combination of soil-forming factors (predictors) corresponds to one and the same soil.

In order to evaluate the changes in the natural and anthropic parameters obtained over the years the thematic materials elaborated previously were used. Therefore, they do not fully reflect (for objective reasons) the complexity of the situation on the ground at the moment.

This finding refers in particular to the forms of use (agricultural, non-agricultural, infrastructure), to the landscapes of the basins, where the pedogenic factors (parental rocks, hydrology, lithology, hydrogeology and vegetation) interact in various ways lead to the emergence of forms of extremely complex degradation (Rozloga, 2013; 2015).

Over the last years, the informational-geographic system has been used in the Republic of Moldova. High-resolution satellite imagery and photogrammetry sets appeared.

The use of the geographic information system (GIS) and the modern equipment in the mapping works took place within the territory of Ialoveni rayon. The research was carried out in 2015-2016 by the collaborators of the Pedological Geological Information System and Precision Agriculture Laboratory and the Soil Improvement and Protection Laboratory of the Institute of Pedology, Agrochemistry and Soil Protection “Nicolae Dimo”.

The following materials were used in the study:
- the maps of the organization of the territory of the Ialoveni rayons at 1: 10000 scale;
- the pedological maps from the Ialoveni rayons at 1: 10000 scale;
- topographical plan sheets at 1: 50000 scale;
- topographical plan sheets at 1: 10000 scale;

- digital map of the land cover of the Republic of Moldova at 1: 50000 scale developed in 2011;
- "Orto-Foto" spatial remote sensing materials at ARFC scale 1: 5000;
- Materials "Land Digital Model (DTM)" of the Land and Cadastre Agency.

These materials have been linked to the national reference system MoldRef-99. Subsequently, the thematic layers ("Hidro", "Roads", "Localities", "Forests", "Forest Strips", etc.) were formed. Also, the structure of the database parameters for the elaborated layers was formed. It was then proceeded to vector the contours of the land according to the categories and subcategories of use, updating them using the Orto-Foto materials, elaborating the digital model of the relief at the scale 1: 1000 and introducing the attribution information for each area. Field research has been carried out to update and correct infrastructure contour boundaries, land degradation forms digitally sized in the office from geospatial remote sensing materials. The work has been done in MapInfo (2D) and ArcGIS (3D).

The research was carried out in accordance with the "Guidelines for Land Surveys for Lands and State Grants" approved in 1991 (Filipciuc, 2007), "Regulation on the content of the land registry documentation" approved by the Government Decision of the Republic of Moldova No. 24 of 11 January 1995 (Arinushkina, 1963).

RESULTS AND DISCUSSIONS

In order to appreciate the natural potential of Ialoveni district, the protection and increase of soil fertility, the development of agricultural enterprises, and therefore the improvement of the socio-economic level of the rural population, it is necessary to study in detail the natural factors: climate, relief, surface waters and pedophrenatics, silvicultural arrangements, soil cover, etc.

Weather conditions. The Republic of Moldova is characterized by temperate continental climate, mild and short winter, hot and long summer with a relatively small amount of atmospheric precipitation. The average annual rainfall in the northern area of Moldova is 550-600 mm (Figure 1, Table 1). In the Center region, rainfall is predominantly low (500-550 mm), and in the South - very low (450-500 mm). The climatic zone determines the frequency of the drought phenomenon. In the North of the country, a severe drought is recorded every 10 years, in the Center area - 2-3 times and in the South region 3-4 times. With global climate change, the phenomenon of drought is more frequent.

This creates a high moisture deficiency in soil and air, the consequences of which lead to a substantial decrease in crop productivity or even crop loss.

The geographic location of the surveyed territory includes area II (72030 ha) and sub-area IIa (6239 ha) pedoclimateric with insufficient humidity (Ursu, 2011).

According to statistical data, the average annual air temperature for zone II is 9.0-9.5ºC and 8.5-9.0ºC for sub-area IIa; the sum of temperatures above 10ºC is 3000-3200ºC and 2900-3000ºC, correspondingly; the duration of the vegetation period with temperatures above 10°C extends over a period of 177-182 days. The cold period (with temperatures below 0°C) averages 174-188 days. Sun days have a duration of 290-320 per year (Table 1, Figure 1).

For the territory of the rayon the materials of the monthly climatic indices were processed during the period 1950-2016 (Agrometeorological Bulletin, 2011). The lowest average mean temperature was set in 1954, which was minus 11.9°C, and the maximum was found in 1992, this being 25.1°C (Figure 2). The average annual temperature is 9.79ºC.

All field and office materials have undergone a conjugate analysis and were studied in the system (Gherasimov et al., 1960). Subsequently, these were used to develop updated cartographic information.

The result of the geoinformation works was finalized with the elaboration of the set of digital thematic maps from the territory of the Ialoveni district. In Figure 3 shows the average monthly temperature over the years with climatic data. The diagram shows that the minimum average temperature during the years corresponds to January, minus 3.46° C, and the maximum - in the months of August to August, reaching the value of 20.4°C. For the normal development of metabolism processes between plants and soil, an optimal water regime is required. In the Republic of Moldova the level of water supply is quite variable over the years; from normal to critical crop development.

Table 1. Characterization of the main agro-climatic indices

Indices	Zone, subzone				
	I	Ia	II	IIa	III
Surface, ha	769912.40	224822.73	1359714.01	124398.72	901282.61
Altitude, m	100-300	200-300	50-200	200-400	10-200
Solar days	290-300	280-290	310-320	290-300	310-320
Hours of sunshine	2050-2100	2000-2050	2100-2200	2100-2150	2200-2300
T° annual average	8.0-8.5	7.0-8.0	9.0-9.5	8.5-9.0	9.5-10.0
$\sum$ T° > 10°	2750-3000	2750-2800	3000-3200	2900-3000	3200-3450
$\sum$ annual rainfall, mm	550-600	550-630	500-550	550-600	450-550
K, h	0.65-0.8	0.7-0.8	0.6-0.65	0.7-0.8	0.5-0.6
No. of drought in 10 years	1-2	1	2-3	1-2	3-4
Vegetation period, days	167-176	166-167	177-182	178-182	179-187
The period of the angels, days	163-179	163-179	174-189	175-188	175-196

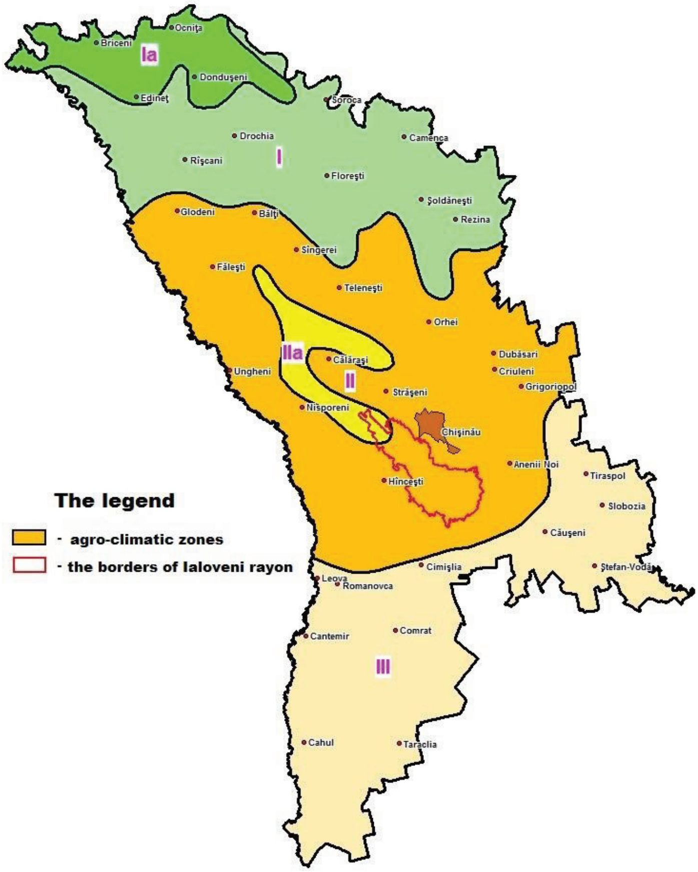

Figure 1. Agro-climatic zones of the Republic of Moldova

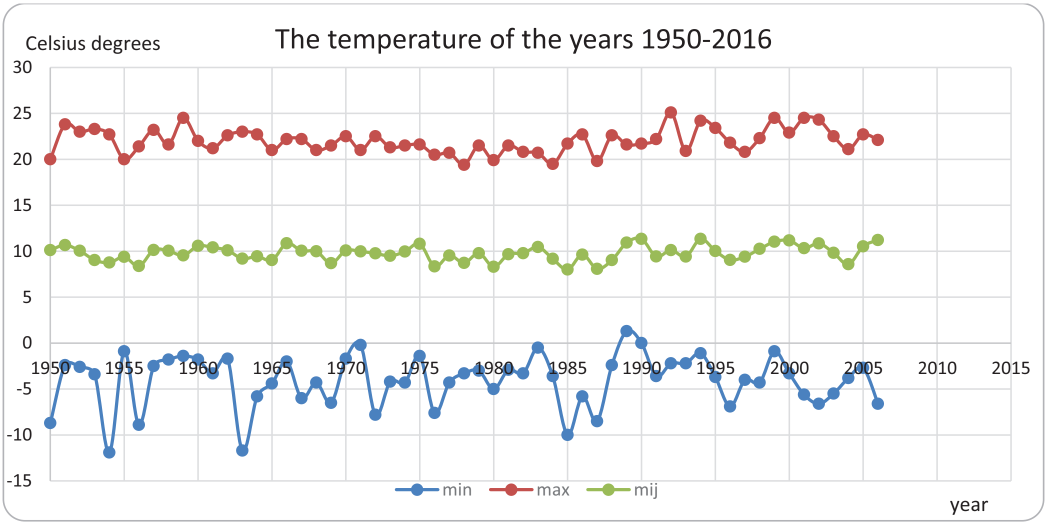

Figure 2. Diagram of annual average temperatures

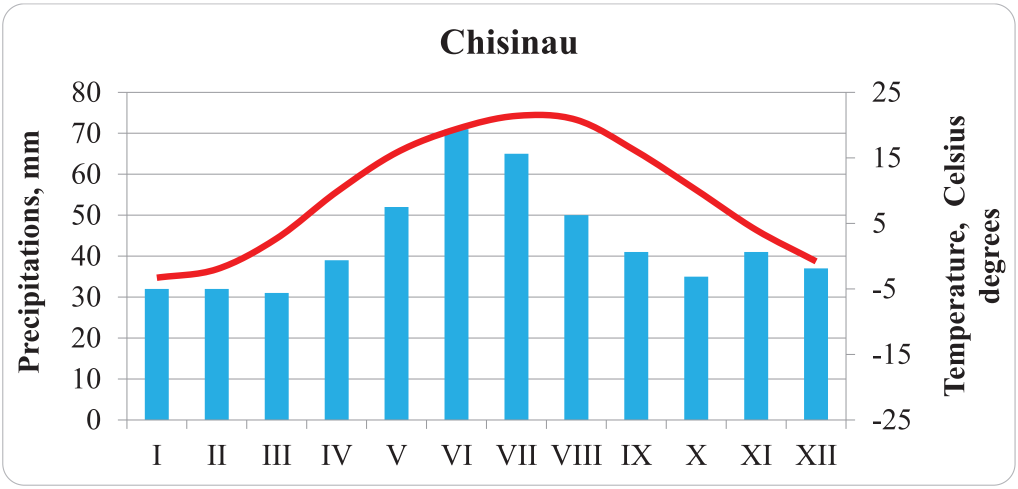

Figure 3. Diagram of atmospheric deposits and annual average temperatures over months

According to the multiannual data on the surveyed area the annual average precipitation is 500-600 mm, and during the period with temperatures above 10°C the atmospheric deposits are 380-400 mm. The number of droughts in 10 years alternates from 1 to 3 times. The value of the hydrothermal coefficient is 0.6 - 0.8, which results in insufficient humidity during the vegetation period of the plants.

The processing of climatic data between 1950 and 2016 shows that the average monthly average atmospheric deposition was detected in June 1952 and consisted of 230 mm. The minimum value was recorded in October 1953, 1969 by 1 mm. The average monthly average of 57-64 years is 46.3 mm (Figure 4).

During 1950-2016 atmospheric depositions had an uneven trend. The amount of prizes varied from 345 mm in 1951 to 774 mm in 1966. The annual average is 555 mm (Figure 5).

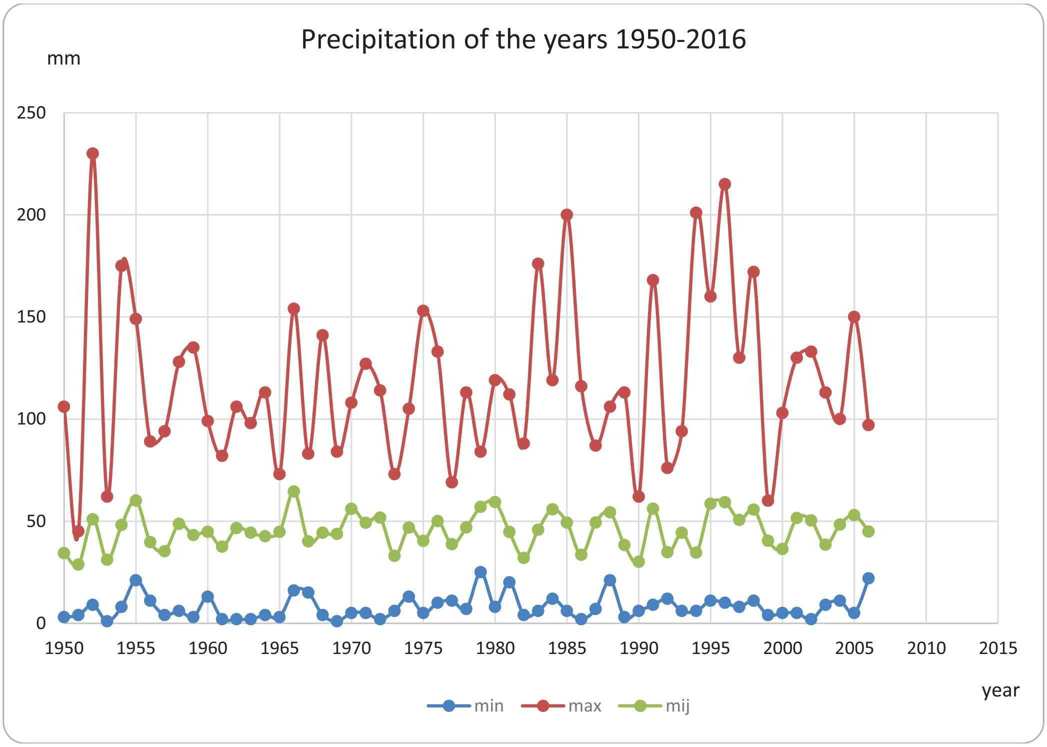

Figure 4. Diagram of annual average atmospheric deposition

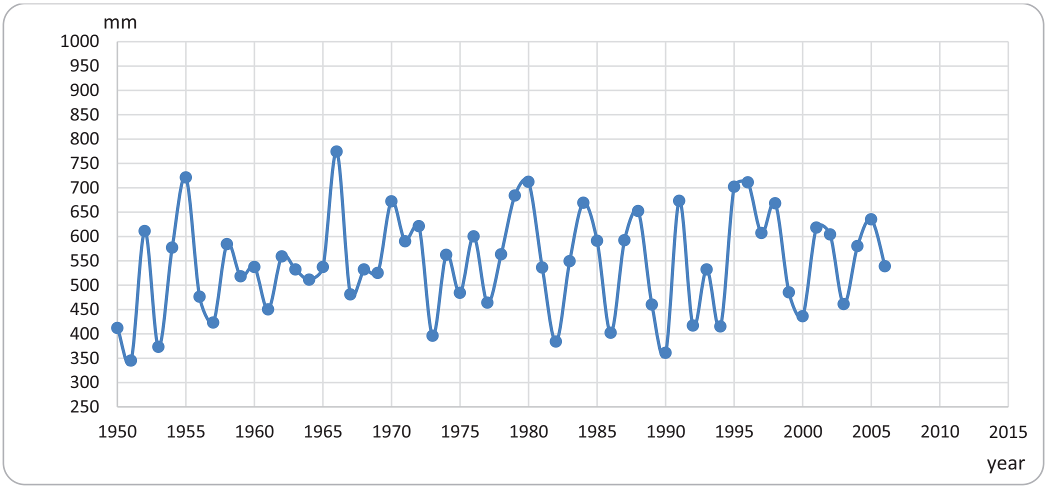

Figure 5. Diagram of the amount of annual atmospheric deposits

From the chart (Figure 3) it is observed that average monthly atmospheric deposits over the years range from 32 mm in March to 73 mm in June.

Digital Relief Model. An increased influence on environmental conditions has its relief. In the Republic of Moldova this was formed as a result of the interaction of the tectonic movements of the Earth's crust and the denudation processes in the contemporary conditions that took place at the end of the neogen. The geomorphologically researched region belongs to Codrii Plateau (55404 ha) and the South Moldavian Plain (22866 ha)

(Ursu, 1980). The territory of the Ialoveni district is characterized by a fragmentary relief and deep valleys (Photo 1). Here predominantly long slopes with a large inclination predominate, favoring the development and development of soil degradation through erosion and landslides. The general division of the basin is southern.

Photo 1. Landscape bazinar

For the object investigated, the Digital Relief Model has been developed that includes specific indexes such as relief plastics, slope classes, exhibition and altitude. From the GRID materials with 48955992 points, the relief plasticity was formed (Figure 6) and extracted the level curves, which make up 40499 in the total length of 168901 km, traced with a numerical equidistance of 10 m (Figure 7).

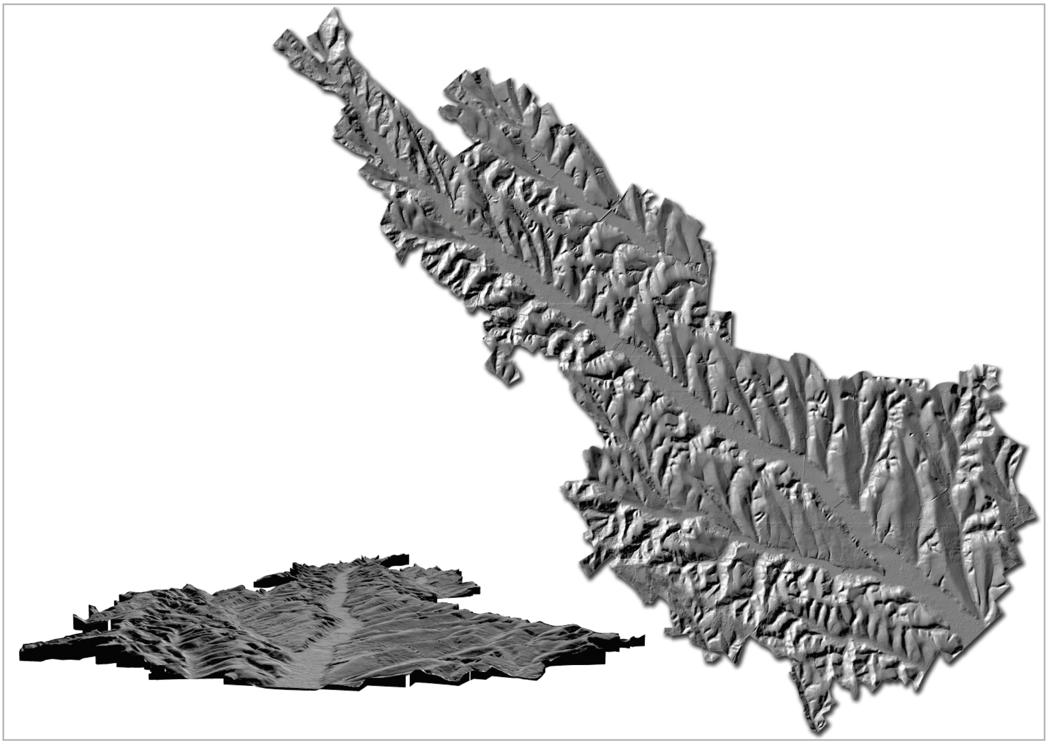

Figure 6. Relief plasticity (DEM)

Each level curve was assigned the absolute value (Z) in the MoldRef-99 reference system. In the case of the georeferenced trapezium it was identified in an altitude with a minimum of 34.7 m and a maximum of 346.5 m, which determines an amplitude of the relief of 325.5 m (Figure 8).

The elevation classes were formed and evaluated over 50 m and are shown in Table 2. Table data show a predominance of the heights class ranging from 120 to 160 m, occupying 29.3% of the territory. In the range of 80 to 200 m the land share is 76.76%.

Using the graphical data of the level curves, the TIN layer of the triangles (200931968 in number) was formed with the sloping slope characteristics and the aspect of the terrain.

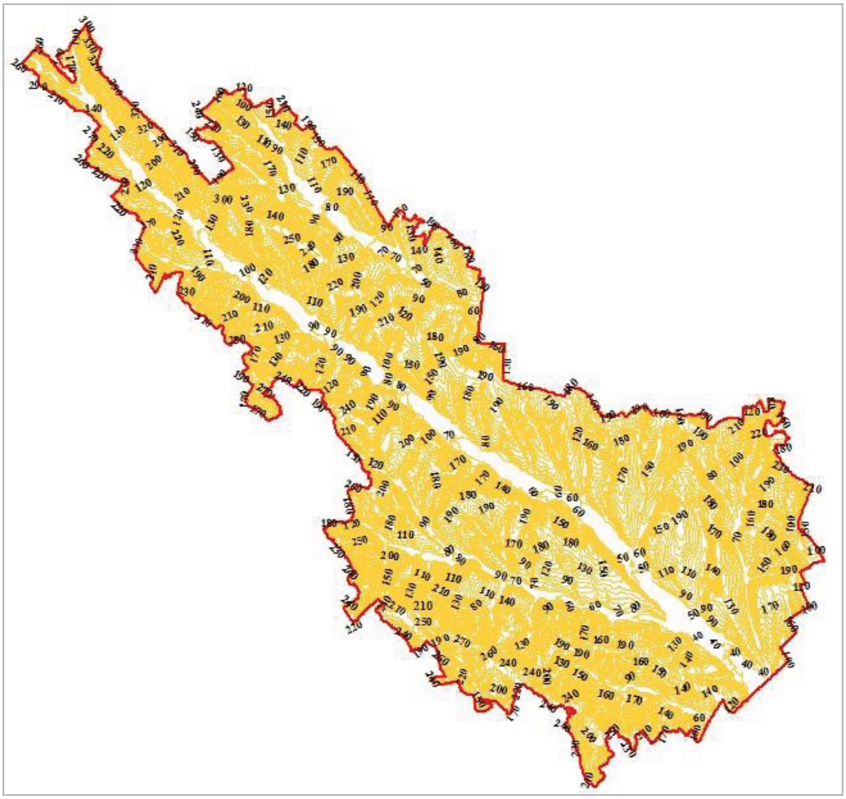

Figure 7. Level curves (10 m equidistance)

Table 2. Cartography of classes by elevation

Altitude range, m	Surface, ha	% of the surface
0-40	316.7324	0.40
40-80	7283.4858	9.30
80-120	18163.5594	23.20
120-160	22988.3524	29.33
160-200	18994.0336	24.23
200-240	6946.5216	8.87
240-280	2759.1877	3.52
280-320	848.8268	1.08
320-360	48.4124	0.06
TOTAL	78306.8155	**100**

From the numerical model of the land, the slope classes, as shown in Figure 1, according to the pedological study methodology were extracted (Florea et al., 1987). Most of the studied area (76.6%) is within the inclination of the slopes from 1º to 10º, where the slopes of 3º-5º predominate by 22.35% (Table 3, Figure 9). In the meadows, land with a slope of up to 1° predominates. The weighted average inclination of the slopes is 8.52º, which presents a very high risk of developing erosion on the surveyed territory (Table 3).

Prevention and control of soil erosion on land on the slopes of the studied area is recommended to be done depending on the size of the slope: - 1º - 3º (11896.83 ha) requires cracking of the slope; spring harvesting and cultivation at a depth of 6-8 cm for pruning crops; 3º-5º (17498.61 ha) requires plowing with plow under 20-22 cm depth and cracking of the autumn plow with hoof; 5º-7º (15500.92 ha) requires the work of the soil with the chisel or the scarifier without turning the furrow to the depth of 20-22 cm with the preservation of the vegetal remains; 7º-10º (15080.74 ha) requires the cultivation of perennial crops and perennial herbs, using the work of the soil with the chisel or the scarifier without turning the furrow to a depth of 20-22 cm with the preservation of vegetal remains and cracking over 10 m across the slope ; 10º-12º (5879.32 ha) requires the cultivation of perennial herbs. In multi-annual plantings, inter-row rooting takes place; >12º (5627.56 ha) requires the establishment of grasslands and forest plantations.

Table 3. Classes according to the tilt of the slopes

The interval	The classes of the slopes	Number of contours	Surface, ha	The average tilt	% of the area of the district	Risk of erosion
0º-1º	Horizontal	3972006	6822.85	0.09	8.71	very weak
1º-3º	Very weak sloping	9778135	11896.83	2.29	15.19	weak
3º-5º	Slightly inclined	29489221	17498.61	4.14	22.35	moderately
5º-7º	Moderately inclined	39035524	15500.92	6.00	19.80	high
7º-10º	Moderately inclined	53874375	15080.74	8.44	19.26	very high
10º-12º	Moderately inclined	27463181	5879.32	10.94	7.51	extremely high
12º-15º	Strongly inclined	23170176	4042.88	13.26	5.16	extremely high
15º-20º	Strongly inclined	10094719	1356.06	16.58	1.73	-
20º-45º	Very strongly inclined	3052796	218.37	25.15	0.28	-
45º-90º	Steep	1001835	10.25	47.76	0.01	-
TOTAL		**200931968**	**78306.82**	**8.52**	**100**	-

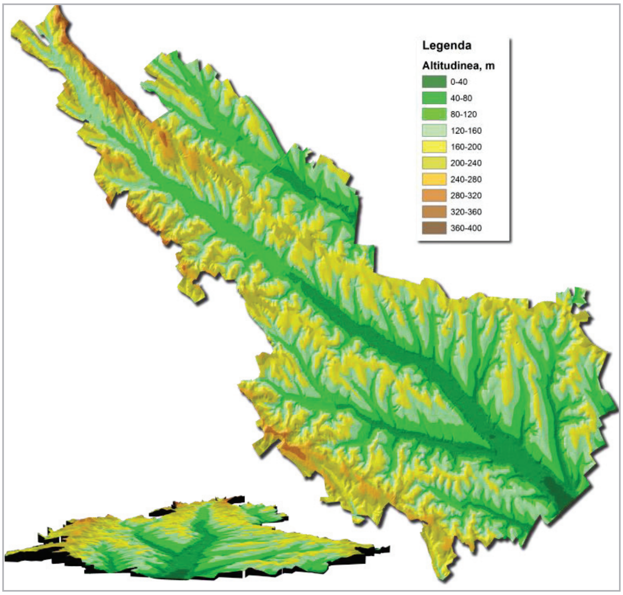

Figure 8. Altitude map of Ialoveni district

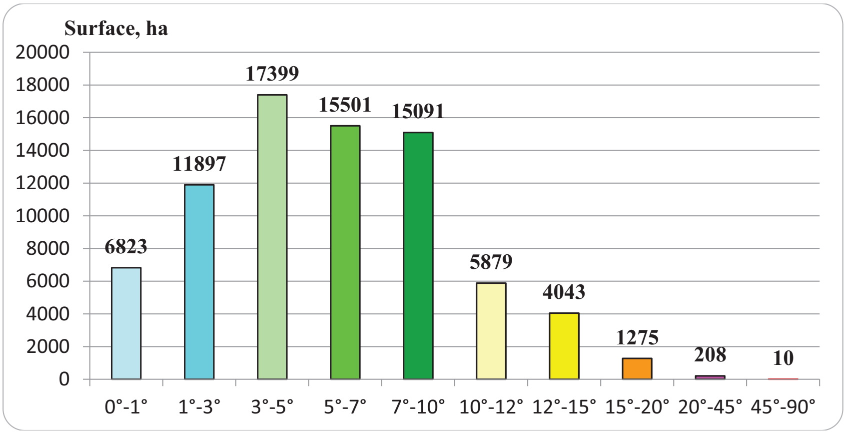

Figure 9. Surface of Ialoveni district

CONCLUSIONS

The researches carried out on the lands within the Ialoveni district were carried out using the geoinformation system. The applied technology has highlighted the deviations of the archival map material from the real situation in the territory. In particular, this refers to administrative boundaries between districts, communes and lands affected by landslides and ravines.

In the structure of land use predominates agricultural ones with a participation of 55% (arable categories predominate by 68% and multiannual plantations by 29%), followed by non-agricultural category - 32% (forests predominate by 54% and non-agricultural pastures by 26%) and 13% - infrastructure (the localities - 64% and roads with 20% with the length of 4684 km).

The hydrographic system is represented by 1669 objects covering 2144 hectares, where lakes with 1623 hectares predominate and 362 hectares of rivers and canals with a length of 568 km. About 2/3 of the surface water has a chemical composition and unsatisfactory quality indices for their use in irrigation.

Of the non-agricultural subcategories, land slopes predominate with 1640 ha - 82% and the ravines with 346 ha - 17% with a 190 km radius.

ACKNOWLEDGEMENTS

This research work was carried out with the support of the Supreme Council for Science and Technological Development of the Academy of Sciences of Moldova and also was financed from Project Partnership No. 15.817.05.33 A.

REFERENCES

Arinushkina, E.B. (1963). *Chemical analysis of soil*. Moscow: Issue ANSRC, 485.

Filipciuc, V. (2007). Soil and water supply to irrigation. In: Drought and methods to minimize adverse consequences. *Annex to "Agriculture of Moldova", Chisinau, 7-8*, 23–30.

Gerasimov, I.P., Glazovskaya, M.A. (1960). *Basics of soil science and soil geography*. M.: Science, 417.

Kashansky, A.D. (1987). *Compilation and use of soil maps*. Ed. M. Agropromizdat, 273.

McBratney, A.B., Mendonca, Santos, M.L., Minasny, B. (2003). On digital soil mapping. *Geoderma, 117*(1-2), 3–52.

Rozloga, Iu. (2013). The spatial positioning of the land infrastructure within the upper Cogilnic catchment area with the use of GIS. "Moldavian chernozems - the evolution, protection and restoration of their fertility": Conf. with internal participation, dedicated to the 60th anniversary of Institute of Pedology, Agrochemistry and Soil Protection "Nicolae Dimo": Collection of papers. Chisinau ("Recklama" Tipography), 290–295.

Rozloga, Iu. (2015). Application of the geoinformatics system for establishing and evaluating erosion processes. *Academy of Sciences, Innovation, Culture and Art "Akademos", Chisinau, ASM, 4*(39), 66–74.

Ursu, A.F. (2011). *Soils of Moldova*. Chisinau: Î.E.P. Science, 323.

Ursu, A.F. (1980). *Soil ecological zoning of Moldova*. Chisinau, Science, 208.

***Agrometeorological Bulletin, (1950-2016). Ch.: Hidrometeo.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

AGROTECHNICAL CHARACTERIZATION OF LANDS FROM THE SOMEŞAN PLATEAU FOR AGRICULTURAL SUSTAINABILITY

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Abstract

The agrotechnical characterization of lands from the Someșan Plateau targets the knowledge of their restrictions and limitations with the purpose to determine the needs and possibilities of increasing the production capacity, under the conditions of a durable agriculture. The purpose of the paper is the technological characterization of lands and the specification of agrotechnical measures to set up durable agricultural technologies, the determination of indices of characterization of rain aggressiveness (Fournier Index, Modified Fournier Index, Angot Pluviometric Index), of the best sowing time and the amount of active biological temperatures, as well as the degree of water ensurance of plants during the vegetation period. Monitoring and variability of climate elements was achieved during 2014-2018, through a network of 10 HOBO microstations, which store soil temperature data electronically (at 10, 30, 50 cm deep) and air (at 1 m hight), soil moisture (at 10 cm depth) and rain gauges. According to the values of the Fournier Index, 31.81% of the soils fit in low risk of rain erosivity. At the same time, the values of the Modified Fournier Index in most of the stations are between 90-120 and fit in a moderate rain risk of aggressiveness. The percentage analysis of the values of this index shows that 12.5% of the values crorespond to a low rain risk of aggressiveness, 58.33% of the values correspond to a moderate rain risk of aggressiveness and 29.16% of the values correspond to a high rain risk of aggressiveness. From the percentage analysis of averages of monthly values of the Angot Index it results that 54.9% of the values are subunit and 45.1% of the values are over unity, indicating the fact that drought intervals prevail over rain ones. The analysis of the best sowing time leads to the recommendation to maintain the reference intervals for the Someșan Playeau, thus: maize, 10-25 April; potato, 10-30 March; clover, 1-10 March. An increase of the amount of active biological degrees and the need to remake the zoning of favorability conditions for certain crops, according to the new microclimate conditions of the area is registered. When it comes to the degree of water ensurance of plants during the vegetation period for the main crops, one can notice that during 2014-2018, in the case of spring row crops, the water need was ensured during an optimal interval of 57.14% from the vegetation period in the case of maize, of 53.49% in the case of potato, of 53.57% in the case of clover and of 47.67% in the case of wheat.

***Key words**: climate indicators, agricultural sustainability, Someșan Plateau.*

INTRODUCTION

The agro-technical characterization of lands targets the knowledge of their restrictions and limitations, with the purpose to determine the needs and possibilities of increasing the production capacity, under the conditions of a durable agriculture. The set up of regional studies, well-documented scientifically, is imperative, taking into consideration the measures to adapt to climate changes and the durable land management.

It is well-known that agriculture is one of the activity sectors sensitive to the variability of climate factors and especially to extreme weather events (drought, floods, storms), considered as significant risk factors influencing negatively both vegetal and animal production. The regional distribution of the impact of climate changes on agricultural production can vary a lot (Donatelli et al., 2012). Thus, for the southern part of Europe, some of the biggest decreases of crops are foreseen, of almost 25%, by 2080 under the conditions of a temperature rise by 5.4°C (Ciscar et al., 2011). Under these conditions, Bindi and Olesen (2011) also take into account, an increase of the risk of failure, especially in the case of unirrigated summer crops.

In the Romanian National Strategy on climate changes 2013-2020, set up by the Ministry of Environment and Climate Changes (2013), it is mentioned that the temperate climate shall be significantly changed within the next 50-100 years. The average daily rate of rainfalls shall be reduced by approximately 20%, foreseeing that they will be in a larger quantity for short periods of time and on reduced surfaces, which can lead to an increase of flood frequency, especially of flash floods, soil and ecosystem degradation. This rainfall predictability varies a lot, from one region to another, according to geographical parameters. The information on the future agrometeorological conditions shall be important in evaluating the impact of climate changes and in the development of adequate strategies to reduce and adapt to climate changes, the policies regarding the land use and in investigating the potential economical responses (Rivington et al., 2013).

The phenomena of dryness and drought generated by the lack of rainfall, tropical heat waves, associated with dry winds, are the most complex climate risks possible in any season which lead to the depletion of the water reserve in the soil, the appearance of the pedological drought, with negative effects upon plant metabolism and, consequently, to drops of the vegetal production (Bogdan, 2005; Povară & Văduva, 2009). The measures which can limit and counteract the drought effects can be applied both locally or regionally, and they can be adapted based on the features of the geographical area and the pedoclimate conditions, where the application of these measures is imposed. Such measures refer to (Rusu et al., 2017): the use of a biological material which is resistant to hydric and thermal stress; the use of agro-technical measures favorable to accumulate, preserve and effectively value water coming from rainfall; the use of a conservative agricultural system based on the protection of soil and avoidance of desertification; identifying areas vulnerable to climate changes and using biological material with biological features and pedoclimate demands specific to the new climate tendencies of the areas vulnerable to climate risks.

The purpose of the paper is the technological characterization of lands and the specification of agro-technical measures to set up durable agricultural technologies, the determination of indices of characterization of rain aggressiveness (Fournier Index, Modified Fournier Index, Angot Pluviometric Index), of the best sowing time and the amount of active biological temperatures, as well as the degree of water ensurance of plants during the vegetation period. The need to determine these indices comes from the existing situation in the agriculture from the Someșan Plateau, where, due to inadequate tillage systems, one can notice a series of negative aspects regarding soil fertility, the quantity and quality of the productions obtained. In this region, accelerated erosivity is not only the result of combined processes of two neotectonic movements (the uplift of the hill compartment and local subsidence), but also of the friability of geological formations, over which a wrong use of lands overlaps.

MATERIALS AND METHODS

Monitoring and variability of climatic elements, from the Someșan Plateau,was achieved during 2014-2018, through a network of 10 HOBO-MAN-H21-002 (On-set Computer Corp., Bourne, MA, USA) stations which store soil temperature data electronically (at 10, 30, 50 cm deep) and air (at 1 m hight), soil moisture (at 10 cm depth) and rain gauges. HOBO Smart Temp (S-TMB-M002) temperature sensors and Decagon EC-5 (S-SMC-M005) moisture sensors were connected to HOBO Micro Stations. Additionally, tipping bucket rain gauges (RG3-M) were deployed to measure rainfall. Data was downloaded from the Micro Stations every four months via laptop computer using HOBOware Pro Software Version 3.7.2. Soil types, land slope and exposition, altitude and geographical coordinates of the locations in which stations were set are shown in Table 1 (Duda, 2018).

How to use the lands and the degree of soil vegetation cover are the most important factors affecting the intensity and the frequency of surface erosivity following the action of rainfall (García-Ruiz, 2010; Nunes et al., 2011; Marin et al., 2015). The soil erosivity, under all its aspects, is one of the restrictive factors with repercussions on the agricultural production, its quality, and last but not least on the production costs, which results in the need to apply the best practices in the use of agricultural lands. The assessment of rain aggressiveness upon the sublayer, respectively fitting the lands in classes with a certain potential of erosivity as well as the susceptibility regarding the start of slope processes can be determined with indices calculated based on the pluviometric regime of the area researched (De Luís et al., 2009; Blaga, 2013).

Table 1. Configuration of stations in the Someșan Plateau

No. Station	Locality (County)	Altitude (m)	Soil type	Exposition	Slope, %
1	Cristorel (Cluj)	404	Preluvosol	N	8-10
2	Borșa (Cluj)	332	Faeoziom	S	2-3
3	Lelești (Bistrița-Năsăud)	606	Regosol	V	25-26
4	Șomcutu Mic (Cluj)	271	Aluviosol	S	2-3
5	Căprioara (Cluj)	416	Preluvosol	S	4-5
6	Almașu (Sălaj)	323	Aluviosol	S	8-10
7	Racâș (Sălaj)	253	Preluvosol	S-E	2-3
8	Șimișna (Sălaj)	256	Preluvosol	N-E	7-9
9	Ileanda (Sălaj)	225	Aluviosol	S	2-3
10	Bunești (Cluj)	209	Preluvosol	N	6-8

The rainfall regime reflects the aggressiveness of rain erosivity on the soil by their volume, duration and intensitaty, their effect is much more stronger when they appear after a long period of drought (Costea, 2012) and which, directly or indirectly, represent the main climate-genetic factor involved in a wide range of geomorphic processes and phenomena (Arghiuș, 2010). The indices used in the analysis are: the Fournier Index (Fournier, 1960), the Modified Fournier Index (Arnoldus, 1980) and the Angot Pluviometric Index.

The Fournier Index (*FI*) uses entry data easily accessible, that is the quantity of rainfall from the rainiest month of the year and the annual quantity of rainfall. The calculation formula of this index is given by the report between these elements, therefore: $FI = Pm^2/P$, where, Pm - is the quantity of rainfall from the rainiest month of the year (mm); and P - the annual quantity of rainfall (mm). The expression analyzes the erosivity capacity by rainfall in a certain territory (Satmari, 2010). The erosivity potential can be characterized based on the K values of the FI (Satmari, 2010; Gabriels, 2006). The conceptual scale (Oduro-Afriyie, 1996) to assess the erosivity risk based on the FI is presented in Table 2 (Meddi, 2013).

Determination of Modified Fournier Index. For the study of degraded lands, a Modified version of the Fournier Index (MFI) by Arnoldus (1980) was introduced, with the following calculation formula:

$$F_M = \sum_{i=1}^{12} \frac{Pi^2}{P}$$

where: Pi - the average quantity of rainfall for month i (mm); P - annual average quantity of rainfall. The classes of rain aggressiveness based on MFI (Yuksel et al., 2008; Blaga, 2013; Meddi, 2013) are presented in Table 3.

Table 2. Evaluation of the risk of erosivity based on the Fournier Index

Class	Soil losses (t/ha/year)	Fournier Index (K)(mm)	Risk of erosivity
1	< 5	< 20	Very low
2	5-12	21-40	Low
3	12-50	41-60	Moderate
4	50-100	61-80	Severe
5	100-200	81-100	Highly severe
6	> 200	> 100	Extremely severe

Table 3. Classes of rain aggressiveness based on Modified Fournier Index (MFI)

Class	MFI	Rain aggressiveness
1	< 60	Very low
2	60-90	Low
3	90-120	Moderate
4	120-160	High
5	>160	Very high

Angot Pluviometric Index (API) is the mathematical expression of the report between the daily average quantity of rainfall in a month (real quantity of rainfall) and the multiannual average quantity, respectively the quantity which would have been its share in the case of uniform division of the quantity of annual rainfall during all days of the year. The poor periods or the surplus from a pluviometric point of view from within a year can be highlighted thanks to this index based on which one can make a delimitation between the droughty months and the rainy ones. Thus, one can highlight the rainy intervals (K>1) and the droughty intervals (K<1). According to the values obtained, the marks of *rainy month* for over unity values and *droughty month* for the subunit values were attributed. They are calculated based on the formula: $K = p/P$; $p = q/n$ and $P = Q/365$; where: q - daily average quantity of rainfall; n - monthly number of days; Q - annual average quantity of rainfall. $K = \frac{p}{q} = \frac{q/n}{Q/365} = \frac{365*q}{Q*n}$. On the whole, this formula highlights the climate shade of each month (the subunit values indicate the droughty months and the over unity ones indicate the rainy months). It can be plotted by using real values (the variation curve has a better accuracy) or generalized (granting the 0.5 value for all the months where the result was subunit and 5.5 for the over unity ones). Blaga (2013) states that based on the values (subunit or over unity) of this index, susceptibility classes were determined to start the slope processes, for linear erosivity processes or for floods. If the values of the API range between 1.0-1.5, there is a very low and low predisposition to start these erosivity processes. In case of values ranging between 1.5-2.0, this predisposition is average. When the values of the API exceed the 2.0 value, there are conditions to start the erosivity processes, and in case of values over 2.5, there are very favorable conditions to start the processes of slope and linear erosivity (Blaga, 2013).

Determination of the optimal sowing period for the main crops in the Someșan Plateau and of the amount of active biological temperatures gives us the possibility to check and compare the sowing periods stated in expert literature with the data resulted from field observations in order to make adequate technological recommendations, adapted to the current conditions in the field. The optimal sowing period differs according to the species cultivated, the types and hybrids cultivated, destination of the crop, orographic conditions of the crop area. The growing phases of the plants are influenced mainly by temperature, which is decisive for germination, therefore, at this stage, every crop plant needs a minimum germination temperature, based on which the crops are grouped in epochs and sowing emergencies (Table 4; Munteanu et al., 2014).

Table 4. The optimal sowing periods of main crops in the Someșan Plateau

Crop and optimum time of sowing in the Someșan Plateau	Optimal time of sowing in the Someșan Plateau	Temperature in soil at sowing depth, ^{0}C
Spring crops		
Epoch I, Emergency I - Clover	1-10.III.	1-3
Epoch I, Emergency II - Potato	10-30.III.	5-8
Epoch II, Emergency I - Maize	10-25.IV.	8-10
Autumn crop		
Wheat, triticale	20.IX.-15.X.	3

Determination of the amount of biological active degrees recorded for the main crops in the Someșan Plateau is calculated only for the vegetation period. In order for a plant to reach maturity, it needs a certain amount of heat expressed as a sum of active biological temperatures (ABT), during the whole vegetation period, based on the following formula: $\Sigma ABT = \Sigma(Tef-Tb)$; where, ΣABT - a sum of active biological temperatures recorded during the vegetation period (thermal constant); Tef – effective temperature expressed as daily average temperature, calculated as an average between the daily maximum temperature and the daily minimum temperature; Tb - basic temperature or the biological threshold (temperature below which there are no more visible growths).

The thermal constants for the main plants cultivated in the Someșan Plateau are presented in Table 5 (Munteanu et al., 2014).

Table 5. Thermal constants of the main crops

Crop	Thermal constants, °C
Wheat	1800-2300
Maize	1700-2500
Triticales	1200-1390
Potato	1500-3000
Clover	800-1100

The determination of the areas which have conditions of thermal stress during the vegetation season is one of the most important parameters regarding the agroclimate conditions of the area.

Determination of the ensurance degree with water for plants during the vegetation period for the main crops. In order to determine the best moisture period for plants (IOUP, %), when the development of plants is made accordingly, the soil capacity for water in the land (CC), the fading coefficient (CO) and the interval of active moisture (IUA) were previously determined.

The capacity for water in the land (CC) is the water at the plants' disposal without being lost by infiltration or evaporation (Rusu et al., 2012) and was determined by indirect method by using the formula: CC = CO + 10 in case of soils with clay-sand texture; +12 in case of soils with clay-argil texture; +13 in case of soils with argil texture; where, CO is the fading coefficient.

The fading coefficient (CO) was determined by the indirect method which is based on the existing correlation between the CO and the hygroscopicity coefficient (CH) by using the following formula: $CO\% = CH \times 1.47$; where: CO - the fading coefficient, %; CH - the hygroscopicity coefficient, %. The fading coefficient is the limit below which the plant development isn't possible due to the inaccesibility of water for plants, resulting in their permanent fading.

The interval of active moisture (IUA) is the main indicator of the potential water reserve of a soil and represents the interval corresponding to the moisture the soil can retain and have at the plants' disposal (Rusu et al., 2012). The values of IUA or of the capacity of useful water (CU_{IUA}) corresponding to this interval were obtained based on the formula: $CU_{IUA} = CC-CO$, where: CU_{IUA} - water capacity, %; CC - land capacity, %; CO - fading coefficient, %.

The best moisture interval for plants (IOUP) was calculated for the interval 60-90% of the IUA value in order to compare with the data recorded in the field of the soil moisture with a view to analyze the moisture need ensured during the vegetation period of the plants.

RESULTS AND DISCUSSIONS

The annual values of the **Fournier Index** calculated for the stations which have a pluviometer, during 2014-2018 ranged between 1.21 at Cristorel station in 2014 and 28.89 at Ileanda station in 2016. As for the distribution of quantities of monthly rainfall recorded at the stations placed in the Someșan Plateau (Table 6), one can notice that the highest quantities were recorded during June (Almașu and Cristorel - 2018; Ileanda - 2016 and 2018; Șomcutu Mic - 2015 and 2018) and September (Almașu and Bunești - 2015; Cristorel - 2015 and 2017; Ileanda - 2015 and 2017). During 2014-2018, the multiannual value of the FI ranges between 11.69 at Cristorel station and 20.85 at Ileanda station (Figure 1).

Table 6. Annual values of Fournier Index (mm) in the Someşan Plateau (2014-2018)

Year/station	Almaş	Buneşti	Cristorel	Ileanda	Şomcutu Mic
2014	18.66672	-	1.21116	-	4.92126
2015	18.97173	21.72168	3.543022	11.21995	20.3932
2016	18.14629	18.3996	9.453125	28.89284	19.102
2017	3.608182	12.0873	25.19018	21.41495	4.165643
2018	27.19145	-	19.05654	21.90851	19.12136
Average	17.31688	17.40286	11.69081	20.85906	13.54069

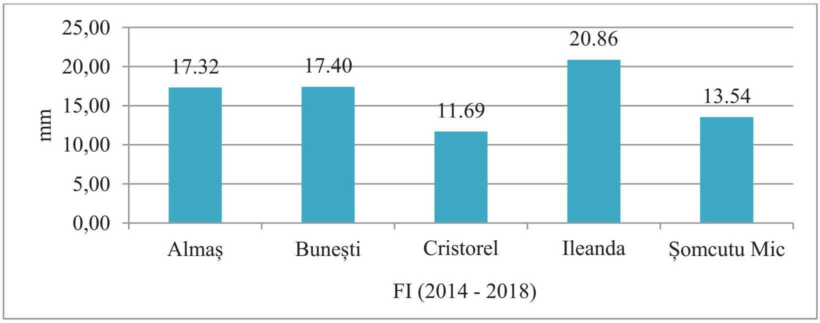

Figure 1. Multiannual value of Fournier Index (FI) from the Someşan Plateau (2014-2018)

In accordance with the values of the FI obtained during 2014-2018 (FI < 20) and of the erosivity classes, by using the Oduro-Afriyie conceptual scale (1996) with 6 classes of erosivity, the soils analyzed at the stations from the Someşan Plateau are submitted to a very low rain erosivity risk, being classified in class 1 of rain erosivity, with soil losses lower than 5 t/ha/year. A higher risk of erosivity was recorded in 2016 at Ileanda station, the FI value of which was bigger (28.89), classifying the soil in class 2 of erosivity, with soil losses ranging between 5-12 t/ha/year. Figure 2 shows the evolution of the K values of FI during 2014-2018 in the Someşan Plateau. From the analysis of the results obtained, one can notice that the FI values calculated reach high values, ranging between 21 and 60, sporadically at all 5 stations analyzed (2015 Buneşti, 2017 Cristorel, 2018 Şomcutu Mic).

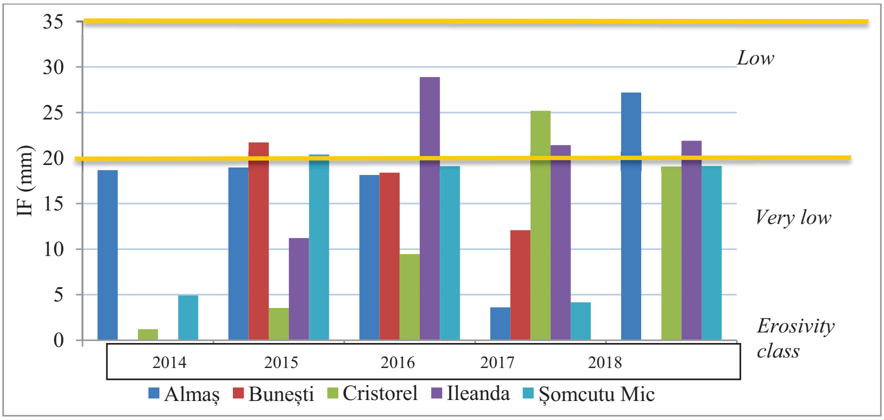

Figure 2. Annual values of FI and erosivity rainfall risk (2014-2018)

The multiannual average of the FI during 2014-2018 is 16.16, which classifies the soils from the Someşan Plateau in class 1, with very low risk of rain erosivity, with soil losses lower than 5 t/ha/year. The percentage analysis of the K values of FI calculated during 2014-2018 shows the fact that 68.19% of the soils fit into class 1, with very low risk of rain erosivity and 31.81%

of the soils fit into class 2, with low risk of rain erosivity.

Arnoldus (1980) considers that the **Modified Fournier Index** version (MFI), is an index which allows for a better approximation of the index of rain erosivity, with which it is in linear correlation (Meddi, 2013).The MFI values calculated during 2014-2018 range between 45, at Cristorel station in 2017 and 216.96 at Almaşu station in 2014, in most stations all MFI values range under between 90-120, which, according to the classes of rain aggressiveness mostly used (Yuksel et al., 2008; Blaga, 2013; Meddi, 2013) the soils fit into class 2 and are submitted to a moderate risk of rain aggressiveness. Also, most of the multiannual values of MFI during 2014-2018 range between 90 and 120, the lowest are obtained at Buneşti station (79.11) and the highest values at Almaşu station (149.69) (Figure 3).

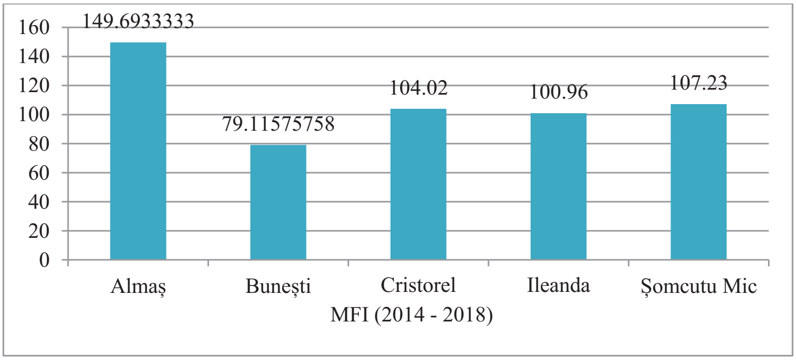

Figure 3. Multiannual value of MFI at stations located in the Someşan Plateau

The evolution of MFI values during 2014-2018 in the Someşan Plateau is represented in Figure 4. From the analysis of the results obtained, one can notice that the highest calculated values of MFI were obtained at Ileanda, Cristorel and Almaşu stations during 2013-2015 and the lowest were obtained at Buneşti and Cristorel during 2015-2018. The percentage analysis of MFI values calculated during 2014-2018 at the stations from the Someşan Plateau shows the fact that 12.5% of the values correspond to class 1, with low risk of rain aggressiveness, the values are lower than 60, and 58.33% of the values classify the soils in class 2, with moderate risk of rain aggressiveness, the values ranging between 90-120 according to the associated scales mostly used and mentioned in expert literature and the rest of 29.16% fit into class 3 of high risk.

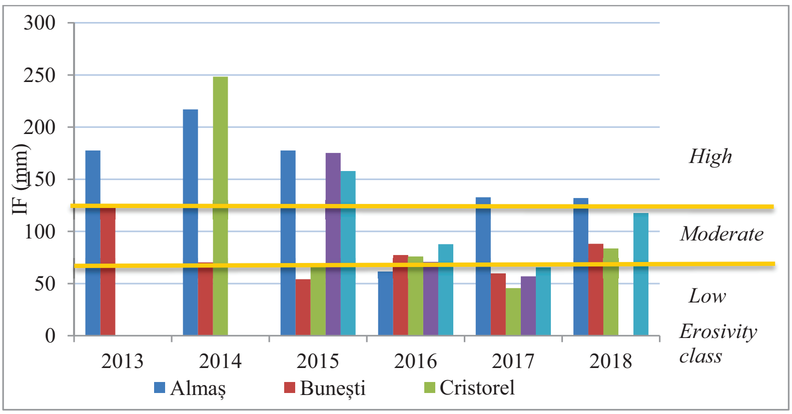

Figure 4. Annual values of MFI and erosivity rainfall risk (2013-2018)

Thus, following the analysis of MFI values calculated during 2014-2018, the conclusion is that most of the soils from the Someşan Plateau are submitted to a moderate risk of rain aggressiveness (58.33%) and to a high risk of rain aggressiveness (29.16%), differentiated based on the morpho-dynamic features of the space analyzed and the duration and intensity of rainfall from the period of time analyzed.

The Angot Pluviometric Index (API). From the analysis of the K values of the API for the stations from the Someşan Plateau which were equipped with a pluviometer, during 2014-2018, one can see subunit values during the summer and winter periods in most of the years and stations and over unity values during the spring and autumn months. The highest values over 2,00 of the API were recorded in 2015 and 2018, when in June it was recorded 2.29 and respectively 2.62. Based on the annual average, all stations have average values, approximately 1.01. The monthly averages of the values of the Angot pluviometric index at the stations analyzed from the Someşan Plateau are represented in Figure 5.

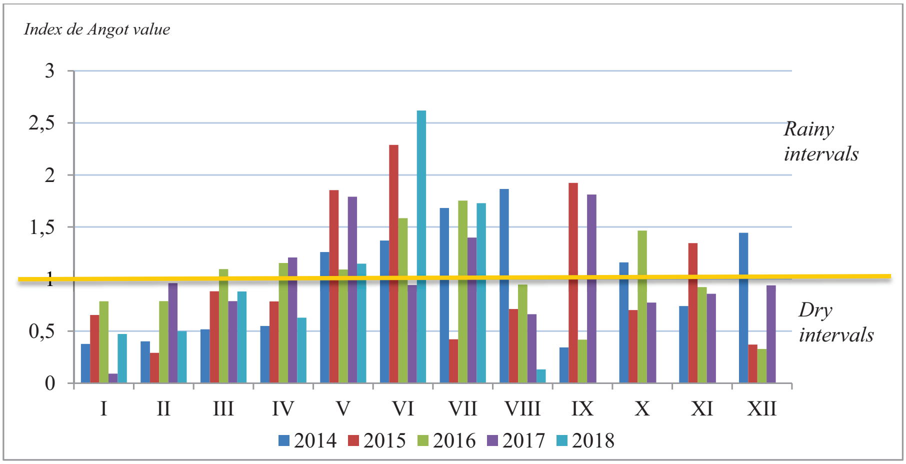

Figure 5. Monthly averages of Angot pluviometric index (2014-2018)

From the percentage analysis of the averages of monthly API values during 2014-2018, at the stations from the Someşan Plateau it results that 54.9% of the values are subunit, while 45.1% of the values are over unity, showing the fact that droughty intervals are predominant to rainy ones. The analysis of the correlation between the multiannual averages of the monthly average amount of rainfall and the multiannual averages of API during 2014-2018 shows a strong, positive and direct correlation between the two parameters, with a correlation coefficient R^2 of 0.77, which indicates the fact that 77% of the API values can be explained in linear relation to rainfall (Figure 6). The index value K >a 2 can highlight the existence of conditions to start slope and linear erosivity processes. During 2015 and 2018, according to the API values, the amount of rainfall was higher compared to the previous year, which indicates the creation of favorable conditions to start the slope and linear erosivity processes (Figure 7).

When it comes to the weight of predisposition in starting slope and linear erosivity processes based on the API values, one can notice that in 59% of the cases there is no risk of rain erosivity, in 21.42% of the cases there is a low and very low predisposition, in 16.07% of the cases favorable conditions to start the erosivity processes are created, and in 3.57% of the cases very favorable conditions of linear rain erosivity were created.

Determination of the optimal sowing period for the main crops in the Someşan Plateau. When establishing the best sowing time, one must take into account the minimum germination temperature of seeds and the amount of grades of useful temperature in every plant. Also, the orographic factors together with the agrometeorological parameters recorded in the crop areas, which refer to the first autumn frost and the last spring frost, temperatures higher than 32°C, the amplitude of temperatures during the summer months or those associated to the vegetation phenophases of plants and last but not least, the risk of developing of agricultural drought are important. Based on the results obtained following the analysis of data recorded in the Someşan Plateau during 2014-2018, one continues to recommend the reference intervals, thus: maize, 10-25 April; potato, 10-30 March; clover, 1-10 March.

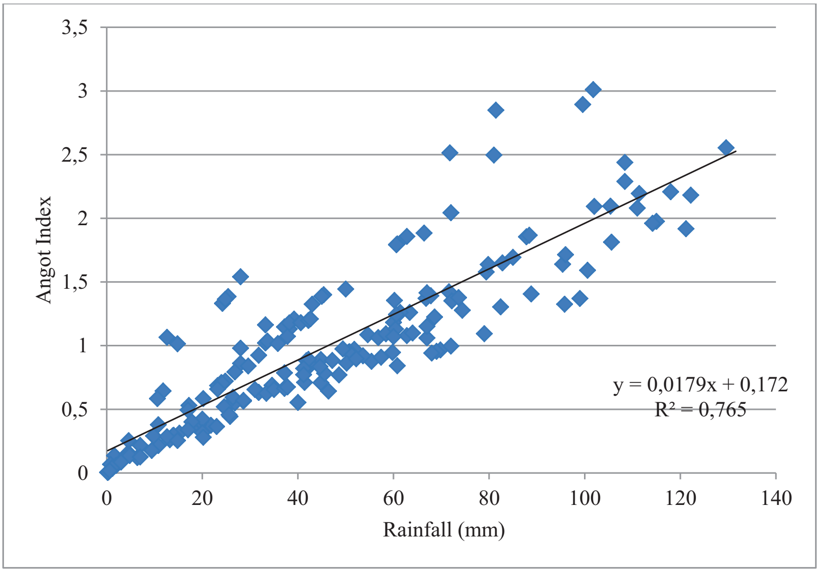

Figure 6. Correlation between the multiannual averages of monthly average quantities of rainfall and the multiannual averages of Angot pluviometric index (2014-2018)

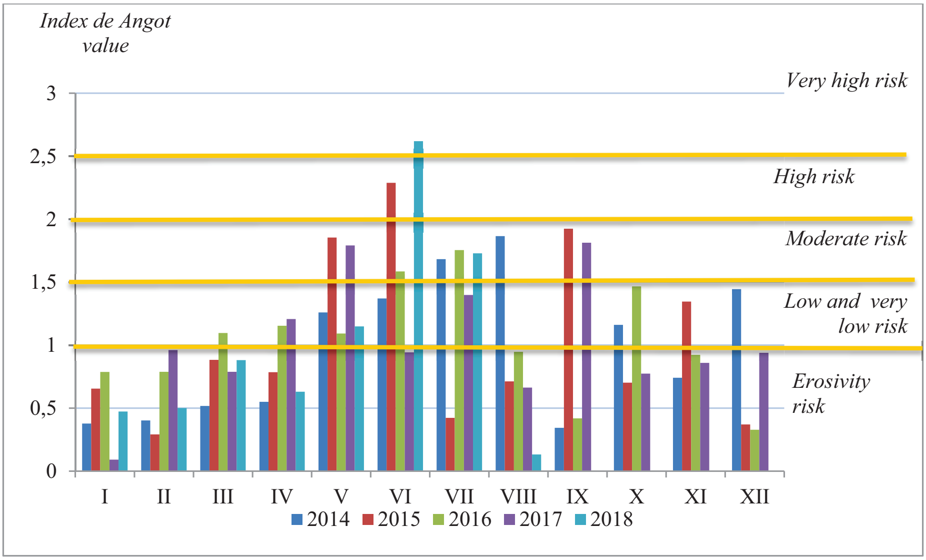

Figure 7. Monthly averages of Angot pluviometric index and the risk of rain erosivity (2014-2018)

The amount of biological active degrees (ABT) recorded for the main crops in the Someşan Plateau. For the calculation of ABT, the longest periods of vegetation, corresponding to the half-late and late types/hybrids in the crops analyzed were taken into consideration, thus: 290 days for autumn wheat (Almaș station) and 320 days the other stations, 180 and 210 days for maize and 180 days for potato. For the crops analyzed, in the case of wheat, during 2015-2017, average values of ABT were obtained, of 1831°C, the values ranging between 2017°C at the Bunești station in 2016 and 1712°C at Ileanda in 2017 (Table 7). These values are close to the need of temperature for the wheat crop mentioned in expert literature.

In the maize crop, during 2013-2018 one can notice that the amounts of active biological temperatures were ensured, which exceeded the limits set up for the III crop area, ensuring thus, the thermal need both for crop area II and for crop area I.

In the case of potato, a vegetation period of 180 days was taken into account, between 10 March and 26 September. During this interval average values of ABT of 1441°C were accumulated to ensure the need of reaching maturity of this crop. The values ranged between 1847°C, at Cristorelstation in 2013 and 1482°C, at Ileanda, 2016.

In the case of clover, on an average, 1683°C are insured, for a vegetation period ranging between 1 March and 30 September. The highest values were recorded at Cristorel, in 2016 (1865°C), and the lowest at Ileanda, in 2016 (1504°C), these values being almost 2 as high as the need of reaching maturity.

Table 7. Value of amount of biological active degrees registered at Almașu, Bunești, Cristorel, Ileanda and Șomcutu Mic for wheat, corn, potato, and cloverleaf (2015-2017)

Station	Year	Wheat/Triticale		Maize		Potato	Clover
		total	pre-winter				
		20.IX-06.VII/06.VIII		10.IV-08.X	10.III-08.X	10.III-26.IX	01.III-30.IX
Almaș	2015	1802,841	532,872	-	-	-	-
	2016	1909,8515	542,7455	1496,4105	1714,19	1517,4865	1735,3615
	2017	1813,579	520,068	1509,3985	1758,061	1506,5095	1776,189
Bunești	2015	1759,0835	430,382	1307,155	1510,1435	1331,1735	1528,252
	2016	2017,277	455,802	1457,2015	1706,26	1461,7415	1727,837
Cristorel	2013	1880,056	259,462	1582,509	1847,742	1587,093	1865,283
	2017	1713,109	438,298	1414,5885	1638,098	1416,0715	1654,7165
Ileanda	2015	1738,594	405,141	-	-	-	-
	2016	1984,185	420,375	1275,224	1482,968	1309,303	1504,486
	2017	1712,4975	416,137	1403,3075	1667,148	1420,9545	1694,12
Șomcutu Mic	2015	1753,877	437,1415	1554,971	1795,4135	1573,3125	1818,9835
	2016	1991,326	461,8135	1301,726	1511,9405	1325,0455	1531,3805
	2017	1733,3465	443,5655	1407,5895	1665,2005	1409,808	1685,67

The ensurance degree with water for plants during the vegetation period for the main crops in the Someșan Plateau. The best moisture time for friends (IOUP, %) is represented by the interval ranging between 60-90% of the active moisture interval (IUA), interval during which the development of plants runs accordingly. The percentage representation of the best moisture time for plants at the stations from the Someșan Plateau during 2014-2018 is showed in Figure 8. During 2014-2018, in the case of spring weeding crops, the need of water was ensured in an optimal interval of 57.14% from the vegetation period in the case of maize, of 53.49% in the case of potato, of 53.57% in the case of clover and of 47.67% in the case of wheat. During 2014-2018, one can notice an increase of extreme climate phenomena, represented by long drought periods correlated with reduced amount of rainfall, which leads to the reduction of the soil capacity to ensure constantly the optimum moisture for plants.

Special attention needs to be paid if the water deficit from the soil is associated to the drought periods during critical times with maximum demands of moisture and temperature, which have a bad influence upon the physiological processes of plants, resulting in decreases of their productivity. In this regard, it is necessary to apply some agricultural technologies which result in maintaining water in the soil, as well as irrigating the crops to ensure the best production potential of crops.

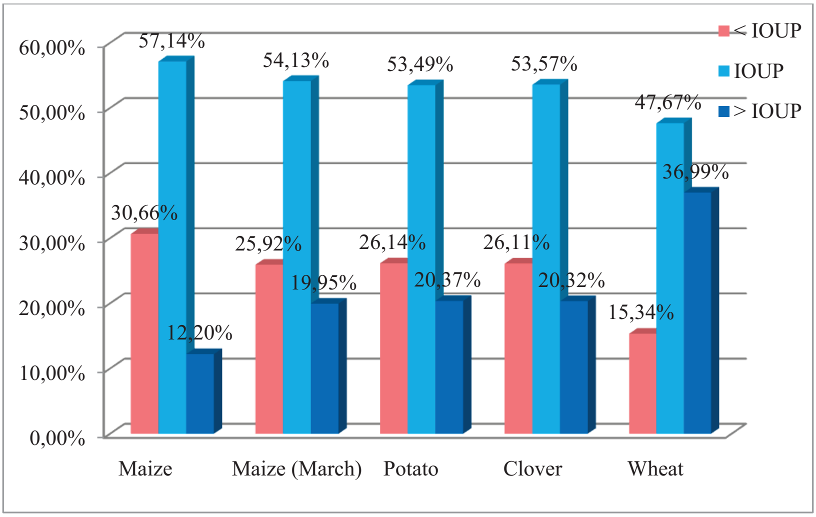

Figure 8. The optimum moisture range for plants (IOUP, %) (2014-2018)

CONCLUSIONS

The determination of the evaluation indices of rain aggressiveness upon the sublayer (Fournier Index, Modified Fournier Index and Angot Pluviometric Index) suggest favorable conditions for the appearance and development of rain erosivity processes. The effect is the more intense when they appear after a period of prolonged drought, especially in March, April, July and August or, in some cases, in October and November. This situation involves the application of some soil tillage systems which incorporate/maintain at surface the vegetral debris, especially during summer and autumn.
As for the degree of water ensuring of plants during the vegetation period for the main crops, one can notice that during 2014-2018, for the spring weeding crops, the water need was ensured during an optimal interval of 57.14% from the vegetation period in the case of maize, of 53.49% in the case of potato, of 53.57% in the case of clover and of 47.67% in the case of wheat. One can notice an increase of extreme climate phenomena, represented by long drought periods correlated with reduced amount of rainfall, which leads to the reduction of the soil capacity to ensure constantly the optimum moisture for plants. Therefore, ensuring irrigations during the droughty periods is a must, especially when temperatures are higher than 32°C, during June-August, which coincides with the critical period, with maximum demands of rainfall for the clover, wheat and maize crops.
The determinations of degrees of active biological temperature and their analysis during 2014-2018 recommend to take into account the cultivation of types and hybrids which best value the conditions from the Someșan Plateau, together with a remaking the zoning of favorability conditions for certain crops, according to the new microclimate conditions of the area.
Following the analysis of parameters regarding rainfall, temperature and soil moisture, we recommend to apply some technological measures which reduce the water losses from the soil and the increase of its reserve by conservative measures.

ACKNOWLEDGEMENTS

This work was supported by a grant of the Ministery of Research an Innovation, CCCDI-UEFISCDI, project number PN-III-P1-1.2-

PCCDI-2017-0056: Functional collaboration model between public research organizations and the economic environment for the provision of high-level scientific and technological services in the field of bio-economy, within PNCDI III.

REFERENCES

Arghiuș, C. (2010). Peak and Piedmont Codrului - geomorphological study. Doctoral Thesis, UBB Cluj-Napoca.

Arnoldus, H.M.J. (1980). An approximation of the rainfall factor in the Universal Soil Loss Equations. In: De Boodt, M. and Gabriels, D., Eds., Assessment of Erosion, John Wiley and Sons, New York: 127–132.

Bindi, M., Olesen, J.E. (2011). The responses of agriculture in Europe to climate change. *Regional Environmental Change, 11*(Suppl. 1), 151–158.

Blaga, I. (2013). The effect of precipitation on the morphodynamics of the mountainous and hilly area of Cluj county. Doctoral Thesis, UBB Cluj-Napoca.

Bogdan, O. (2005). Characteristics of hazards, climatic risks on the territory of Romania. *Natural and Anthropogenic Hazards, 5*(23), 26–36 .

Ciscar, J.C., Iglesias, A., Feyen, L., Szabó, L., Van Regemorter, D., Amelung, B., Nicholls, R., Watkiss, P., Christensen, O.B., Dankers, R., Garrote, L., Goodess, C.M., Hunt, A., Moreno, A., Richards, J., Soria, A. (2011). Physical and economic consequences of climate change in Europe. *Proceedings of the National Academy of Sciences of the United States of America, 108*(7), 2678–2683.

Costea, M. (2012). Using the Fournier Indexes in estimating rainfall erosivity. Case study-The Secaşul Mare Basin. Air and water components of the environment. The Cluj University Press, Cluj-Napoca, 313-320.

De Luís, M., González-Hidalgo, J.C., Longares, L.A., (2009). Is rainfall erosivity increasing in the Mediterranean Iberian Peninsula?. *Land Degradation and Development, 21*(2), 139–144.

De Martonne, E. (1926). Une nouvelle fonction climatologique: L'indice d'aridité. La Meteorologie, 449–458.

Donatelli, M., Srivastava, A.K., Duveiller, G., Niemeyer, S. (2012). Estimating impact assessment and adaptation strategies under climate change scenarios for crops at EU27 scale. In: International Environmental Modelling and Software Society (iEMSs) [Seppelt R., Voinov A.A., Lange S., Bankamp D. (eds.)], Manno, Switzerland, 404–411.

Fournier, F. (1960). Climat et Erosion. PUF, Paris.

Gabriels, D. (2006). Assessing the Modified Fournier Index and the Precipitation Concentration Index for some European Countries. Soil Erosion in Europe, John Wiley & Sons Eds.

García-Ruiz, J.M. (2010). The effects of land uses on soil erosion in Spain: a review. *Catena, 81*, 1–11.

Duda, B.M. (2018). Monitoring of the thermal and hydric regime in Someşan Plateau and the technological characterization of the land for the main agricultural crops. Doctoral Thesis, USAMV Cluj-Napoca.

Marin, D.I., Rusu, T., Mihalache, M., Ilie, L., Nistor, E., Bolohan, C. (2015). Influence of soil tillage system upon the yield and energy balance of corn and wheat crops. *Agrolife Scientific Journal, 4*(2), 43–47.

Meddi, M. (2013). Seddiment transport and rainfall erosivity evolution in twelve basins in central and western Algeria. *Journal of Urban and Environmental Engineering, 7*(2), 253–263.

Muntean, L.S, Cernea, S., Morar, G., Duda, M.M., Vârban, D.I., Muntean, S., Moldovan, C. (2014). *Fitotehnie.* Editura Risoprint, Cluj-Napoca.

Nunes, A.N., De Almeida, A.C., Coelho, C.O.A. (2011). Impacts of land use and cover type on runoff and soil erosion in a marginal area of Portugal. *Applied Geography, 31*, 687–699.

Oduro-Afriyie, K. (1996). Rainfall erosivity map for Ghana. *Geoderma, 74*, 161–166.

Povară, R., Văduva, I. (2009). Meteo-climatic risks in the Timiş Plain with impact on agriculture. Geographic Forum. *Studies and Researches of Geography and Environmental Protection, 8*(8), 82–85.

Rivington, M., Matthews, K.B., Buchan, K., Miller, D.G., Bellocchi, G., Russell, G. (2013). Climate change impacts and adaptation scope for agriculture indicated by agro-meteorological metrics. *Agricultural Systems, 114*, 15–31.

Rusu, T., Coste, C.L., Moraru, P.I., Szajdak, L.W., Pop, A.I., Duda, B.M. (2017). Impact of climate change on agro-climatic indicators and agricultural lands in the Transylvanian Plain between 2008-2014. *Carpathian Journal of Earth and Environmental Sciences, 12*(1), 23–34.

Satmari, A. (2010). *Practical applications of biogeography*. Editura Eurobit, Timişoara, 85.

Yuksel, A., Gundogan, R., Akay, A.E. (2008). Using the Remote Sensing and GIS Technology for Erosion Risk Mapping of Kartalkaya Dam Watershed in Kahramanmaras, Turkey. *Sensors, 8*, 4851-4865.

***National Strategy of Romania on Climate Change 2013-2020, Ministry of Environment and Climate Change: http://mmediu.ro/new/wp-content/uploads/2014/02/Strategia-Nationala-peSchimbari-Climatice-2013-2020.pdf

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

PEDOFUNCTIONAL EFFECTS INDUCED BY ALGALIZATION OF TYPICAL CHERNOZEM HUMUS MODERATED WITH CYANOPHYTA ALGAE NITROGEN FIXATORS UNDER IRRIGATION

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Abstract

Algalization - administration procedure in the soil of living algal cultures in order to restore trophic chains within soil microbiocenoses, which have a beneficial impact on all levels of soil ecosystem organization. At the lower levels (bio-cellular, ionic-mineral and elementary particle) involves biochemical processes and organic-mineral with the formation of active substances which determines the structural-aggregate organization of soil mass which involves several evolutionary stages: 1. Elementary particle $\frac{algae}{microbiota}$ *elementary particle → microaggregates (< 0.25 mm); 2. Microaggregates* $\frac{algae}{microbiota}$ *elementary particle → aggregate (1-0.25 mm); 3. Aggregate (1-0.25 mm)* $\frac{humic}{substances}$ *aggregate (1-0.25 mm) → aggregate 7-1 mm. Within these processes three groups of structural aggregates with different pedogenetic stability and functions are formed.*

***Key words**: algalization, structural-aggregate organization, pedogenetic stability.*

INTRODUCTION

In the literature, the place and role of algae in pedogenesis is more frequently examined in the context of the early stages of pedogenesis process. In this sense, algae are assigned the role of principal source of organic matter necessary for the operation of microbial "biocenoses". Recent research, however, has shown that, as the pedogenesis process and soils evolve the role of algae does not decrease but on contrary increases. In this context, it was established that algae directly participate in several biochemical and physical processes that occur in soils: the accumulation of organic substances and nitrogen fixation from the atmosphere, decomposition of primary and secondary minerals, redistribution and accumulation of biophile elements and aggregation - structuration. Mechanisms and quantitative expression of these processes are less studied.

This implies the need for systematic studies in the field. In light of above this paper aims to study the pedofunctional effects induced by alagalization of typical chernozem humus moderated with cynophyta algae nitrogen fixators under irrigation.

MATERIALS AND METHODS

Research was conducted during 2016-2018 in the central area of Moldova. Soils within experimental land are represented through typical chernozems humus moderated loam clay with relatively high potential for aggregation. Irrigation method used is drip irrigation.

The water source is conventionally suitable for irrigation. Experience scheme:

1. Control;
2. Algogenic preparation *Nostoc flagelliforme* (3 kg/ha);
3. Algogenic preparation *Anabaena minima* (3 kg/ha).

Structural-aggregation analysis was performed by Savvinov method. For calculating the quality indices of structural-aggregate have been applied relations:

$$Ks = \frac{A}{B} \quad (1)$$

where:

Ks - coefficient of structure;

A - aggregates content with dimensions 10-0.25 mm;

B - summary content of aggregates > 10 mm and < 0.25 mm.

$$Kafi = \frac{A}{B} \quad (2)$$

where:
Kafi - coefficient of structure;
A - aggregates content with dimensions 7-1 mm;
B - summary content of aggregates > 7 mm and < 1 mm.

$$Kc = \frac{A}{B} \quad (3)$$

where:
Kc - aggregate stability index;
A - aggregates content 1-0.5 mm fractionated in water;
B - aggregates content 1-0.5 mm dry fractionation.

RESULTS AND DISCUSSIONS

Algalization-soil administration procedure in the soil of living algal cultures in order to restore trophic chains in soil biocenosis and support for pedofunctional processes has come to the attention in the relatively recent research (Lupascu & Jigau, 1998; Jigau, 2009; Jigau et al., 2018 a; 2018 b.; Dobrojan et al., 2016).

The specified research has shown that algalization has impact on pedofunctional processes at all levels of structural-functional of soil ecosystem.

At bio-cellular level cyanophyta algae nitrogen fixators in their capacity as nucleus of cyano-bacterial coenoses lead to the formation of organisms responsible for decomposition of various groups of organic substances (geno-metabolic chains), which decompose the consecutive organic residues by enriching organic matter with nitrogen in a closed circuit of substances and energy.

At ionic-molecular level algae-bacterial coenoses creates a vital environment which favours starting the humification processes of algae-bacteria dead mass with the formation of active organic substances represented by macromolecular organic compounds and pseudohumic substances.

Elementary particle level involves organic-mineral reactions with the formation of organic-mineral compounds (Figure 1).

Processes specified quantitatively and qualitatively materialize in the “aggregate” hierarchical level, which involves two groups of structural-aggregate organization:

a) Direct - binding of elementary particles and microaggregates by means of agglutinated substances secreted in the algae activity process (gels and coagulants);
b) Indirect - are interleaved with algae activity within the carbon circuit in the system: external biocoenosis (upper plants) and internal biocoenosis (soil biota).

Through this prism of ideas cyanophyta algae provide carbon and nitrogen circuits in the soil:

1. Short duration - within biological circuit of chemical elements in the trophic chain algae - microbiota - algae.
2. Long duration - in the process of formation and humus accumulation.

The circuit ensures development of plant root system and enhancing its role in the soil mass structure with the formation of bullet aggregates (7-3 mm).

In the long duration circuit, humic substances forming with the cations of metals and clay minerals stable compounds that are associated in grain structural formations (3-0.5 mm). Within these mechanisms, the grain structural aggregates are produced in situ formed with participation of soil internal biocoenosis.

This is intensive in the layer 0-30 cm. This implies that the aggregation activity of internal biocoenosis is intercorrelated with the activity of structuration of root system plants. The interaction between plants and algae in this process has direct character algae - plants or indirect plant - algae - microbiota - algae - plant. The role of algae in this sense implies stimulation of microbial activity, part of humification process. In this respect, organic substances in the soil follows to be perceived as bio-organic substances which include non-specific organic matter (organic residues) in various decomposition phases, specific organic substances (humic substances) and microbiota associated with these two groups of substances (Jigau et al., 2018).

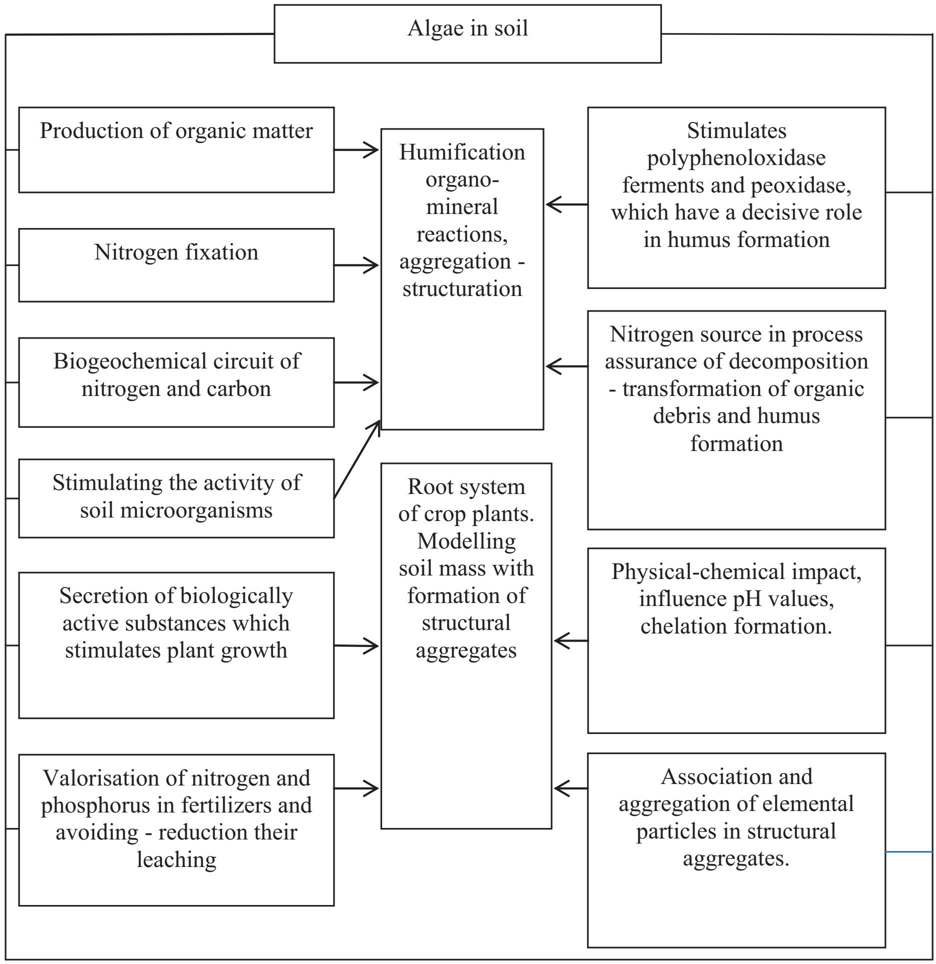

Figure 1. Place of algae in aggregation structure of soils

The biocoagulation of structural-aggregation process of pedomatrix with participation of algae includes several evolutionary stages:

1. Elementary particle $\frac{algae}{microbiota}$ elementary particle → microaggregates (< 0.25 mm);

2. Microaggregates $\frac{algae}{microbiota}$ elementary particle → aggregate (1-0.25 mm);

3. Aggregates (1-0.25 mm) $\frac{humic}{substances}$ aggregates (1-0.25 mm) → aggregates 7-1 mm.

Within these processes three groups of structural aggregates are formed:

a) Low stability - 7-5 mm;
b) With moderate stability - 5-1 mm;
c) With increased stability - 1-0.25 mm.

Through this prism of ideas aggregates > 7 mm have an abiotic origin. Their formation is determined by physico-mechanical processes.
Intensification of formation processes and humus accumulation contributes to the intensification of soil invertebrate's mesofauna biological activity, and respectively of coprogenic structuring processes, increasing over time (Table 1).

Table 1. Post-action of algal preparations on invertebrate mesophane activity in typical chernozem humus moderated (irrigated regime), mean values, layer 0-50 cm (un/m^2)

Version	Year											
	Beginning of growing season						End of growing season					
	2016		2017		2018		2016		2017		2018	
	Copr	Cerv	Copr	Cerv	Copr	Cerv	Copr	Cerv	Copr	Cerv	Copr	Cerv
Control	29	37	28	38	27	35	33	42	36	41	33	39
Nostoc flageliforme	32	39	33	43	35	45	39	49	40	52	47	58
Anabaena minima	36	43	35	41	37	44	65	77	67	80	76	94

According to Kovda (1973) the formation and reproduction of humus reserves in soil involves intercalated action mechanisms of invertebrate's mezofauna and soil microbiomes. The earthworms assimilate preferentially soluble humic substances and slightly subject to mineralization of organic matter in various stages of decomposition and forming coprolites enriched in carbon and nitrogen. This is due the overlap, in time of biological activity of earthworms and microbiomes responsible for formation processes and humus accumulation: during the active nutrition of earthworms in soil, microbiological activity is intensified. This leads to the formation of intermediate products which are framed in the resynthesis process and humification.

The data presented in Table 1 highlights the intensification of intervertebrate mesofauna activity both in the administration of *Nostoc flagelliforme* culture as well as in the case of culture with *Anabaena minima*. An increase in time of this index allows us to consider the soil slowly the trophic chains are restored algae microbiomes materialized in the intensification of formation and humus accumulation. The decisive role in the structuration - aggregation of soil mass returns to biocoagulant processes determinate intercalating action of root system and coagulation processes.

From Table 2 we find that in the early stages of growing season agronomical valuable aggregate content (10-0.25 mm) in all three investigates variants is excellent. However, already at this stage we find the positive influence of algae microflora on the structural-aggregation process. The maximum effect of this process is within *Nostoc flagelliforme* version, both with aggregate content 10-0.25 mm and 5-1 mm. During vegetation, this variant stands out strongly by the Control version through content of these groups of aggregates. The effects of aggregation structure determined by *Anabaena minima* species are later; these are intensifying over time and presenting excellent values at the end of growing season. In the *Nostoc flagelliforme* version in the aggregates composition 10-0.25 mm predominates 5-1 mm. Their content makes up 54.32 % of soil mass or 64.47 % of agronomic valuable aggregates. In the case of *Anabaena minima* version the aggregate content 5-1 mm does not differ essentially from the Control variant.

Table 2. Dynamics of structural quality indices of typical chernozem humus moderated under algalization conditions (Irrigated regime) (Mean values, layer 0-50 cm)

Terms of sampling	Version	Agronomical valuable aggregate content (10-0.25 mm)	Precious agronomic aggregate content (5-1 mm)	Coefficient of structure (Ks)	Coefficient of structure (Kafi)	Aggregate stability index (Kc) %
May	Control	80.34	46.54	4.38	1.35	318
	Nostoc flagelliforme	85.58	54.32	8.12	2.05	492
	Anabaena minima	87.73	46.42	5.71	1.45	347
June	Control	72.33	39.56	3.95	1.37	426
	Nostoc flagelliforme	80.06	50.33	4.63	1.68	542
	Anabaena minima	73.50	42.73	2.96	1.23	411
July	Control	72.07	37.85	3.30	1.13	396
	Nostoc flagelliforme	79.75	47.91	4.44	1.16	458
	Anabaena minima	77.13	41.85	4.03	1.18	387
August	Control	71.80	36.14	2.65	0.88	306
	Nostoc flagelliforme	79.45	45.49	4.24	1.09	375
	Anabaena minima	80.76	40.98	5.09	1.13	363

Table 3. Dynamics of aggregate content > 7 and 7-1 mm in the aggregate composition of typical chernozem humus moderated under algalization conditions (Irrigated regime) (Mean values, layer 0-50 cm)

Terms of sampling	Version	Aggregate content > 7 mm	Aggregate content 7-1 mm	
			7-1 mm	% from ∑ 10-0.25 mm
May	Control	28.39	58.95	73.21
	Nostoc flagelliforme	24.02	64.89	75.82
	Anabaena minima	26.04	58.41	72.30
June	Control	28.19	55.43	70.44
	Nostoc flagelliforme	21.65	60.23	74.86
	Anabaena minima	31.55	53.29	72.57
July	Control	29.50	50.87	69.65
	Nostoc flagelliforme	20.82	54.43	71.37
	Anabaena minima	30.05	53.09	69.08
Augu st	Control	30.81	46.31	68.85
	Nostoc flagelliforme	19.99	55.57	67.88
	Anabaena minima	28.55	52.89	65.59

During the vegetation period in all variants in the structural - aggregate composition predominated 7-1 mm. With maximum capacity of their formation is characterized
Nostoc flagelliforme where their content in the first half of vegetation periods makes up (75.82-74.84 %).

Anabaena minima version through the content of these aggregates differs insignificantly from Control version. In the second half of vegetation period, aggregate content 7-1 mm is reduced to all variants. However, for algalized variants their content is higher than Control version. This allows us to conclude that aggregates 7-1 mm formed with participation of cyanophyta algae have greater aggregate stability. In support of this assertion it's coming Aggregate Stability Index (Kc) and dynamic indices of aggregate stability (Table 4).

From the last we find that in case of algalized versions there are attested hydrostatic aggregates with a diameter 7-5 mm throughout the whole vegetation period, their content, however, ranging from 2-4%.

As the aggregate diameter is reduced their hidrostability increases in all variants.

Table 4. Dynamics of aggregate stability of typical chernozem humus moderated under algalization conditions (Irrigated regime) (Mean values, layer 0-50 cm)

Terms of sampling	Version	Hydrostabile aggregate content, %				
		Aggregate content. mm				
		7-5	5-1	1-0.25	> 0.25	< 0.25
May	Control	-	17.34	41.43	58.77	41.23
	Nostoc flagelliforme	3.62	18.19	42.13	63.30	36.70
	Anabaena minima	3.33	19.74	43.32	65.45	34.55
June	Control	-	17.82	32.48	51.56	48.44
	Nostoc flagelliforme	3.89	20.94	47.28	72.11	27.89
	Anabaena minima	2.36	20.12	38.08	60.56	39.44
July	Control	-	15.31	32.61	47.92	52.08
	Nostoc flagelliforme	2.33	25.73	44.31	72.13	24.87
	Anabaena minima	2.32	19.12	39.76	31.20	38.80
August	Control	-	16.94	32.16	49.10	50.90
	Nostoc flagelliforme	2.23	27.53	45.81	75.57	24.43
	Anabaena minima	1.97	18.83	40.51	62.31	37.69

With maximum hidrostability is characterized aggregates 1-0.5 mm which make up the active reserves for aggregation process. Based on their content we find that maximum structural potential characterizes *Nostoc flagelliforme*.

CONCLUSIONS

Algalization intensifies the entire chain processes and soil functioning mechanisms which materializes in the aggregation of soil mass structure. It is necessary to distinguish two stages intercalated:

Processes and mechanism carried out at lower levels (bio-cellular, ionic-molecular, elementary particle) to organize the soil ecosystem with the formation of active substances;

Processes and mechanisms of structural-aggregate organization.

Structural aggregation with the participation of algae leads to the formation of three groups of structural aggregates with aggregate stability and different pedofunctional functions.

REFERENCES

Dobrojan, S., Salaru, V., Melnic, V. (2016). *Cultivarea algelor*. CEP USM, 172.

Jigau, G. (2009). *Geneza si fizica solurilor*. CEP USM, 164.

Jigau, G., Salaru, V., Dobrojan, S., Tofan, E., Blidari, A., Placinta, N. (2018a). Place of cynophyta algae in support and achievement of the structural – aggregation processes of humifer moderated typical chermozems. *Factori si Procese Pedogenetice din Zona Temperata, Iasi, Al. I. Cuza University.*

Jigau, G., Salaru, V., Blidari, A., Dobrojan, S., Tofan, E., Gatman G., Placinta, N. (2018). Vision on the structural – aggregation mechanisms with the participation of cyanophyta alegae. *Seria Agronomie, Iasi.*

Lupascu, G., Jigau G., Verlan, M. (1998). *Pedologie Generala*. Iasi, 298.

Kovda, V.A. (1973). *Osnovi ucenia o pocvah*. Moskva: Nauka, 468.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF *NO-TILLAGE* PRACTICES AND GREEN MANURE TREATMENT ON SOIL BIOTA OF THE ORDINARY CHERNOZEM IN THE SOUTHERN ZONE OF THE REPUBLIC OF MOLDOVA

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Abstract

Statistical parameters of the biological properties of ordinary chernozem under different land management in the southern zone of the Republic of Moldova have been elaborated. The number of Lumbricidae family and total invertebrates in conditions of no-tillage and vetch as a green manure increased on average by 3.0-6.0 and by 2.2-2.5 times respectively compared with plowing. The base mass of invertebrates and earthworms in the ordinary chernozem under arable is concentrated in the 0-10 cm layer, in conditions of no-tillage and green manure treatment - in the 0-20 and 0-30 cm layer. The application of conservative tillage and green fertilizers led to the restoration of the total number of invertebrates and the Lumbricidae family, the annual growth rate is of 36.1 and 12.4 ex m^{-2} accordingly. The positive action of no-tillage and vetch as a green fertilizer on biota is manifested by the growth of microbial biomass on average by 13.9% and activation of dehydrogenase, polyphenoloxidase and peroxidase by 15.3%, 28.9% and 6.4%. Soil biological parameters did not reach the level of the soil under natural vegetation.

Key words: *biota, parameters, ordinary chernozem, no-tillage, green manure.*

INTRODUCTION

The soil cover of the Republic of Moldova is subjected to processes of desertification and anthropic degradation (Cerbari et al., 2010). The biological degradation of arable soils is interconnected with the dehumification processes, compaction and destruction of the soil structure. The current state of the biota in the arable soils of the Republic of Moldova is characterized by a significant reduction of abundance, biomass, activity and diversity, as compared to the soils of standard natural ecosystems (Senicovscaia et al., 2012). The values of most biological indicators in zonal soils decrease in the following sequence: natural and long-term fallow land → arable land under ryegrass and the organic farming system with the incorporation of manure and crop residues → arable land without fertilizers.

A major reason for the deterioration of soil biological properties and for the decrease of humus content under arable agriculture is annual tillage, which aerates the soil and breaks up aggregates where microbes are living. Previous research has demonstrated that the conventional tillage system leads to structural aggregates destruction in the soil, and therefore of biota' habitat (Senicovscaia et al., 2008). Annual plowing destroys agronomically valuable aggregates in the top layer of chernozems and their hardening in big sized soil particles in underlying 10-20 cm layer, thereby forming the soil compaction. The microbial biomass content in soil aggregates decreases according to the same regularity (Lungu et al., 2017).

The negative effects of conventional tillage have led to necessity of application of alternative soil management practices. These techniques are known as *"conserving/preserving tillage"*. It should be noted that the concept of conservation processing includes many processes from direct seeding and *no-tillage* to the deep cultivation without turning of soil layers. The implementation of soil tillage technologies should be based on the detailed knowledge of characteristics of soil. Necessarily should be taken into account such important factor as soil biological properties in cases when soil conservation tillage is applied.

Previous studies have demonstrated the effectiveness of *no-tillage* technology in

conditions of the northern zone of the Republic of Moldova (Senicovscaia et al., 2015).
The way to support the functioning of soil biota, to increase the level of biodiversity and resistance of soils used for a long time as arable is the cultivation of leguminous cultures as a green manure (Senicovscaia, 2013). This practice has several positive aspects from the microbiological point of view. It provides nutrient-rich organic matter for the microbial community which easily converts organically bound nutrients in plant residues to easily available nutrient form to the crops. Green manure application enhances the biodiversity of soil microorganisms. Green manuring with legumes has added advantage due to the capacity of legumes to fix atmospheric nitrogen and to decompose easily. Presumably this technique will improve the functioning of edaphic fauna.
In connection with foregoing, the evaluation of the biota recovery process in degraded chernozems, including *no-tillage* soil conservative system and the application of leguminous crops as a green fertilizer, is an actual problem.
Such research corresponds to the tasks of soil bio-testing for biodiversity conservation and development of soil quality standards.
The purpose of the research was to determine the influence of *no-tillage* practices and green manure application on biota of the ordinary chernozem for the improvement of its soil quality and environmental certifications.

MATERIALS AND METHODS

Site. The experimental site is located in the southern zone of the Republic of Moldova, in Natcubii-Agro SRL, Larga Noua village, Cahul region. Agricultural lands are located on the quasi-horizontal surface of the highest terraces of the Prut River. The absolute altitude of the site is about 120-121m.
The technology of *no-tillage* has been implemented on the area of about 2.000 hectares during 2 years before the application of green manure treatment.
Vetch was used twice as a green manure. Vetch was planted in the September of 2014 and its green mass in the amount of 26 t ha^{-1} was plowed under disc in May, 12, 2015 (Figure 1). On the same plot, a vetch was sown again, and its green mass was introduced in the amount of 17 t ha^{-1} into the soil by disking in September, 30, 2015. The experimental plot occupied 0.15 ha.

Figure 1. Field with a vetch in the southern zone of the Republic of Moldova, in Natcubii-Agro SRL, Larga Noua village, Cahul region

Land management practices with application of *no-tillage* with green manure treatments (plot 3) have been compared with the long-term conventional tillage with plowing to a depth of 25-27 cm (plot 1) and 55-year-old fallow plot.
Soil. The soil is the ordinary chernozem, subjected to deep plowing 40 years ago and used for plowing in the next 20 years. The profile of the arable chernozem demonstrates the strong compaction of the arable layer in depths of 5-20 cm and massive structure (Figure 2, a). As a result of the incorporation into the soil of the green mass of vetch (Figure 2, b), the physical quality of the layer 0-20 cm has evolved from the unfavorable to very favorable. The soil penetration resistance has become low and extremely low and the structure turned out to be agronomically favorable.

a b

Figure 2. The profiles of ordinary chernozem in conditions of long-term conventional tillage (a) and *no-tillage* with green manure treatments (b)

The initial values of the humus content in the soil of plot 1 constitute 2.43 %, pH = 7.9, the content of mobile phosphorus - 1.09 mg 100 g^{-1} soil, mobile potassium - 17.7 mg 100 g^{-1} soil, nitrates ($N\text{-}NO_3$) - 0.24 mg 100 g^{-1} soil in the 0-35 cm layer. The humus content in the soil of plot 3 before green manuring constitutes 2.49%, pH = 8.0, the content of mobile phosphorus - 1.47 mg 100 g^{-1} soil, mobile potassium - 19.7 mg 100 g^{-1} soil, nitrates ($N\text{-}NO_3$) - 0.24 mg 100 g^{-1} soil in the 0-35 cm layer.

Soil samples were collected from the 0-10, 10-20 and 20-30 cm layers of the experimental plots during 2015-2018.

Status of invertebrates was identified from the test cuts by manual sampling of the soil layers to the depth of soil fauna occurrence. The diversity at the family level was categorised according to Gilyarov and Striganova (1987).

Microbiological properties. The microbial biomass carbon was measured by the rehydration method (the difference between C extracted with 0.5 M K_2SO_4 from fresh soil samples and from soil dried at 65-70°C for 24 h (Blagodatsky et al., 1987). K_2SO_4 - extractable organic carbon concentrations in the dried and fresh soil samples were measured simultaneously by dichromate oxidation. K_2SO_4-extractable carbon was determined at 590 nm using a "СФ-103" spectrophotometer. To estimate the contribution of microbial carbon to the total content carbon, the ratio between the carbon of the microbial biomass and the total carbon expressed in % was used (Kennedy & Papendick, 1995). Reserves of microbial biomass have been calculated taking into account the carbon content of the microbial cell and the bulk density of soils.

Enzymatic activity. The potential enzymatic activity was determined in samples of the air-dry soil. The urease activity was measured by estimating the ammonium released on incubation of soil with buffered urea solution by colorimetrical procedure (Haziev, 2005). The dehydrogenase activity was determined by the colorimetric technique on the basis of triphenylformazan (TPF) presence from TTC (2,3,5-triphenyltetrazolium chloride) added to the soil (Haziev, 2005). The polyphenoloxidase and peroxidase activities were determined by the colorimetric technique using hydroquinone as a substrate (Karyagina & Mikhailovskaya, 1986).

Soil biological indices were evaluated by analysis of variance. Statistical parameters of soil invertebrates were calculated taking into account the depth of soil fauna occurrence, microorganisms and enzymes - for the layer of 0-30 cm.

RESULTS AND DISCUSSIONS

Invertebrates. Application of *no-tillage* with vetch as a green fertilizer has a favorable effect on the edaphic fauna of the ordinary chernozem. This effect has been noted both by the average values of indicators and by the confidence intervals. The number of invertebrates increased on average from 21.7 to 130.1 ex. m^{-2}, the number of earthworms - from 18.5 to 55.6 ex m^{-2} in comparison with the plot with arable tillage. The biomass of invertebrates rose from 4.4 to 10.8 ex m^{-2}, the biomass of *Lumbricidae* family - from 4.3 to 9.3 ex m^{-2} (Table 1).

The dominant position in the complex of invertebrates occupies the *Lumbricidae* family. The share of earthworms in the total abundance of invertebrates in the arable soil constitutes 85.3 % and their biomass - 97.7%. The share of *Lumbricidae* family in the total population on the plot with conservation tillage and application of green manure constitutes 42.7%, their biomass - 86.1%. These data indicate that when *no-tillage* and green fertilizer were used, other species appear in the complex of edaphic fauna of the ordinary chernozem.

The average weight of one exemplar of *Lumbricidae* family in the arable soil constitutes from its total absence to 0.24 g, on the plot with *no-tillage* and green manure - 0.20-0.33 g. The base mass of invertebrates (80.0%) and fam. *Lumbricidae* (88.0%) in the ordinary chernozem under arable is concentrated in the 0-10 cm layer according to the average data. When green fertilizers were applied together with the *no-tillage* soil conservation system, the most invertebrates (93.9%) are concentrated in the 0-20 cm layer and earthworms (93.2%) - in the 0-30 cm layer (Figures 2, 3). Chernozem in conditions of *no-tillage* and green manure management is characterized by an active thick layer of soil.

Table 1. Statistical parameters of biota in the ordinary chernozem under different land management in the southern zone of the Republic of Moldova (2015-2018)

Index	Plot 1, arable 25-27 cm					Plot 3, *no-tillage* and green manure					55-year-old fallow plot
	mean value	min	max	V, %	confidence interval ($P \leq 0.05$)	mean value	min	max	V, %	confidence interval ($P \leq 0.05$)	mean value (n=3)
Invertebrates (n=7)											
Number of invertebrates, ex m^{-2}	21.7	0	48.0	75.9	6.5-36.9	130.1	32.0	272.0	78.6	35.4-224.8	448.0
Number of *Lumbricidae* fam., ex m^{-2}	18.5	0	40.0	83.5	4.2-32.8	55.6	16.0	96.0	54.3	27.6-83.6	340.0
Biomass of invertebrates, g m^{-2}	4.4	0	10.0	92.4	0.6-8.2	10.8	7.0	19.2	41.1	6.7-14.9	84.0
Biomass of *Lumbricidae* fam., g m^{-2}	4.3	0	9.6	93.2	0.6-8.0	9.3	5.2	18.8	54.6	4.6-14.0	74.8
Microorganisms (n=33-45)											
Microbial biomass, µg C g^{-1} soil	204.1	144.1	258.7	16.6	192.1-216.1	232.4	126.9	486.9	43.9	201.8-263.0	415.6
Enzyme activity (n=21-45)											
Urease, mg NH_3 10 g^{-1}soil 24 h^{-1}	3.9	2.9	5.7	22.6	3.3-4.5	3.8	2.8	5.3	17.4	3.5-4.1	5.5
Dehydrogenase, mg TPF 10 g^{-1} soil 24h^{-1}	1.57	0.95	2.35	23.9	1.43-1.71	1.81	0.96	3.53	34.7	1.63-1.99	2.79
Polyphenoloxidase, mg 1,4-p-benzoquinone 10 g^{-1} soil 30 min^{-1}	18.0	6.5	28.0	35.6	15.7-20.3	23.2	12.5	31.0	25.0	21.5-24.9	20.1
Peroxidase, mg 1,4-p-benzoquinone 10 g^{-1} soil 30 min^{-1}	28.3	23.0	34.0	11.1	27.1-29.5	30.1	24.5	36.0	11.2	29.0-31.2	31.3

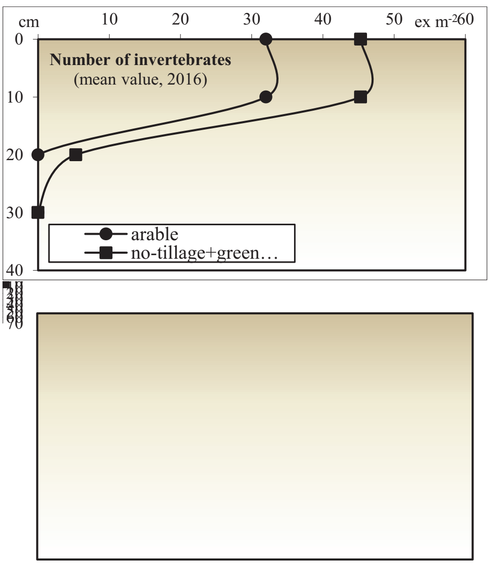

Figure 2. Profile distribution of invertebrates in the

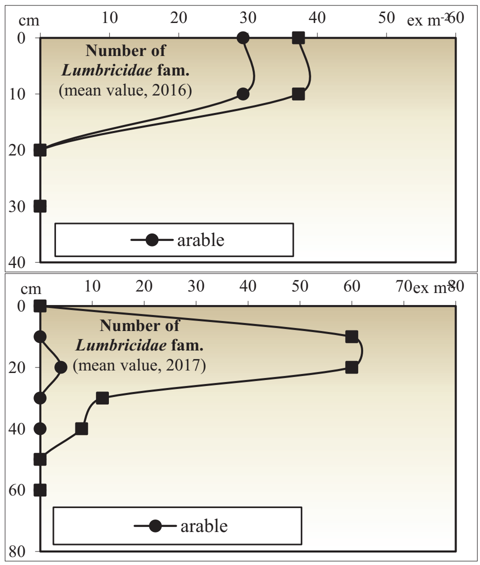

Figure 3. Profile distribution of earthworms in the ordinary chernozem under different land management

Ordinary chernozem in conditions of *no-tillage* and green manure is characterized by a high diversity of invertebrates in comparison with the soil under arable.

This soil contains 4-8 families of invertebrates, whereas under *arable* only 3-5 families depending on the year of sampling (Table 2).

Lumbricidae family is prevalent in soils with both types of tillage. *Lumbricus terrestris* was a typical representative of *Lumbricidae* family. In addition to *Lumbricidae* family, other species of the *Enchytraeidae, Elateridae, Julidae, Araneidae, Carabidae, Coccinellidae* and *Formicidae* families in faunal samples have been identified.

Table 2. Biodiversity of invertebrates (ex m^{-2}) in the ordinary chernozem under different land management (mean value, 16.05.2018)

Invertebrate family	Plot 1, arable 25-27 cm	Plot 3, *no-tillage* and green manure
Lumbricidae	34.6	24.0
Enchytraeidae	0	5.3
Elateridae	5.3	2.6
Julidae	5.3	8.0
Araneidae	0	2.7
Scarabaeidae	10.7	0
Carabidae	2.7	2.7
Coccinellidae	0	8.0
Geophilidae	0	0
Formicidae	0	+
Specii neidentificate	5.4	0

Indices of invertebrates' number and biomass in the soil of both plots are characterized by the strong variability. There is a tendency of decrease of the variation coefficient from the arable soil (75.9-93.2 %) to the soil under *no-tillage* and green manure (41.1-78.6 %) by faunal indicators. Taking into consideration the increase in the total level and decrease in the amplitude of the biological parameters oscillations in soil with *no-tillage* and green manure, this testifies to the enhance of soil's ecological stability against anthropogenic impacts.

Microorganisms. The application of *no-tillage* technologies and vetch stimulates the restoration of microorganisms in the ordinary chernozem. According to statistical data, the use of *no-tillage* with green manure lead to the slight increase of microbial biomass content on average from 204.1 to 232.4 μg C g^{-1} soil (Table 1). Indices of microbial biomass in the soil of both plots are characterized by the medium and strong variability.

The main changes have been occurred in the soil layer of 0-20 cm. Microbial biomass increased from 219.3 μg C g^{-1} soil in the arable soil to 276.3 μg C g^{-1} soil in conditions of *no-tillage* and green manure (Table 3).

The share of microbial carbon in the total carbon in the plot under arable constitutes on average 1.55% and in the plot with *no-tillage* and green manure - 1.67%.

Table 3. The content and reserves of microbial biomass in the ordinary chernozem in conditions of different land management

Plot, variant	Depth, cm	Microbial biomass, μ g C g^{-1} soil	C_{MB}/C_{org}, %	Reserves of MB, kg ha^{-1}	
				in layers	in the 0-20 cm layer
25.04.2016					
Plot 1, arable 25-27 cm	0-10	235.1	1.66	573.6	1147.3
	10-20	235.1	1.66	573.7	
Plot 3, *no-tillage* and green manure	0-10	310.2	1.85	732.1	1414.3
	10-20	281.9	1.71	682.2	
29.05.2017					
Plot 1, arable 25-27 cm	0-10	216.4	1.52	528.0	1056.0
	10-20	216.4	1.52	528.0	
Plot 3, *no-tillage* and green manure	0-10	225.0	1.34	558.0	1240.9
	10-20	271.0	1.61	682.9	
16.05.2018					
Plot 1, arable 25-27 cm	0-10	194.2	1.35	431.1	1012.8
	10-20	218.7	1.56	581.7	
Plot 3, *no-tillage* and green manure	0-10	397.9	2.41	970.9	1409.7
	10-20	171.4	1.09	438.8	

The application of *no-tillage* contributes to the increase of microbial biomass reserves from 1072.0 to 1355.0 kg ha^{-1} in the 0-20 cm layer. Reserves of microorganisms' biomass in the 0-30 cm layer are 1492.7-1862.7 kg ha^{-1} in the plot with plowing and 1678.7-2012.2 kg ha^{-1} in the plot with *no-tillage* and green manure.

Enzymatic activity. A soil management with the application of *no-tillage* and the introduction of green fertilizers created favorable conditions for the improvement of the enzymatic activity in the soil. Dehydrogenase activity in the ordinary chernozem increased on average by 1.2 times and polyphenoloxidase activity - by 1.3 times under the influence of *no-tillage* and green fertilizers (Table 1). In contrast, activities of urease and peroxidase on average have not shown statistically significant changes. Although in some periods the activity of urease and peroxidase on the plot with conservation technology was significantly higher than on the plot with arable tillage. The strongest impact on enzymes was recorded in the 0-20 cm layer.

Urease, dehydrogenase and peroxidase indices are characterized by the medium variability, polyphenoloxidase - by the strong variability.

The variation coefficients of the urease and polyphenoloxidase activities in the chernozem under conservation tillage and green manure are lower than in the arable chernozem.

CONCLUSIONS

The application of *no-tillage* practices with the introduction of green fertilizers in the southern zone of the Republic of Moldova improves the conditions for functioning of biota in the ordinary chernozem at the level of high values of parameters. The number of invertebrates increases on average by 6.0 times, *Lumbricidae* family - by 3.0 times, biomass - by 2.5 and 2.2 times as compared with conventional tillage. The annual growth rate of invertebrates and earthworms constitutes 36.1 and 12.4 ex m^{-2} accordingly. Biomass of microorganisms increases on average by 13.9%, dehydrogenase, polyphenoloxidase and peroxidase activities - by 15.3%, 28.9% and 6.4% in comparison with the arable plot. The base mass of invertebrates and fam. *Lumbricidae* in conditions of *no-tillage* and green manure treatment is concentrated in the 0-20 and 0-30 cm layers, maximum values of microbial biomass and enzyme activity have been recorded in the soil layer of 0-20 cm. Biological indications show a tendency to decrease the coefficient of variation from the arable soil to the soil under application of *no-tillage* with vetch as a green fertilizer. This indicates an improvement of soil's ecological stability to anthropogenic impacts and increases the homogeneity of the topsoil, although biological parameters do not reach the level of soils under the natural vegetation.

REFERENCES

Blagodatsky, S.A., Blagodatskaya, E.V., Gorbenko, A.J., Panikov, N.S. (1987). Rehydration method for the determining of the microbial biomass in the soil. *Pochvovedenie*, *4*, 64–71. Moscow.

Cerbari, V. (coord.) et al. (2010). Monitoring soil quality of the Republic of Moldova (database, conclusions, prognosis, recommendations). Kishinev: Pontos.

Gilyarov, M.S., & Striganova, B.R. (1987). Quantitative methods in soil zoology. *Nauka*, Moscow.

Haziev, F.H. (2005). Methods of soil enzymology. Russian Academy of Sciences, Ufa.

Karyagina, L.A., & Mikhailovskaya, N.A. (1986). Determination of polyphenoloxidase and peroxidase activities in the soil. *Journal of the Academy of Sciences of BSSR, 2,* 40– 41.

Kennedy, A.C., & Papendick, R.I. (1995). Microbial characteristics of soil quality. *J. Soil and Water Conservation*, *50*(3), 243–248.

Lungu, M., Senicovscaia, I., Ciolacu, T. (2017). Soil microbial biomass content into leached chernozem structural aggregates in different lands management systems. *Scientific Papers. Series A. Agronomy, LX*, 119–124.

Senicovscaia, I. (2013). Restoration of biota in arable chernozems by green manuring and perennial grasses. *Scientific Papers, Series A. Agronomy*, *LVI*, 86–91.

Senicovscaia, I., Marinescu, C., Balan, T., Cerbari, V., Boincean, B. (2008). Biota and its interdependence with the physical properties of typical chernozems under different agricultural conditions. *Bulletin of the ASM. Life sciences*, *305*(2), 170–179. Kishinev.

Senicovscaia, I. et al. (2012). Methodological instructions on the assessment and increase of the soil biota stability in conditions of the degradation processes intensification. Pontos, Kishinev.

Senicovscaia, I., Filipchiuk, V., Tsintsari, A. (2015). Impact of different soil tillage practices on biota of chernozems in the northern zone of the Republic of Moldova. *Scientific papers. Series A. Agronomy, LVIII*, 95–100.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE EFFECT OF SOME *ALS* INHIBITING HERBICIDES IN CHAMBIC CERNOZEM SOIL

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Abstract

The herbicides with ALS (inhibiting the enzyme acetolactate synthase) are rather unique in their ability to control weeds through both foliar uptake and through root uptake as they are also biologically active in soil. This provides control of both emerged weeds and those that emerge after time of application. Other benefits include low application rates and low mammalian toxicity. Carryover of a herbicide beyond the year of application can be of benefit in controlling weed growth in subsequent years, but can also be of concern in causing injury to sensitive crop species that may be grown in the years following application. The paper present the results concerning the study on the residual effect of some herbicides (inhibiting the enzyme acetolactate synthase - ALS) applied for weed control in sunflower crops in relationship with the climatic conditions. The phytotoxicity and persistence of the herbicides in soil was assessed using the mustard root inhibition bioassay. In earlier stage of mustard vegetation the treatments withimazamox herbicide had a residual effect, but over time this effect was significantly reduced. The chemical composition of the soil was not affected by the applied herbicides. The results indicate that for wheat was not residual effect for any of treatments used.

***Key words**: herbicides with ALS, residual effect, phytotoxicity, mustard, bioassay.*

INTRODUCTION

Acetolactate synthase (ALS)-inhibiting herbicides have been used extensively in agricultural production mainly because of their remarkable efficacy at very low application rates. However, it has been recognized that the ALS-inhibitors are the most resistance-selective herbicide group. ALS-herbicides were first introduced in the early 1980s, and since then, rapid increase in incidence of resistance to these herbicides has been reported; more weeds have become resistant to ALS-inhibiting herbicides than to any other herbicide mode of action (Whitcomb, 1999).

The rate of pesticide breakdown in field conditions depends on soil moistureand temperature, which are very important factors in determining the rate of pesticide breakdown. There are different mechanisms which determine the environmental fate of a herbicide such as volatilization, breakdown from sunlight (photolysis), leaching etc. However, the two main mechanisms of herbicide degradation are microbial and chemical hydrolysis. These two processes are dependent on soil water and temperature. However, soil moisture is more important for herbicides that require microbes for its degradation (Streck, 2005). Soil microbesthrive in warm soils, which results in faster degradation. It is estimated that there is a two to three-fold increase in chemical half life with a 10°C decrease in temperature (Walker, 1987).

In field conditions, soil properties such as organic matter content, soil moisture, soil texture, and soil pH play also an important role in the carryover potential ofresidual herbicides (Walker, 1987). With Clearfield® technology, herbicide adsorption to organic matter may reduce its bioavailability and the moisture holdingcapacity of high organic matter soils makes them conducive to increased microbialactivity. The importance of soil organic matter in reducing carryover potential hasbeen shown in studies conducted on sulfosulfuron and flucarbazone (Moyer & Hamman, 2001; Eliason et al., 2002). The effect of clay content on herbicide residues is similar to organic matter in that it tends to adsorb the herbicide as well as improve the water holding capacity. Soil pH is another factor affecting the residual characteristics of some herbicides in field conditions. A low soil pH (less than 7.0) tends to increase the persistence of imidazolinone herbicides such as imazethapyr, imazamethabenz, and imazamox. Imidazolinone herbicides tend to be more adsorbed under acidic (lowsoil pH) conditions, which reduces their availability for microbial degradation (Loux & Reese, 1992). Extended carryover of imidazolinone herbicides in acidicsoils may also be related to their sorption-desorption characteristics.

ALS-herbicides inhibit biosynthesis of branched-chain amino acids and affect primarily root growth of susceptible plants through inhibition of cell division at the root tips. Therefore, measuring root length reduction of sensitive plant species is the most common detection approach used in bioassays for ALS-inhibiting herbicides.

Benefits of the bioassay are that whole-plant bioassays show biological effects of herbicides present, often at levels below chemical detection thresholds. They can be more useful than chemical detection methods due to interactions with soil organic matter, pH, soil moisture and soil texture. Finally, it is the only risk management tool available to growers at this time.

The objective of this study was to determine using bioassay, the persistence of imazamox herbicide inhibiting the enzyme acetolactate synthase (ALS) (applied for weed control in sunflower crops) in relationship with the climatic conditions and different soil caracteristics.

MATERIALS AND METHODS

The experience was carried out at SC Profarma Holding SRL from Fundulea (situated in south-east part of Romania) in the years 2017 and 2018 under non-irrigated conditions on cambic chernozem soil**.**

The experiment was designed by the block method, in three replications, size of the experiment plot was 25 m^2.

Treatments with herbicides based on imazamox were applied to sunflower using technologies and specific resistant hybrids**.**

Soil samples were collected after crop harvest, in three repetitions, from 0-20 cm depth of soil, (Table 1)**.**

To estimate the persistence in soil of studied herbicides was used the bioassay method developed by Eliason et al. (2004) and modified by Petcu et Oprea (2013). It was used mustard (plant sensitive to studied herbicides) and winter wheat (because is known that wheat is sown frequently after sunflower). Seeds were sown at a depth of 2 cm every 2.5 cm along the pot wall. Six seeds of similar size were selected and placed in transparent pots filled with 150 g soil (Figures 1, 2).

Table 1.Calendar and schedule of soil sampling

Date of harvesting soil samples	Treatments
October 12, 2017	**C -** Control, no herbicide
	T1- Pulsar ® 40, dose of 1.2 l/ha
	T2- Generic dose of 1,2 l/ha
	T3- Pulsar Plus, dose of 1.6 l/ha
	T4 - Pulsar Plus, dose of 2 l/ha)
October 14, 2018	**C -** Control, no herbicide
	T1- Pulsar ® 40, dose of 1.2 l/ha
	T2- Generic dose of 1,2 l/ha
	T3- Pulsar Plus, dose of 1.6 l/ha
	T4 - Pulsar Plus, dose of 2 l/ha)

The seeds were carefully placed vertically, embryo downwards and facing the wall to facilitate root growth along the transparent wall.

After sowing, the clear pots were wrapped in aluminum foil and placed in dark-colored paper bags to exclude light from the developing.

The plants were watered with the same amount of tap water avoiding dry and kept at a room temperature of 20-25°C.

Figure 1. Experience with mustardin the firs days

Figure 2. Experiences with mustard

On the ten day after seeding the plants were manually removed from the soil (Figure 3) and the length of root and shoot was measured.

The reduction rate was calculated according to the formula:

RL = (1-Lt/Lo)*100, where:

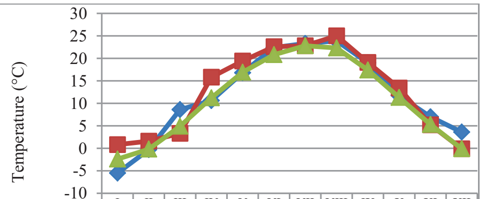
Lt is root length from soil with treatments and Lo is root length from control soil.

Figure 3. Winter wheat plants removed from soil (pots)

RESULTS AND DISCUSSIONS

The average of temperature during April and May 2018 exceeded the normal of the zone with 4.6°C and 2.5°C respectively, while in 2017 in the same period the temperatures were lower by 0.6 and 0.2°C, respectively.

On average, temperatures in 2018 exceeded normal area by 1.6°C and in 2017 only 0.9°C (Figure 4).



	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
2017	-5,5	-0,3	8,6	10,6	16,8	22,4	23,3	23,9	19	11,7	7	3,6
2018	0,8	1,6	3,3	15,8	19,4	22,6	22,8	25	19,1	13,4	5,2	-0,1
MAA	-2,4	-0,2	4,9	11,2	16,9	20,8	22,8	22,3	17,4	11,3	5,3	-0,1

Figure 4. Monthly average temperature (2017, 2018) and multiannual average temperatures (1960 - 2018)

The years of experimentation were totally different from the viewpoint of quantity and monthly repartitions of rainfall.

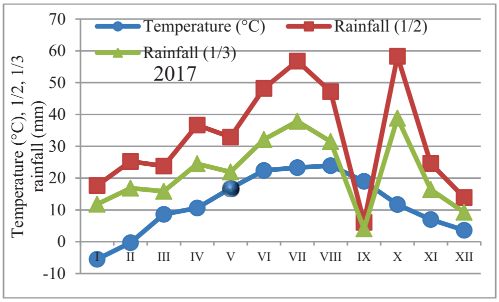


Figure 5. The climate diagram for 2017 (the method for the climate diagram consists in the graphical representation of 1/2 and 1/3 of the total value of recorded rainfall and average temperature)

In 2017 was a moisture deficits in September, as shown in Figure 5. As compared cu 2017, the year 2018 was a year with two water deficits.

First moisture deficits was from April up to May and the second ones was from august to middle of octomber and both of them were combinated also with heat stress (Figure 6).

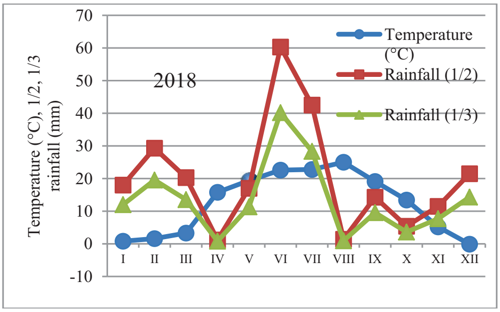


Figure 6. Climato diagram for 2018

Analysis of variances evidentiate that length of mutard roots was very significantly ($P < 0.001$) affected by year of soil sampling and significantly ($P < 0.5$) for the treatments (Table 2).

Table 2.Analyses of variance for length of mutard root

Source of variance	Degree of freedom	Mean square	S2	F factor and significance
Factor A (year)	1	2707	2707	243.9***
Error	2	22.2	11.1	
Factor B (treatment)	4	1065	266	7.37*
A x B	4	321	80	2.22
Error	16	577	36	

***significant differences for P<0.001,* significant differences for P<0.5.

A higher inhibiting effect on the length of root was derived from the T2 and T4 variants in both years of experimentation.

In the 2017 the length of roots from T1 and T3 variants were almost similar to untreated mustard, indicating that there was no detectable herbicide in the soil and the explanation coud be the higher quantity of precipitations registered in this year (Figure 7).

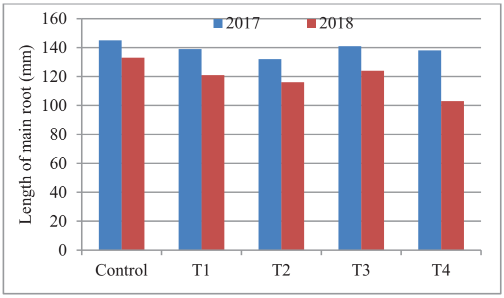


Figure 7. The effect of imazamox on lenght of mustard root

The negative effect on the height of plants was lower in both years of experimentation as compared with the effect on the length of root (Figure 8).

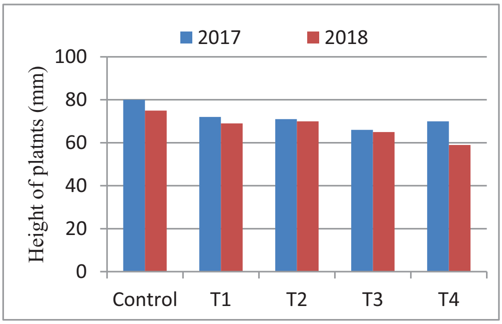


Figure 8. The effect of imazamox on height of mustard plants

Although in earlier stage of mustard vegetation the treatments with imazamox herbicide had a low residual effect on height of plants, but over time this effect was significantly reduced (Figure 9).

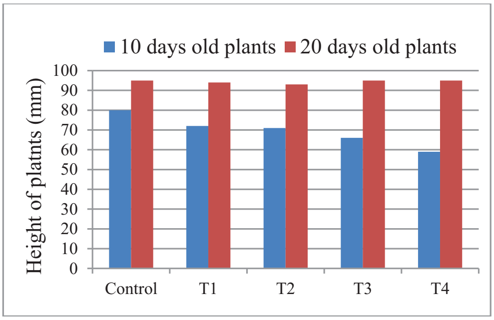


Figure 9. The effect of imazamox on height of mustard plants

The precipitation influence both transport and the herbicides degradation (microbial and chemical hydrolysis) (Streck, 2005; Ghinea et al., 2007).

Data from the literature show that in general, breakdown occurs by soil microbes and breakdown occurs more rapidly and herbicide activity increases as soil pH increases. The soil reaction in our experiences was slightly acidic from 6.06 to 6.36, with little difference between the treatments but a reduction of pH is observed in 2018 in all herbicidal variants (Table 3).

In terms of total nitrogen content, it was noted that soil from experimental plots had 0.141-0.150% total nitrogen (Nt) and organic matter was a few percent higher in 2017 (year without strong hydric stress) (Table 3).

The literature shows that variations in soil pH can influence how long a herbicide will persist. Loux and Reese (1992) (quate by Petcu Victor & Oprea Georgeta, 2013) found that imidazolinone herbicides adsorbed increases as organic matter (OM) increases and as soil pH decreases. All factors increasing microbial activity also increase herbicide degradation (warm, moist soils). Degradation increases in soils with pH above 6.5 (Imi) or 7 (TPS) because herbicide molecules are not adsorbedand are in soil solution for plant uptake and microbial breakdown, (www.cof.orst.edu).

Table 3. The nitrogen and soil reaction. Fundulea, 2017 and 2018

Year	Treatment	N t (%)	Organic matter (%)	Ph
2017	**Control**	**0.145**	**2.98**	**6.36**
	T1	0.143	2.65	6.33
	T2	0.149	2.60	6.35
	T3	0.150	2.75	6.43
	T2	0.149	2.65	6.51
2018	**Control**	**0.141**	**2.65**	**6.25**
	T1	0.140	2.43	6.21
	T2	0.144	2.44	6.12
	T3	0.145	2.30	6.16
	T2	0.141	2.31	6.25

ALS inhibitors are highly plant active through both foliage and root uptake. This ability to be active in the soil and be taken up through the root system is beneficial for the control of weeds that emerge after the date of application. In years of reduced herbicide degradation in the soil due to reduced temperatures or soil moisture, some ALS inhibitors or their metabolites can persist into the following growing seasons (Hall et al. 1999; Hill B. Dal, 1998). This prolonged persistence can potentially injure sensitive crops grown in rotation such as canola and lentils (non-Clearfield® varieties), mustard, or sugar beet (Moyer & Hamman, 2001).

Persistence of phytotoxic levels of a herbicide for more than 1 year can be a problem with some herbicides. Herbicide residues are most likely to occur following years with low rainfall because chemical and microbial activity needed to degrade herbicides are limited in dry soil. Crop damage from herbicide residues can be minimized by applying the lowest herbicide rate required for good weed control, by using band rather than broadcast applications, and by mouldboard plowing before planting the next crop. Moldboard plowing reduces phytotoxicity of some herbicides by diluting the herbicide residue in a large volume of soil, (www.cof.orst.edu).

In order to established if studied herbicides affect the previous crops it was analyzed the reaction of winter wheat plant to herbicides studied.

The results showed no significant differences in terms of main root length (Table 4).

Table 4. Analyses of variance for length of winter wheat root

Source of variance	DF	Mean square	S2	F factor and significance
Factor A (year)	1	28.03	28.03	1.83
Error	2	29.8	14.93	
Factor B (treatment)	4	277	69	4.55
A x B	4	66.13	16.53	1.08
Error	16	243	15.21	

In case of winter wheat, during the dry year 2018 as compared with 2017, the persistence of herbicides studied was higher, the decrease of length of wheat roots being 0.54-8.74% from control compared with that of 2017 when, the winter wheat root lengths varied from 0 to 5.40% from the control (Table 5).

Table 5. The length of winter wheat root

Year	2017	2018	2017	2018
	Length of root (mm)		Rate of reduction (%)	
Control	185	183	100	100
T1	185	182	0	0.54
T2	178	175	3.78	4.37
T3	180	176	2.70	3.82
T4	175	167	5.40	8.74

The explanation is that the humidity and heat accelerates biodegradation, but also facilitates the leaching, while a dry climate, on the contrary, increases persistence as non-biological degradation and biodegradation are less intense.

CONCLUSIONS

The root length bioassay is suitable for assessment of susceptibility/resistance of wild mustard populations to ALS-inhibiting herbicides.

Our study evidentiate that biodegradation of herbicides depends by climatic conditions from year of experimentations and soil properties. On wheat (as in mustard) the reduction in root length as a treatment effect was higher in the dry year. But from a statistical point of view, our results revealed no significant differences between the type of herbicide used and untreated check. This suggests that the herbicides studied do not significantly affect the growth of this plant.

Results from the bioassay should not be interpreted alone. Interpretation needs to include other tools and information such as label recommendations, rainfall restrictions, pH, organic matter, soil texture, and perhaps most importantly, grower and agronomist experience.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Eliason, R., Schoenau, J.J., Szmigielski, A.M., Laverty, W.M. (2004). Phytotoxicity and persistence of flucarbazone-sodium in soil. *Weed Sci*., *52*. 857–862.

Ghinea, L., Stefanic, G., Popescu, A., Oprea, G. (2007). Research in the soil chemistry and biology. *Analele INCDA Fundulea*, *LXXV*, 405–428.

Hall, L., Beckie, H., Wolf, T.M. (1999). How herbicides work: biology to application. *Alberta Agriculture, Food and Rural Development*. Edmonton, 133,

Hill, B.D., Moyer, J.R., Inaba, D.J., Doram, R. (1998). Effect of moisture on quincloracdissipation in Lethbridge soil. Can. *J. Plant Sci., 78*, 697–702.

Moyer, J.R., and Hamman, W.M. (2001). Factors affecting the toxicity of MON 37500 residues to following crops. *Weed Technol., 15*, 42–47.

Strek, H.J. (2005). The science of Dupont's soil residual herbicides in Canada. In: R.C. Van Acker (Ed.), *Soil Residual Herbicides: Science and Management. Topics in Canadian Weed Science*, *3*, 31–44. Sainte-Anne-de Bellevue, Quebec. Canadian Weed Science Society- Société canadienne de malherbologie.

Petcu, V., and Oprea, G. (2013). Determination of the persistance in soil of herbicides inhibiting the enzyme acetolactate synthase (ALS) by bioassay. *Soil Science*, *XLVII* (2), 33–40. ISSN 0585-3052.

Walker, A. (1987). Herbicide persistence in soil. *Rev. Weed Sci., 3*, 1–18.

Watson, P.R. (2005). Soil residual herbicide bioassays: Science and practice. In: *Soil Residual Herbicides: Science and Management*. Ed. Rene C. Van Acker. Quebec. Canada.

Whitcomb, C.E. (1999). An introduction to ALS-inhibiting herbicides. *Toxicol. Ind. Health*., *15*, 232–240.

http://www.cof.orst.edu/cof/fs/kpuettmann

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF SOIL TILLAGE AND HYBRID MATURITY GROUP UPON THE MAIZE GRAIN YIELD FROM MOARA DOMNEASCĂ, ILFOV COUNTY

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Abstract

Sustainable agriculture is an increasingly used concept, as it also includes the minimum or zero tillage technologies. This implies the complete absence of ploughing, disking and other crop maintenance works. This paper presents the mean of the research carried out between 2014-2016 at the Moara Domnească Teaching Farm located in the Ilfov County and belonging to the University of Agronomic Sciences and Veterinary Medicine of Bucharest. The biological material consisted in several early maize hybrids belonging to the following groups: FAO- 270, 280 (Sunergy, PR39B76), FAO- 340, 380 (Bonito, PR37N01) and FAO- 400, 500, 510 (Olt, Sumbra, P0216). The experimental variants for the soil tillage system were: ploughing 20 cm (control), chiselling 20 cm, chiselling 40 cm, disking 10 cm, disking/ploughing 20 cm, disking/chiselling 40 cm. Production ranged between 4886.0 kg/ha (in the FAO 270, 280) and 6626.2 kg/ha (FAO 340, 380), and recorded the highest values of 7034.8 kg/ha in hybrid P0216.

Key words*: soil tillage, Zea mays* L.*, hybrids, yield.*

INTRODUCTION

Plant cultivation technologies include an array of interdependent elements, and make up an organized whole which ensures that farming practices work and meet the intended purpose.
The interactions between the various technological elements, the diversity of environmental conditions and the large number of cultivated plants motivate the wide range of technologies and the differences between them. These technologies are differentiated according to the climatic specificity and the relief of the agricultural areas; within them, the differences depend on the soil particularities, the cultivated plant, the agricultural machinery system and the impact of the technological elements on the environment (Guş, 2003).
Minimum soil tillage involves the replacement of ploughing in crop technology and of soil tillage partly, which leads to soil better preservation. That system was first implemented in the US, then in England and it gradually expanded throughout Europe.
The results obtained in Virginia (1972) showed that the no-tillage system ensured the same effect on the yield treated with lower doses of phosphorus and potassium fertilizers and the loss of water was lower owing to the protective layer (Budoi, 1996). Reduced soil tillage in maize increased after the use of broad spectrum herbicides (Şarpe et al.,1996; Ulinici et al., 1967).
Research on minimum soil tillage has been carried out in Romania, too. Promising results have been obtained so far in the technology of maize, sunflower, soybean and other plants, by carrying out autumn tillage. In spring a single pass of the aggregate on the field prepares the germinating bed concurrently with the application of fertilizers, herbicides and sowing (C. Pintilie et al., 1985).
Unconventional soil tillage, together with other basic elements of sustainable agriculture, have direct influence on crop plants (Guş, 2001; Marin, 2008; Rusu, 2009; Pop, 2011).
Maize is the most commonly studied plant in terms of crop requirements in the minimum tillage or no-tillage system (Hallauer and Colvin, 1985; Wright,1990; Uri, 2000). The selection of the hybrid type for this agricultural system has been a constant concern for the breeders (Hallauer and Colvin, 1985; Newhouse and Crosbie, 1986; Hesterman et al.,1988; De Felice et al., 2006).

The research carried out by A. Canarache and Elisabeta Dumitru at ICPA in 1991 was based on the centralization of the pedoclimatic data collected from OSPA throughout Romania, and showed that about 42% of the arable land met moderate and favorable conditions for the unconventional soil cultivation system (15% under favorable conditions and 27% under moderately favorable conditions) (Guş, 2003).

Research carried out by Marin D.I. on the red preluvosol soil of Moara Domnească, Ilfov County, between 2008 and 2010, showed that the average yield of grain maize ranged from 7,450 kg/ha in the conventional system and 7,248 kg /ha in the nonconventional system (in the 40 cm chisel version).

Soil tillage based only on disc harrows resulted in lower yields compared to other soil tillage variants, i.e. 6,355 kg/ha. The results obtained by soil tillage based on chiselling at 40 cm in depth recommend this variant as a technological alternative of soil tillage for the application of the sustainable agriculture system.

MATERIALS AND METHODS

Research was carried out on the experimental field of the Moara Domnească Teaching Farm, Ilfov County, belonging to U.S.A.M.V. Bucharest.

The experiment included the following factors:

Factor A - soil tillage based on the following variants: a_1-ploughing at 20 cm in depth (control - A20 cm), a_2-chisseling 20 cm (C20 cm), a_3-chiselling 40 cm (C40 cm), a_4-disking 10 cm (Disc), a_5-disking/ploughing 20 cm (D/A20 cm), a_6-disking/chiselling 40 cm (D/C40 cm), disking in succesion disking/chiselling 40 cm and disking/ploughing 20 cm was performed in the pre-emergent plant, winter wheat.

Factor B - early maize hybrids: Sunergy, PR39B76 (FAO- 270, 280);

- semi early hybrids: Bonito, PR37N01 (FAO-340, 380);
- semi late hybrids: Olt, Sumbra, P0216 (FAO-400, 500, 510).

Soil tillage was carried out in the last decade of September.

Maize hybridw were sown in the second decade of April by SPC 6 seed drill.

Weeds are a risk factor for agricultural crops as they prevent normal growth of the latter. Thus, for control purposes, the treatments applied were based on such herbicides as Dual Gold in a dose of 1.4 l/ha (*s-metolaclor* 960 g/l) - pre-emergent and Dicopur Top 1.0 l/ha (344 g/l acid 2.4 D + 120 g/l *dicamba*) + Titus 25DF, 50g/ha (25% *rimsulfuron metil*) - post-emergent). Two mechanical hoeing works were also performed during the growing season.

Fertilization provided annual doses of $N_{120}P_{60}K_{60}$ kg d.m./ha of the complex fertilizer NPK 15:15:15 and urea.

Harvesting was carried out in the second decade of September, depending on the climatic conditions which are an important factor influencing plant production. The data recorded by the Găneasa Weather Station were used for the characterization of the climatic conditions. The climatic condition between 2014 and 2016 had direct influence on the growth of the crop plants. The evolution of the thermal regime and rainfalls during the research period oscillated in comparison with the multiannual mean values. From a meteorological viewpoint, 2014 and 2016 were characterized by variations in temperature and particularly rainfalls (Table 1). For the maize growing period (April-August), the average temperature medie was 19.9ºC, i.e. 1.7ºC higher than the normal value in the area, while the sum of rainfalls was 381.2 mm and 343.4 mm, respectively, which was close to the normal value in the area. It is important to note that rainfalls in July and August (2014) were much lower than the normal values in the area.

Less favourable to maize crop were the climatic conditions in 2015, when the rainfalls during the maize growing season (April-August) recorded 146.0 mm, lower than the normal value of 203.3 mm in the area, while the average temperature of 20.1ºC was higher by 1.9ºC than the normal value in the area.

RESULTS AND DISCUSSIONS

The influence of soil tillage on the grain yield in the maize crop

The results concerning the influence of basic soil tillage on the maize grain yield are presented in Table 2.

Table 1. Climatic conditions of Moara Domnească, Ilfov County (2014-2016)

Month	Average temperature (ºC)						Rainfalls (mm)					
	2013-2014	**2014-2015**	**2015-2016**	**Average 2013-2016**	**Normal**	**Diff. from normal value**	**2013-2014**	**2014-2015**	**2015-2016**	**Average 2013-2016**	**Normal**	**Diff. from normal value**
October	14.0	11.7	10.7	12.1	**11.0**	1.1	81.7	64.2	70.0	71.9	**35.8**	36.1
November	8.3	5.3	7.7	7.1	**5.3**	1.8	17.6	49.1	110.6	59.1	**40.6**	18.5
December	-0.2	0.8	3.1	1.2	**0.4**	0.8	1.2	84.6	1.8	29.2	**36.7**	-7.5
January	-0.5	-1.1	-3.4	-1.6	**-3.0**	1.4	33.2	33.4	62.6	43.0	**30.0**	13.0
February	1.2	2.0	1.7	1.6	**-0.9**	0.7	7.6	21.4	35.6	21.5	**32.1**	-10.6
March	8.9	6.3	7.5	7.5	**4.4**	3.1	37.3	65.6	67.8	56.9	**31.6**	25.3
April	13.4	11.7	14.3	13.1	**11.2**	1.9	116.0	2.0	64.6	60.8	**48.1**	12.7
May	19.3	18.6	15.8	17.9	**16.5**	1.4	88.0	33.6	71.0	64.2	**67.7**	-3.5
June	19.9	20.9	22.4	21.0	**20.2**	0.8	113.0	56.8	114.8	94.8	**86.3**	8.5
July	22.8	25.2	24.1	24.0	**22.1**	1.9	38.0	5.2	4.2	15.8	**63.1**	-47.3
August	24.1	24.4	23.0	23.8	**21.1**	2.7	26.2	48.4	88.8	54.4	**50.5**	3.9
September	18.4	18.8	18.9	18.7	**17.5**	1,2	60.6	86.0	83.2	76.6	**33.6**	43.0
Average/Sum April-August	19.9	20.1	19.9	19.7	**18.2**	1.5	381.2	146.0	343.4	290.2	**349.3**	-59.1
Average/Sum October-September	12.4	12.0	12.1	12.1	**10.5**	1.6	620.4	550.3	775.0	648.5	**556.1**	92.4

Table 2. Influence of soil tillage on maize grain yield, Moara Domnească, Ilfov County, average 2014-2016

Soil tillage variant/ Hybrids	Yield	A20	C20	C40	Disking	D/A20	D/C40
PR39B76	kg/ha	6313.4	5703.7	5772.7	5224.4	6406.9	5969.2
	%	100	90	92	83	101	94
	Diff. kg/ha	Mt	-609.7 ^{ooo}	-540.7 ^{ooo}	-1089.0 ^{ooo}	93.5	-344.2 ^{o}
	$LSD_{5\%}$= 252.3 kg/ha $LSD_{1\%}$= 358.7 kg/ha $LSD_{0.1\%}$= 519.4 kg./ha						
SUNERGY	kg/ha	5477.2	4903.2	5068.1	4547.6	5505.7	5168.4
	%	100	89	92	83	101	94
	Diff. kg/ha	Mt	-574.0 ^{ooo}	-409.1 ^{oo}	-929.6 ^{ooo}	285	-308.8 ^{o}
	$LSD_{5\%}$= 239.6 kg/ha $LSD_{1\%}$= 331.8 kg/ha $LSD_{0.1\%}$= 457.9 kg./ha						
PR37N01	kg/ha	6754.7	5968.4	6192.7	5407.7	6865.8	6283.1
	%	100	88	91	80	101	93
	Diff. kg/ha	Mt	-786.3 ^{oo}	-562.0 ^{o}	-1347.0 ^{ooo}	111.1	-471.6 ^{oo}
	$LSD_{5\%}$= 405.7 kg/ha $LSD_{1\%}$= 576.8 kg/ha $LSD_{0.1\%}$ = 835.2 kg./ha						
BONITO	kg/ha	6147.1	5344.6	5676.6	5019.2	6386.7	5578.2
	%	100	87	92	82	104	91
	Diff. kg/ha	Mt	-802.5 ^{ooo}	-470.5 ^{oo}	-1127.9 ^{ooo}	239.6	-568.9 ^{oo}
	$LSD_{5\%}$= 274.8 kg/ha $LSD_{1\%}$= 390.7 kg/ha $LSD_{0.1\%}$= 565.7 kg./ha						
OLT	kg/ha	6386.5	5715.2	6125.1	5079.3	6378.1	6213.0
	%	100	89	96	80	100	97
	Diff. kg/ha	Mt	-671.3 ^{ooo}	-261.4	-1307.2 ^{ooo}	-8.4	-173.5
	$LSD_{5\%}$= 264.8 kg/ha $LSD_{1\%}$ = 376.5 kg/ha $LSD_{0.1\%}$= 545.1 kg./ha						
SUMBRA	kg/ha	6157.7	5617.7	5850.3	4975.1	6341.3	5971.9
	%	100	91	95	81	103	97
	Diff. kg/ha	Mt	-540.0 ^{ooo}	-307.4 ^{o}	-1182.6 ^{ooo}	183.6	-185.8
	$LSD_{5\%}$= 225.8 kg/ha $LSD_{1\%}$ = 321.0 kg/ha $LSD_{0.1\%}$ = 464.9 kg./ha						
P0216	kg/ha	6930.1	5902.3	6328.1	5508.3	7034.8	6442.1
	%	100	85	91	80	101	93
	Diff. kg/ha	Mt	-1027.8 ^{ooo}	-602.0 ^{oo}	-1421.8 ^{ooo}	104.7	-488.0 ^{oo}
	$LSD_{5\%}$= 335.2kg/ha $LSD_{1\%}$= 476.6 kg/ha $LSD_{0.1\%}$ = 690.1kg./ha						

The application of minimum tillage resulted in various yields, depending on hybrid.

Between 2014 and 2016 the average yield of maize grain in the seven hybrids under study ranged between 4,975.1 kg/ha (hybrid Sumbra, disking 10 cm) and 7,034.8 kg/ha (hybrid P0216, disking/ploughing 20 cm). In the control (ploughing 20 cm) the highest average yield was recorded in the hybrid P0216, i.e. 6,930.1 kg/ha, followed by PR37N01 (6754.7 kg/ha) and Olt (6386.5 kg/ha). The average yields recorded in the control variant ranged between 5,477.2 kg/ha (hybrid Sunergy) and 6,930.1 kg/ha (hybrid P0216).

The hybrid P0216 recorded the highest average yield both in the traditional worked variant and in unconventional system (6,442.1 kg/ha - disking/chiselling 40). The closest to the control were the yields obtained from minimum tillage and were recorded in the variants chiselled at 40 cm in depth, i.e. 91% (hybrids PR37N01, Bonito and P0216) and 97% (hybrids Olt and Sumbra). The chiselled variants at 20 cm in depth resulted in yields of 85% (hybrid P0216 - 5,903.2 kg/ha) and 91%, respectively (hybrid Sumbra - 5,617.7 kg/ha) compared with the control ploughed at 20 cm in depth. The yields obtained from the seven hybrids in the disked variant recorded highly significant negative values, compared with the control ploughed at 20 cm in depth. They ranged between 4,975.1 kg/ha (hybrid Sumbra) and 5508.3 kg/ha (hybrid P0216), i.e. between 80 and 83% of the values recorded by the control.

Influence of the hybrid maturity group on the grain yield in the various variants of soil tillage.

The hybrids grown in Romania are classified into five groups of early ripeness according to the growing period, as follows: very early hybirds (FAO classification 100-199); early hybrids (FAO 200-299); semi early hybrids (FAO 300-399); semi late hybrids (FAO 400-499 and 500-599) and late hybrids (>600). Hybrid zoning is based on the thermal constant, which represents the sum of temperatures over 10°C throughout the entire growing season.

The comparative analysis of the maize hybrid maturity groups (Table 3) shows that, compared with the early hybrids (the control group), higher yields were recorded in both semi early and semi late hybrids. Compared with the control group, semi late hybrids recorded yields ranging between 5,187.5 kg/ha (disked variant) and 6,584.7 kg/ha (variant disking/ploughing 20), i.e. 106 and 110%, respectively, of the control value. In the conventional system, the yields ranged between 6,450.9 kg/ha (semi early hybrid group - variant ploughing 20 cm) and 6,626.2 kg/ha (semi early hybrid group - variant disking/ploughing 20 cm). In the nonconventional system, the semi late hybrid group was conspicuous, compared with the control group, by its increased yield, i.e. 680.7 kg/ha. This showed that the semi late hybrids were better suitable for southeastern Romania.

Table 3. Influence of hybrid maturity group on maize grain yield in various soil tillage systems, Moara Domnească, Ilfov County, average 2014-2016

FAO group/ Soil tillage	FAO 270-280 (Early)			FAO 340-380 (Semi early)			FAO 400- 510 (Semi late)		
	kg/ha	%	Diff. kg/ha	kg/ha	%	Diff. kg/ha	kg/ha	%	Diff. kg/ha
A20	5895.3	100	Mt	6450.9	109	555.6**	6491.4	110	596.1***
C20	5303.4	100	Mt	5656.5	106	353.1*	5745.0	108	441.6**
C40	5420.4	100	Mt	5934.6	109	514.2 *	6101.1	112	680.7***
Disc	4886.0	100	Mt	5213.4	106	327.4	5187.5	106	301.5*
D/A20	5956.3	100	Mt	6626.2	111	669.9**	6584.7	110	628.4***
D/C40	5568.8	100	Mt	5930.6	106	361.8*	6209.0	111	640.2***
				$LSD_{5\%}$= 340.2 kg/ha $LSD_{1\%}$ = 483.7kg/ha $LSD_{0.1\%}$= 700.4 kg./ha			$LSD_{5\%}$= 275.2 kg/ha $LSD_{1\%}$= 391.9 kg/ha $LSD_{0.1\%}$ = 566.7 kg./ha		

The average results regarding the influence of soil tillage system on the Thousand Grain Weight (TGW) and Standard Mass per Storage Volume (SMPSV) in the seven maize hybrids grown in both traditional and minimum tillage systems are shown in Tables 4 and 5.

The values analysed were influenced by the climatic conditions, grown hybrids and soil tillage system. The TGW of the seven hybrids under study ranged between 253.1 g (hybrid Sunergy - disking) and 279.0 g (hybrid P0216 - disking/ploughing 20 cm).

In the traditional system, hybrid P0216 ranged between 277.7 g in variant A20 cm (control) and 279.0 g in variant disking/ploughing 20 cm, followed by hybrids PR37N01 (278.5 g, variant disking/ploughing 20 cm) and hybrid Olt, respectively, that recorded 273.8 g in variant ploughing 20 cm.

The lowest valued of TGW were recorded in the variants disked at 10 cm in depth and chiselled at 20 cm in depth of the hybrids Sunergy and Bonito. The closest to the control ploughed at 20 cm in depth were the values recorded in the following hybrids: PR39B76 (269.2 g - variant disking/chiselling 40 cm), Olt (272.4 g - variant disking/chiselling 40 cm) and Sumbra (269.1 g - variant disking/chiselling 40 cm), i.e. 99% of their values.

Table 4. Influence of basic soil tillage on thousand grain weight (TGW) in maize crop, Moara Domnească, Ilfov County, average 2014-2016

Soil tillage variant/ Hybrids	TGW (g)	A20	C20	C40	Disc	D/A20	D/C40
PR39B76	TGW (g)	271.9	266.9	267.8	262.8	273.4	269.8
	%	100	98	98	96	101	99
	Diff. (g)	Mt	-5.0	-4.1	-9.1 ^{o}	1.5	-2.1
	$LSD_{5\%}$= 9.1 g $LSD_{1\%}$= 14.1 g $LSD_{0.1\%}$= 20.4 g						
SUNERGY	TGW (g)	263.3	257.0	261.1	253.1	263.4	260.8
	%	100	97	99	96	100	99
	Diff. (g)	Mt	-6.3	-2.2	-10.2	0.1	-2.5
	$LSD_{5\%}$= 10.4 g $LSD_{1\%}$= 14.8 g $LSD_{0.1\%}$= 21.4 g						
PR37N01	TGW (g)	277.0	270.1	272.0	262.9	278.5	272.8
	%	100	97	98	95	101	98
	Diff. (g)	Mt	-6.9	-5.0	-14.1 ^{o}	1.5	-4.2
	$LSD_{5\%}$= 10.1 g $LSD_{1\%}$= 14.4 g $LSD_{0.1\%}$ = 20.9 g						
BONITO	TGW (g)	270.9	261.2	264.6	255.0	271.6	265.8
	%	100	96	97	94	100	98
	Diff. (g)	Mt	-9.7	-6.3	-15.9 ^{o}	0.7	-5.1
	$LSD_{5\%}$= 11.3 g $LSD_{1\%}$ = 16.1 g $LSD_{0.1\%}$= 23.4 g						
OLT	TGW (g)	273.8	267.3	270.3	260.4	273.6	272.4
	%	100	97	98	95	100	99
	Diff. (g)	Mt	-6.5	-3.5	-13.4 ^{o}	-0.2	-1.4
	$LSD_{5\%}$= 9.7 g $LSD_{1\%}$ = 13.8 g $LSD_{0.1\%}$= 20.0 g						
SUMBRA	TGW (g)	270.1	264.6	268.2	260.1	272.5	269.1
	%	100	98	99	96	101	99
	Diff. (g)	Mt	-5.5	-1.9	-10.0 ^{o}	2.4	-1.0
	$LSD_{5\%}$= 8.9 g $LSD_{1\%}$ = 12.6 g $LSD_{0.1\%}$ = 18.3 g						
P0216	TGW (g)	277.7	270.8	272.3	264.3	279.0	273.2
	%	100	97	98	95	100	98
	Diff. (g)	Mt	-6.9	-5.4	-13.7 ^{o}	1.3	-4.5
	$LSD_{5\%}$= 10.0 g $LSD_{1\%}$= 14.3 g $LSD_{0.1\%}$ = 20.7 g						

On average for 2014-2016, the Standard Mass per Storage Volume (SMPSV) recorded high values in the traditional variants where the highest values were recorded in the hybrids P0216 (73.5 kg/hl, variant disking/ploughing 20 cm) and PR37N01 (73.1 kg/hl, variant disking/ploughing 20 cm). This exceeded the control by 0.6 and 0.8 kg/hl, respectively.

The values resulted from nonconventional tillage recorded between 90 and 99% of the control.

The chiselled variant at 20 cm in depth recorded values that ranged from negative to highly significant negative, compared with the control (ploughing 20 cm).

The disking of reddish preluvosoil resulted in lower values of the SMPSV, compared with the other soil tillage variants. They ranged between 63.7 kg/hl (hybrid Bonito) and 67.3 kg/hl (hybrid PR39B76), which were significantly negative, compared with the control.

Table 5. Influence of basic soil tillage on Standard Mass per Storage Volume (SMPSV) in grain maize crop, Moara Domnească, Ilfov County, average 2014-2016

Soil tillage variant/ hybrids	SMPSV (kg/100 l)	A20	C20	C40	Disc	D/A20	D/C40
PR39B76	SMPSV (kg/hl)	71.2	68.4	69.6	67.3	71.7	70.5
	%	100	96	97	94	100	99
	Diff. (kg/hl)	Mt	-2.8 ^{oo}	-1.6	-3.9 ^{oo}	0.5	-0.7
	LSD$_{5\%}$= 1.9 kg/hl LSD$_{1\%}$= 2.8 kg/hl LSD$_{0.1\%}$= 4.0 kg/hl						
SUNERGY	SMPSV (kg/hl)	66.3	64.2	65.4	63.8	66.0	65.5
	%	100	97	98	96	100	99
	Diff. (kg/hl)	Mt	-2.1 ^{o}	-0.9	-2.5 ^{o}	-0.3	-0.8
	LSD$_{5\%}$= 2.1 kg/hl LSD$_{1\%}$= 2.9 kg/hl LSD$_{0.1\%}$= 4.3 kg/hl						
PR37N01	SMPSV (kg/hl)	72.5	68.9	70.9	66.4	73.1	70.7
	%	100	95	98	91	101	97
	Diff. (kg/hl)	Mt	-3.6 ^{oo}	-1.6	-6.1 ^{ooo}	0.6	-1.8
	LSD$_{5\%}$= 2.4 kg/hl LSD$_{1\%}$= 3.5 kg/hl LSD$_{0.1\%}$ = 5.0 kg/hl						
BONITO	SMPSV (kg/hl)	71.0	66.1	68.2	63.7	70.9	69.1
	%	100	93	96	90	100	97
	Diff. (kg/hl)	Mt	-5.0 ^{ooo}	-2.8 ^{o}	-7.3 ^{ooo}	-0.1	-1.9
	LSD$_{5\%}$= 2.3 kg/hl LSD$_{1\%}$= 3.2 kg/hl LSD$_{0.1\%}$= 4.7 kg/hl						
OLT	SMPSV (kg/hl)	71.1	68.2	69.5	66.0	71.4	70.6
	%	100	96	97	93	100	99
	Diff. (kg/hl)	Mt	-2.9 ^{oo}	-1.6	-5.1 ^{ooo}	0.3	-0.5
	LSD$_{5\%}$= 2.0 kg/hl LSD$_{1\%}$= 2.9 kg/hl LSD$_{0.1\%}$= 4.2 kg/hl						
SUMBRA	SMPSV (kg/hl)	71.1	68.4	69.5	65.8	71.5	70.6
	%	100	96	98	92	101	99
	Diff. (kg/hl)	Mt	-2.7 ^{o}	-1.6	-5.3 ^{ooo}	0.4	-0.5
	LSD$_{5\%}$= 2.0 kg/hl LSD$_{1\%}$= 2.9 kg/hl LSD$_{0.1\%}$= 4.2 kg/hl						
P0216	SMPSV (kg/hl)	72.7	68.9	70.9	66.2	73.5	71.0
	%	100	94	97	91	101	97
	Diff. (kg/hl)	Mt	-3.8 ^{oo}	-1.8	-6.5 ^{ooo}	0.8	-1.7
	LSD$_{5\%}$= 2.4 kg/hl LSD$_{1\%}$= 3.4 kg/hl LSD$_{0.1\%}$ = 5.0 kg/hl						

CONCLUSIONS

The climatic conditions between 2014 and 2016 manifested direct influence on the development of the crop plants. The evolution of the thermal regime and rainfalls throughout the same period oscillated, compared with the multiannual average values.

The average maize grain yield of the seven hybrids studied between 2014 and 2016 ranged between 4,975.1 kg/ha (hybrid Sumbra, disking 10 cm) and 7,034.8 kg/ha (hybrid P0216, disking/ploughing 20 cm).

The yields that were the closest to the control were obtained from minimum tillage in the following variants: chiselling 40 cm and disking/chiselling 40 cm, i.e. between 91% (in hybrids PR37N01, Bonito and P0216) and 97% (in hybrids Olt and Sumbra).

Concerning the influence of hybrid maturity group on the maize grain yield, the highest yield was recorded in the semi late hybrids, i.e. 680.7 kg/ha (chiselling 40 cm), compared with the control.

The thousand grain weight of the seven hybrids studied recorded values between 253.1 g (hybrid Sunergy - disking) and 279.0 g (hybrid P0216 - disking/ploughing 20 cm).

The standard mass per storage volume (SMPSV) on average for the period between 2014 and 2016 recorded high values in the conventional variants, compared with the nonconventional ones. In the sonventional system, the values of the standard mass per storage volume ranged between 66.0 kg/hl (hybrid Sunergy - disking/ploughing 20 cm) and 73.5 kg/hl (hybrid P0216 - disking/ploughing 20 cm).

Considering the results obtained during the three years of research, we recommend cisseling at 40 cm in depth as an alternative to traditional soil tillage in the Moara Domnească area, the Ilfov County.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

De Felice, M., Carter, P., Mitchell, S. (2006). Influence of Tillage on corn yield in the USA and Canada. *Crop Insights., 16*(11). Pioneer Hi-Breed. Johnston. I.A.

Guş, P., Rusu, T., Bogdan, Ileana, Haţegan, M. (2001). *Sisteme neconvenţionale de lucrare a solului*. Ed. Risoprint. Cluj-Napoca.

Guş, P., Rusu, T., Stănilă, S. (2003). *Lucrările neconvenționale ale solului și sistema de mașini*. Editura Risoprint. Cluj-Napoca.

Hallauer, A.R., and Colvin, T.S. (1985). Corn hybrids response to four methods of tillage. *Agronomy Journal, 77*, 547–550.

Hesterman, O.B., Pierce, F.J., Rossman, E.C. (1988). Performance of commercial hybrids under convențional and no-tillage system. *J. Prod. Agric*., *1*, 202–206.

Marin, D.I., Rusu, T., Mihalache, M. (2008). The effect of non-conservation tillage up on the edaphic component of the Agricultural Ecosystem. *Analele Universităţii din Craiova. Lucrări ştiinţifice - Agricultură. Montanologie. Cadastru*, 399–402. ISSN 1841-8317.

Marin, D.I., Mihalache, M., Ciontu, C., Ilie, L., Bolohan, C. (2011). Influența sistemului de lucrare a solului asupra culturilor de mazăre. grâu și porumb. USAMV Cluj. *5th Internațional Symposium - Soil Minimum Tillage System*, 111–118. Ed. Risoprint Cluj-Napoca.

Newhouse, K.E., and Crosbie, T.M. (1986). Interacțion of maize hybrids with tillage system. *Agronomy Journal*, *78*, 951–954.

Pintilie, C., Timariu, Gh., Romosan, St. (1985). *Agrotehnică.* Editura Didactică și Pedagogică. București.

Pop, A.I., Guş, P., Rusu, T., Bogdan, I., Moraru, P., Cîmpean, P. (2011). Influence of soil tillage system on weeding degree and corn yield. *Scientific Papers. Series A. Agronomy, LIV*, 230–234. ISSN 1222-5339.

Rusu, T., Guș, P., Bogdan, I., Moraru, P.I., Pop, A.I., Clapa, D., Marin, D.I., Oroian, I. and Pop, L.I. (2009). Implications of Minimum Tillage Systems on Sustainability of Agricultural Production and Soil Conservation. *Journal of Food. Agriculture & Environment, 7*(2), 335–338.

Uri, N.D. (2000). Perception on the use of no-till farming in producțion agriculture in United States: an analysis of survey results. *Agricultural Ecosystem and Enviroment, 77*, 263–266.

Wright, D.L. (1990). Starter fertilizer placement in No-Till corn. Florida *Agr. Exp. Station Journal*, *9943*, 27–29.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOIL REQUIREMENTS AND ENVIRONMENTAL CONSIDERATIONS FOR PENNYROYAL (*Mentha pulegium* L.): A CASE STUDY FROM ROMANIA

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Abstract

Although there are many studies on mint species, too few establish the link between its growth and the environment in which it grows. Between 7 and 22 August 2017, at Ilganii de Sus village (Nufăru commune, Tulcea County), on Sulina Danube branch, five soil samples taken from the base of some populations of pennyroyal plants (Mentha pulegium L.) were analysed. The sampling points were chosen according to the different habitats encountered at Ilganii de Sus (ruderal sites, grazed pastures, flooded ditches, sandy beaches on the banks of Arhipenco channel), with different soil texture.
Surrounded by other mint species and cocklebur, pennyroyal can be seen as a potential environmental weed in the study area. We found that the highest values of humus content, potassium and phosphorus levels correspond to a zone located at the western edge of the village, where household waste and poultry manure are deposited. The pH values of soil at Ilganii de Sus varied between 7.30 and 8.57; meanwhile, salts levels indicated a non-saline soil type. As far as we know, this report brings the first data concerning the soil features and environmental conditions for Mentha pulegium in a Romanian landscape.

Key words*: habitat, Ilganii de Sus, Mentha pulegium, Pennyroyal, soil.*

INTRODUCTION

Pennyroyal is the most common vernacular name used for *Mentha pulegium* L., a member of Lamiaceae family native to Europe, North Africa and Middle East (Miraj & Kiani, 2016).
Worldwide, *Mentha pulegium* was studied intensively so far from several points of view: chemical characterization, antibacterial and antioxidant activities of essential oil (Silva et al., 2015); pharmacological effect (Miraj & Kiani, 2016); insecticidal activity (Zekri et al., 2013; Rocha et al., 2015); melliferous characteristics (Ion & Ion, 2007); therapeutic effects (Brahmi et al., 2017); abortifacient agent (Gerenutti et al., 2014); determination of mineral content (Paşca et al., 2017); botany, ecology and food uses (Batsatsashvili et al., 2016); histo-anatomical and physiological features (Andro, 2012); classification according to geographical location (Kanakis et al., 2011); genetic diversity (Fadhel & Boussaïd, 2004). However, as Mansoori (2014) noticed, only few reports are available on environment condition e.g. soil properties related to mint growth.
Holzer (2012) labelled the pennyroyal as unpretentious species, which prefers "good soil", grows in the sun to half shadow, and is sensitive to frost.
Taking into consideration the general reputation of mint species in traditional medicine, *Mentha pulegium* is beneficial in terms of its use as medicinal plant for refreshing teas or essential oil in cosmetics. However, some restrictions are imposed when it comes to pennyroyal. Thus, studies have revealed that a volatile component named pulegone present in *M. pulegium* is hepatotoxic and affects uterine function (Bakerink et al., 1996; Hadi et al., 2017; Stringaro et al., 2018). Unfortunately, for pennyroyal oil poisoning there is no antidote (https://www.poison.org/; https://livertox.nih.gov/).
In Romania, the local names of *M. pulegium* are known as "busuicoul cerbilor", "menta franţuzească", "izma proastă" or "menta puricilor".
This mint species was reported before in the vegetation of several Romanian Counties, as: Bihor (Gavra, 2015), Botoşani (Tănase & Ştefan, 2010), Caraş-Severin (Prodan et al.,

2010), Călăraşi (Ion & Ion, 2007), Dolj (Răduţoiu et al., 2014), Galaţi (Oprea, 2004), Ilfov (Anastasiu & Liţescu, 2012), Maramureş (Jiboc, 2014), Mureş (Domokos & Cristea, 2013), Suceava (Tomescu & Chifu, 2009), Vaslui (Irimia & Danu, 2010), Timiş (Neacşu et al., 2015).

In Tulcea county, *M. pulegium* was already mentioned in Danube Delta Biosphere Reserve (D.D.B.R.) including Ilganii de Sus (Covaliov et al., 2012). In a previous report, we recorded pennyroyal at Ilganii de Jos (Dobrin et al., 2013).

Figure 1. Different habitats for populations of *Mentha pulegium* at Ilganii de Sus (August 2017)

Grigore (2008) included *M. pulegium* on the list of salted plants of Romanian flora. Ion and Ion (2007) stated that *M. pulegium* is a mezohidrophyte species with pretty dense populations in the water meadows and grasslands around the Danube wall.

At Ilganii de Sus (Tulcea County), pennyroyal cohabits with other spontaneous mint species, such as water mint *Mentha aquatica* L. and horsemint *Mentha longifolia* (L.) Huds. However, compared to the latter species, *M. pulegium* is rather abundant, especially in the grassy surroundings of Ilganii de Sus (Figure 1). Most often, pennyroyal shares its vast territory with cocklebur (*Xanthium* sp.), being challenged to withstand an environment dominated by this invasive plant.

The soil features in Ilganii de Sus is less known. In a monograph concerning this village, it is mentioned that there are two types of predominant soils: alluvial soils in the dry, habitable and arable area, respectively gleiosoils in the wet area. Alluvial soils have varied sandy-clay texture, are carbonate and poor in organic matter (Moţoc and Manole, 2015).

Since pennyroyal is so frequent in the studied area and generally in the Danube Delta, the interest of our research has focused on type of soil where *M. pulegium* prefers to grow, so that in future it will be a reference for those searching for the ecological aspects, caring and using this mint species in a broader sense.

MATERIALS AND METHODS

Five soil samples were obtained from the following sampling points (SP) at Ilganii de Sus, Tulcea County (Figure 2):

SP 1 (45°11′50.90″N/28°56′50.70″E);
SP 2 (45°11′58.65″N/28°57′2.35″E);
SP 3 (45°11′39.83″N/28°58′10.33″E);
SP 4 (45°11′31.60″N/28°56′1.59″E);
SP 5 (45°11′43.98″N/28°57′44.11″E).

Soil samples (S1-S5) were taken from the base of pennyroyal populations in each point (SP1-SP5), from a depth of 0-20 cm.

The choosing of these points was based on different habitats encountered at Ilganii de Sus (grazed pastures, flooded areas, sandy beaches on the banks of Arhipenco channel, ruderal places) where soil texture varies.

The methods used for investigation of soil properties are presented in Table 1.

Figure 2. Survey area for soil samples (map source: Google Earth)

Table 1. Soil analyses, methods and instrumentation

Analyses	Method	Apparatus
pH_{H2O} (1:2.5)	potentiometry	Hanna pH-meter
Total soluble salts	conductometry	Hach sens Ion 7
Potassium (mobile form), K_{AL}	flame emission spectrometry	Sherwood 410
Phosphorus (mobile form), P_{AL}	spectrophotome try	CECIL 2041 spectrometer
Humus content	Walkley-Black-Gogoaşă	-

RESULTS AND DISCUSSIONS

The features of each sample soil are indicated in Table 2.

Table 2. Soil agrochemical parameters (Ilganii de Sus, August 2017)

Soil sample	pH	Soluble salts $dS \cdot m^{-1}$	P_{AL} mg/kg	K_{AL} mg/kg	Humus %
S1	7.75	0.2927	28.00	140	1.372
S2	8.08	0.1849	56.00	228	3.993
S3	8.43	0.2143	49.33	80	0.998
S4	7.30	0.2493	87.62	400	7.613
S5	8.57	0.1789	62.00	216	1.372

It can be noticed that pH values range of between 7.30-8.57, being considered as very weak to strong alkaline soil reaction. The analyses concerning salt contents indicated that samples correspond to a non-saline soil type.

While Fadhel and Boussaid (2004) have specified that *M. pulegium* grows best in hydromorphic soils with a pH of 5 to 8.5, DeBaggio and Tucker (2009) recorded for this species a pH range 4.8-8.2, with a mean of 6.9. Thus, the pH levels of our soil samples show higher values than average mentioned by specialised literature.

Mobile form of phosphorus (P_{AL}) was classified as middle content for S1, high content for S2, S3 and S5 and very high for S4. Potassium content (K_{AL}) was middle for S3, normal for S1, high for S2 and S5 and very high for S4 (Madjar, 2008).

In the monograph of Ilgani village it is mentioned that alluvial soils have at most 5% humus (Moţoc & Manole, 2015). Concerning humus content, our results indicated wide variations, the highest value being found for S4 which is considered as good content (Davidescu & Davidescu, 1999).

With respect of humus, phosphorus and potassium content, S4 proves the highest values of all sample soils. This indicates that in SP 4, which corresponds to a ruderal area, located at the western edge of the Ilganii de Sus village, the soil is strong alkaline. This result could be explained by the accumulation, in that specific area, of many domestic wastes, bird feathers and poultry manure, that seem to contribute to the increase in phosphorus and potassium content of the soil. In addition, it was already demonstrated that poultry litter represent a good organic fertilizer for plants (Kobierski et al., 2017).

Categorized as least concern by IUCN Red List of Threatened Species, pennyroyal is a hemicryptophyte that typically occurs in freshwater wetlands, seasonally inundated grassland, ponds, ditches, roadsides, disturbed sites, ephemeral watercourses and abandoned fields, particularly on reasonably fertile soils and usually in very short open vegetation often which is grazed (De Belair et al., 2014; http://naturalhistory.museumwales.ac.uk/; http://www.iucnredlist.org/; http://www.cabi.org/isc/).

This perennial species is suitable for sandy, loamy and clay soils and can grow in heavy, sometimes silty clay soil; it can grow in semi-shade or without shade, preferring moist soil (http://www.pfaf.org/; http://www.cabi.org/).

Considering the report of Covaliov et al. (2012), pennyroyal is a perennial species, flowering from July to September, with a frequent spreading degree. According to Ion and Ion (2007), *M. pulegium* is defined, among other characteristics, by the flowering time and duration correlated to the level of soil moisture.
In the studied areas, most often, pennyroyal was found next to cocklebur (*Xanthium* sp.), an invasive plant that dominates the pastures of the village, where cattle go daily to the pasture. Besides, pennyroyal itself was regarded as a potential environmental weed in some regions of the world and was characterized as a plant with aggressive spreading nature, which takes over the land where it grows (http://www.floralencounters.com/Seeds/; https://keyserver.lucidcentral.org/weeds/). This could also be the case of the habitats studied by us in the present research. While some authors emphasized the poisonous role of pennyroyal for cattle forage (Amsberry & Meinke, 2008), others mentioned that in animal therapy, *Mentha pulegium* has proved its tonic effect on the uterus of ruminants (Laudato & Capasso, 2013).
It is known that pennyroyal can survive some drought, likes full sun, rich soil and some moisture (http://www.floralencounters.com/). The weather in August 2017 was characterized by very high temperatures (even 35^0C) that have attracted the sharp drying of areas usually flooded at Ilganii de Sus. As Koetlisi (2013) suggested, a water deficit and drought stress may result in smaller leaf and reduce yield of aromatic plants.

CONCLUSIONS

This paper discusses for the first time some soil characteristics at Ilganii de Sus from Tulcea County, a region where pennyroyal grows as an aromatic plant with pest potential.
Mentha pulegium was found in different types of habitats, as: ruderal areas, flooded ditches or sandy beaches.
The highest values of humus content, pH, phosphorus and potassium were associated to the western part of the study area, which correspond to a zone rich in domestic waste and poultry manure.
Soil agrochemical parameters determined at Ilganii de Sus, Tulcea County, reveals growth requirements of *Mentha pulegium*, soils with slightly to strongly alkaline pH, low to moderate humus content and moderate to very high content in mobile forms of phosphorus and potassium.

ACKNOWLEDGEMENTS

This article was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Amsberry, K., Meinke, R. (2008). Evaluating allelopathic effects of pennyroyal (*Mentha pulegium*) on two native plant species. *Oregon Department of Agriculture, Native Plant Conservation Program*, https://www.oregon.gov/ODA/shared/Documents/Publications/PlantConservation/AllelopathyEffectsPennyroyal.pdf.

Anastasiu, P., Liţescu, S. (2012). Preliminary study on the flora of the Snagov Lake Natural Reserve and its surroundings. *Acta Horti Bot. Bucurest*, *39*, 69–90.

Andro, A.R. (2012). Physiological and biochemical comparative research on taxa of the genus *Mentha* L, PhD thesis, "Alexandru Ioan Cuza" University of Iaşi Faculty of Biology.

Bakerink, J.A., Gospe, S.M., Dimand, R.J., Eldridge, M.W. (1996). Multiple Organ Failure After Ingestion of Pennyroyal Oil From Herbal Tea in Two Infants. *Pediatrics, 98*(5), 944–947.

Batsatsashvili, K., Mehdiyeva, N., Fayvush, G., Kikvidze, Z., Khutsishvili, M., Maisaia, I., Sikharulidze, S., Tchelidze, D., Aleksanyan, A., Alizade, V., Paniagua Zambrana, N.Y., Bussmann, R.W (2016). *Mentha aquatica* L., *Mentha longifolia* L., *Mentha pulegium* L., *Ethnobotany of the Caucasus, European Ethnobotany*, R.W. Bussmann (ed.).

Brahmi, F., Khodir, M., Cjibane, M., Duez, P. (2017). Chemical Composition and Biological Activities of Mentha Species, *Aromatic and Medicinal Plants - Back to Nature*, http://www.intechopen.com/books/.

Covaliov, S., Doroftei, M., Negrea, B.M. (2012). Assessment of vegetal resources in Danube Delta, Romania. *AAB Bioflux*, *4*(2), 57–72.

Davidescu, V., Davidescu, D. (1999). *Compendium agrochimic*. Ed.Academiei Române.

DeBaggio, T., Tucker, A.O. (2009). *The Encyclopedia of Herbs: A Comprehensive Reference to Herbs of Flavor and Fragrance*. Timber Press, Inc. London.

De Belair, G., Rhazi, L., Lansdown, R.V. (2014). *Mentha pulegium*. *The IUCN Red List of Threatened Species* 2014: e.T164256A42395474. http://dx.doi.org/10.2305/IUCN.UK.2014-1.RLTS.T164256A42395474.en.

Dobrin, I., Săndulescu, E.B., Stavrescu-Bedivan, M.M. (2013). Summer field trip to Sfântu Gheorghe branch at Ilganii de Jos (Tulcea County, Romania): a naturalistic approach. *Agrolife Scientific Journal*, *2*(2), 79–82.
Domokos, E., Cristea, V. (2013). The woody vegetation in the middle stream of the Niraj Valley (Romania, Mureş County). *Plant Develop*, *20*, 149 – 162.
Fadhel, N.B., Boussaïd, M. (2004). Genetic diversity in wild Tunisian populations of *Mentha pulegium* L. (*Lamiaceae*). *Genetic Resources and Crop Evolution*, *51*(3), 309–321.
Gavra, C. (2015). Study regarding the halophilous vegetation of the Crişurilor Plain (North Western Romania). *Analele Universității din Oradea, Fascicula: Ecotoxicologie, Zootehnie şi Tehnologii de Industrie Alimentară, XIV/A*, 339–349.
Gerenutti, M., Modesto, L., Carrara, V.A., Megalhaes, S.A., de Freitas, N.P., Silva, M.G. (2014). Maternal exposure to aqueous extract of *Mentha pulegium* L.inducing toxicity to embryo development in rats. *Afr. J. Pharm. Pharmacol*., *8*(22), 609–614.
Grigore, M.N. (2008). *Halofitotaxonomia. Lista plantelor de sărătură din România*. Iaşi: Ed. Pim.
Hadi, M.Y., Hameed, I.H., Ibraheam, I.A. (2017). *Mentha pulegium*: Medicinal uses, Anti-Hepatic, Antibacterial, Antioxidant effect and Analysis of Bioactive Natural Compounds: A Review. *Research J. Pharm. and Tech., 10*(10), 1827–1831.
Holzer, S. (2012). *Permacultura. Ghid practic pentru agricultura la scară mică*. https://cartidintei.files.wordpress.com/2013/05/01-sepp-holzer-permacultura-ghid-practic-pentru-agricultura-la-scarc483-micc483-v-compactc483.pdf
Ion, N., Ion, V. (2007). Melliferous charactheristics of spontaneous Lamiaceae species, identified in the Danube Valley. *Lucrări ştiinţifice Zootehnie şi Biotehnologii*, *40*(2), 71–79.
Irimia, I., Danu, M.A. (2010). Contributions to the paludal vegetation study from the Vaslui river basin. *AAB Bioflux*, *2*(1), 1–8.
Jiboc (Coste), A.M. (2014). Study of Jumco Inflexi-Menthetum Longifoliae association in the Oas Mountains (North Western Romania). *Analele Universității din Oradea, Fascicula: Protecţia Mediului, XXIII*, 457–462.
Kanakis, C.D., Petrakis, E.A., Kimbaris, A.C., Pappas, C., Tarantilis, P.A., Pollisiou, M.G. (2011). Classification of Greek *Mentha pulegium* L. (Pennyroyal) samples, according to geographical location by Fourier Transform Infrared Spectroscopy. *Phytochemical Analysis, 23*(1), 34–43.
Kobierski, M., Bartkowiak, A., Lemanowicz, J., Piekarczyk, M. (2017). Impact of poultry manure fertilization on chemical and biochemical properties of soils. *Plant Soil Environ*., *63*(12), 558–563.
Koetlisi, A.K. (2013). Influence of Soil Texture, Water Management and Fertilizer N on the Biomass Production and Antimicrobial Properties of *Mentha longifolia* L. Master Thesis, 124.
Laudato, M., Capasso, R. (2013). Useful plants for animal therapy. *OA Alternative Medicine, 1*(1), 1.
Madjar, R.M. (2008). *Agrochimie, planta şi solul*, Editura INVEL-Multimedia.
Mansoori, I. (2014). The effect of plant density and harvesting time on growth and essential oil of peppermint (*Mentha piperita* L.). *Journal of Medical and Bioengineering, 3*(2), 113–116.
Miraj, S., Kiani, S. (2016). Study of pharmacological effect of *Mentha pulegium*: A review. *Der Pharmacia Lettre*, *8*(9), 242–245.
Moţoc, C., Manole, G., 2015. *Monografia satelor Partizani şi Ilganii de Sus*. Iaşi: Zoom print and copy center.
Neacşu, A., Arsene, G.G., Faur, F., Imbrea, I., Nicolin, A. (2015). Vegetal comunities from the Class *Isoëto-Nanojuncetea* BR.-BL. et TX. 1943 from Banat. *Research Journal of Agricultural Science*, *47*(3), 192–201.
Oprea, A. (2004). Forest vegetation in the Tecuci Plain (Galaţi county). *Buletinul Grădinii Botanice Iaşi*, *12*, 53–74.
Paşca, C., Mărghitaş, L., Dezmirean, D., Bobiş, O., Bonta, V., Chirilă, F., Matei, I., Fiţ, N. (2017). Medicinal Plants Based Products Tested on Pathogens Isolated from Mastitis Milk. *Molecules*, *22* (1473), 1–16.
Prodan, M., Dăneţ, C., Imbrea, I. (2010). Family Lamiaceae: Main valorisable medicinal and aromatic species in the Aninei Mountains (Caras-Severin County, Romania). *Research Journal of Agricultural Science*, *42*(4), 129–132.
Răduţoiu, D., Stan, I., Costache, I. (2014). *Oenanthe pimpinelloides* L. in the flora of Romania. *Contribuţii Botanice*, *XLIX*, 61–66.
Rocha, D., Novo, M., Matos, O., Figueiredo, A.C., Delgado, M., Cabral, M.D., Liberato, M., Moiteiro, C. (2015). Potential of *Mentha pulegium* for mosquito control. *Revista de Ciências Agrárias*, *38*(2), 155–165.
Silva, L.F., Cardoso, M., Batista, L.R., Gomes, M., Avelar Rodrigues, L.M., Selvati Rezende, D., Teixeira, M.L., Sousa Carvalho, M., Santiago, J., Nelson, D.L. (2015). Chemical Characterization, Antibacterial and Antioxidant Activities of Essential Oils of *Mentha viridis* L. and *Mentha pulegium* L. *American Journal of Plant Sciences*, *6*. 666–675.
Stringaro, A., Colone, M., Angiolella, L. (2018). Antioxidant, Antifungal, Antibiofilm, and Cytotoxic Activities of *Mentha* spp. Essential Oils. *Medicines* *5*(112), 1–15.
Tănase, C., Ştefan, N. (2010). Contributions to the study of flora and vegetation from Basin Valea Morisca. Analele ştiinţifice ale Universităţii „Al. I. Cuza“ Iaşi tomul LVI, fasc. 1, s. II a. *Biologie vegetală*, 87–98.
Tomescu, C.V., Chifu, T. (2009). The Vascular Flora from the Suceava River Basin (Suceava County). *Analele Universității „Ştefan Cel Mare“ Suceava*, *1*, 31.
Zekri, N., Amalich, S., Boughdad, A., Alaoui, El Belghiti, M., Zair, T., 2013. Phytochemical study and insecticidal activity of *Mentha pulegium L.* oils from Morocco against *Sitophilus oryzae. Mediterranean Journal of Chemistry, 2*(4), 607–619.

***https://www.poison.org/articles/2016-mar/pennyroyal-oil.
***https://livertox.nih.gov/Pennyroyal.htm.
*** http://www.pfaf.org/.
***http://naturalhistory.museumwales.ac.uk/.
*** http://www.iucnredlist.org/.
***http://www.cabi.org/isc/.
***http://www.floralencounters.com/Seeds/.
***https://keyserver.lucidcentral.org/weeds/.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESEARCH ON THE ACTION OF ROM-AGROBIOFERTIL NP BIOFERTILIZER ON AGRICULTURAL CROPS

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Abstract

The use of intensive chemistry in the agricultural field was a first cause of pollution in the agricultural field. In order to move towards sustainable, environmentally friendly agriculture, farmers took into account the use of alternatives to chemical fertilizers, green, green and non-polluting alternatives. Thus, so-called ecological fertilizers, green fertilizers, soil-friendly, agricultural crops, production and, implicitly, for humans and animals have emerged. The alternative to chemical fertilizers is fertilizer based on bacterial cultures. In agriculture, the main source of nutrition is the nutrients. These are the basis for the growth and development of cultural plants, and they are indispensable to any form of life. On the other hand, the amount of nutrients at some point in time may decrease or increase depending on certain pedo-climatic factors, soil typology, the ability of plants to return some of the nutrients consumed (by the decomposition of crop residues) and so on.

Key words: *biofertilisers, Azospirillium lipoferum, Azotobacter chroococcum, Bacillus megaterium, Rom-Agrobiofertil NP.*

INTRODUCTION

Biofertilizers are organic products that consist of living microorganisms (bacteria, fungi, fungi etc.), these biofertilizers providing the necessary microelements in the soil, better growth and development of agricultural plants, as well as obtaining healthy and higher yields and quantity of products) compared to chemically fertilized crops. Biofertilizers, being liquid substances, can be applied either on soil, seeds or on the surface of plants so that the microorganisms in their composition fit the best. Once these microorganisms are fixed on the organs of the plants, together with their activity there will be a contribution of elements necessary for the growth and development of the agricultural crops (Sunita et al., 2018).

Rom-Agrobiofertil NP is a fertilizer based on three bacterial strains: *Azospirillium lipoferum, Azotobacter chroococcum* and *Bacillus megaterium*. Each bacterium has its own role as well as action on soil and crops. Thus, *Azotobacter chroococcum* has the role of capturing atmospheric nitrogen on the basis of certain resources and exchanges of energy between the soil and the environment, fixing in the soil. In addition to this role, this bacterium also has the role of metabolizing phosphates in the soil, assimilating root exudates, and counteracting certain plant-damaging bacteria, existing bacteria in the soil (Levandovschi et al., 2017).

Azospirillium lipoferum has the role of metabolizing organic matter from soil to nutrients assimilable by plants (metabolism of cellulose, hemicellulose, lignin etc.). At the same time, as azotobacter, azospirillium is designed to capture nitrogen in the atmosphere and to place it in the soil. Under certain environmental and soil conditions, *Azospirillum* can positively influence plant growth, crop yields and N-content of the plant. This plant stimulatory effect exerted by *Azospirillum* has been attributed to several mechanisms, including biological nitrogen fixation and auxin production (Steenhoudt et al., 2000).

The mode of action of *Azospirillum* is most probably composed of multiple mechanism. The increased use of the various biological process in soil will decisively contribute to

make agriculture more productive with less harm to the environment (Attilla et al., 2010).
The last bacterium of the biofertilizator, *Bacillus megaterium*, plays a role in the decomposition of soil detriment, the solubility of insoluble phosphates from the soil into soluble phosphates, but also produces certain organic acids necessary for the growth and development of plants (lactic acid, glutamic acid etc.) and enzymes and minerals the role of acting as growth incentives for plants and agricultural production (Sbîrciog et al., 2017).
The isolation of phosphate solubilizing bacterial strains exhibiting high ability to solubilize soil phosphorus is a matter of great interest with high applicability. The use of phosphate solubilizing bacteria as inoculants simultaneously increase phosphate uptake by the plant and increase crop yield, Strains from the genera *Pseudomonas*, *Bacillus* and *Rhizobium* species are the most powerful phosphate solubilises (Patel et al., 2016).
Research on soil has found that in order to replace the chemical elements in the composition of classical fertilizers, researchers can use numerous bacteria found in the soil structure. Thus, following laboratory and open field tests, it has been found that by combining bacteria, much better results can be obtained than when applying a chemical fertilizer. Using these research and results on a large scale, researchers have created biofertilizers to reduce soil pollution, increase crop production, and protect human health.

MATERIALS AND METHODS

The first research on biofertilizers of Romvac Company S.A. was carried out at a research farm in agriculture, namely Vegetable Development Research Station Buzău. Within this resort, alongside Romvac collaborators Dr. Biol. Floarea Burnichi (Scientific Secretary of SSC Buzau) and Drd. Ing. Constantin Petre (Head of Mechanization Department of Vegetable Development Research Station Buzău) established the crops to which the Rom-Agrobiofertil NP biofertilizer was applied (cabbage and tomatoes).
Thus, in November 2017 the semicircular cabbage culture was ished, the "Buzau" variety in field tomatoes "Buzău 1600" and "Florina 44". In the spring of 2018 the first tranche of this biofertilizator was administered at a dose of 5 l/ha (3 types of product x 5 = 15 l/ha) for each crop.
The second research on biofertilizers of Romvac Company S.A. was carried out at a research farm in agriculture, namely Research and Development Station for Plant Culture on Sands Dăbuleni. Within this resort, alongside Romvac collaborators Dr. Ing. Aurelia Diaconu (General Manager of Research and Development Station for Plant Culture on Sands Dabuleni) and Dr. Ing. Milica Dima established the crops to which the Rom-Agrobiofertil NP biofertilizer was applied.

RESULTS AND DISCUSSIONS

Developing unit. Vegetable Development Research Station Buzau. *Main features*. The seedlings have a green, green-violet coloration. At planting, the seedling must have 40 days. Characteristic of seedling planting is the height of the plant, a mean height of 55-60 cm.
The diameter of the leaf rosette is high, ranging from 90-100 cm. The position of the rosette leaves is half-erect. The leaf is medium to large (27-29 cm). The base leaves are between 30 and 35 cm high, with a 12-15 cm long petiole, with 1-3 bracts. The leaf is greenish-bluish, elongated to the basal leaves and rounded to the middle leaves and top of the rosette.
The edges of the tongue are medium-wavy, slightly creased. The surface of the tongue is slightly embossed, with a prominent rib. The leaf is well covered with pruin. The diameter of the head records a medium variation with variation limits between 23 and 25 cm.
The height of the head has a medium variability, with limits between 17 and 19 cm. The shape index is between 0.7-0.9, with globular-capped head forms. Head weight fluctuates between 2.0-3.5 kg. The head is full, covered with leaves. Position of the rosette after the formation of the head: does not cover the head. The outer cochlea is 4-6 cm. The inner cochlea is medium, between 5.0 and 6.5 cm. The color of the leaves inside the head is yellowish-white. Leaves inside the head are very fine and thin. Semi-late variety, with a vegetation period of 135-140 days. The range from planting to maturity is 95-100 days. The

production potential is 70-80 tons/ha, under normal crop conditions, it is easy to achieve 50-60 tons/ha.

Crack resistance: 10-15 days after consumption maturity. This variety of cabbage is cultivated in all parts of the country, in southern areas it is cultivated in successive culture. "Buzău" variety shows good resistance to pathogen attack. The growing period of this variety is: at maturity (130 days) 14.06-20.10 and planting - maturity (95 days) 18.07-20.10.

Production destination (Table 3): a well-known and well-known variety for fine leaves, very well-suited for fresh consumption or preserved by marinating (from the leaves of this variety, from the 1950s to the 1960s, traditional sausages are prepared).

Experimental variants: V_1 - Unfertilized (Control - Table 1, Figure 1) and V_2 - Rom-Agrobiofertil NP 5 l/ha (3 x 5 = 15 l - Table 2, Figure 2).

Table 1. Scheme of treatment for autumn cabbage culture "Buzău" variety

Scheme of cultivation of cabbage seedlings in the field					
No. treat.	**Date of application of the treatment**	**Applied product type**	**Name bacterial strain**	**Approved dose**	**The dose used**
1	4/25/2018	Organic fertilizer	*Azospirillum lipoferum*	5.0 l/ha	5.0 l/ha
		Organic fertilizer	*Azotobacter chroococcum*	5.0 l/ha	5.0 l/ha
		Organic fertilizer	*Bacillus megaterium*	5.0 l/ha	5.0 l/ha

Table 2. Biometric data recorded in autumn cabbage seed crop "Buzău" variety

Variant	**No. of the main shells**	**Average length of silicone**	**No. of silicone / plant media**	**No. seed / silica medium**	**Average seed / plant**	
					Nr. seeds / plant	**g/pl**
V_1 – Control	21	6.25	209	24	2021	8.31
V_2 - Rom-Agrobiofertil NP 5 l/ha x 3 = 15 l/ ha	23	7.80	424	29	6564	44.4
Growth like V_1 Control (%)	9.52%	24.8%	102.87%	20.83%	224.79%	434.3%

Table 3. Seed production at Autumn Cabbage semicircle "Buzau" variety, kg/ha

Variant	**Average seed yield kg/ha**	**Selling price/kg**	**Total value of the seeds lei**	**MMB g**	**Number of seeds/1 g**
V_1 – Control	422.46	500	211230	4.11	243.0
V_2 - Rom-Agrobiofertil NP 5 l/ha	1171.76	500	585880	6.76	147.8
Growth like V_1 Control (%)	177.4%			64.5%	

Developing unit. Vegetable Development Research Station Buzau. The year of approval of this variety was in 1977.

Main features. Semi-sweet variety of tomatoes for fresh consumption. The period of vegetation from the east to the consumer maturity of the first fruits is 125-130 days.

The plants have indeterminate growth, the height of 120-130 cm, vigorous, with characteristic green leaf. The fruit is large, firm, with an average weight of about 200 g (150-220 g), spherical, taste particularly pleasant due to its balanced sugar-acidity ratio. The surface of the fruit is smooth, uniform red. It is tolerant to the attack of the main pathogens.

Economic efficiency. The production potential is 60-80 t/ha; if improved technology is applied, production can grow up to 150 t/ha.

Scope. Fruits are intended for fresh consumption, in which case they are harvested in the spring stage and for industrialization (juices, paste etc.). It can be cultivated successfully in all areas favorable to tomato culture, being recommended primarily for households.

Potential beneficiaries. Vegetable Units and Private Producers.

Figure 1. Semi-cabbage crop, "Buzău" variety - control batch

Figure 2. Semi-sweet cabbage crop, "Buzău" variety - fertilized with Rom-Agrobiofertil NP

Figure 3. Semi-sweet cabbage crop, "Buzău" variety - differences between control and variant fertilized with Rom-Agrobiofertil NP

Results obtained in field tomato variety "Buzău 1600"

The culture was set up in May, in a gauntlet system, on the scrubber. Rom-Agrobiofertil NP was administered in spring, the second decade of May, in vegetation at a dose of 5 l/ha (3 x 5 = 15 l/ha). *Cultivation desimetry*: 30000 plants/ha. Fruit weight: 120-135 g. Experimental variants: V_1 - Control, V_2 - Fertilization variant with Rom-Agrobiofertil NP 5 l/ha x 3 types.

Developing unit. Buzau Vegetable Research and Development Station. *Variety*: tomatoes "Buzău 1600". *Characteristics of tomato variety,* "Buzău 1600" (Figure 4): Semi-sweet variety, vegetation period (sunrise on consumption) 125-130 days, is tolerant to attack by major pathogens and use in households. *Economic efficiency:* production of 60-80 t/ha, increase production to approx. 150 t/ha. *Usage area*: fresh consumption, for industrialization (juices, paste etc.).

It presents a positive response to Rom-Agrobiofertil NP biofertilizer, increase by approx. 1.4% against the control group of the crop plants, increase by approx. 6.8% against the control group, weight of the fruit on the plant with approx. 26.2%, the weight of the fruit in the variant fertilized with Rom-Agrobiofertil NP by approx. 6.8% (Table 4).

Table 4. Biometric data recorded on the "Buzău 1600" field tomato crop located on the trellis

Variant	Height of plant (cm)	No. inflorescences/ plant	Medium length (cm)	No. fruit / plant	Average fruit weight (g)	Total fruit production (t/ha)	TGW (g)	No. of seeds / 1 g
V_1 – Control	145.9	7.3	21.8	8.4	123.9	31.2	2.7	372.7
V_2 - Rom-Agrobiofertil NP 5 l/ha x 3	148.0	7.8	23.8	10.6	132.3	42.1	3.5	293.0
Growth with V1 Control (%)	**+1.4**	**+6.8**	**+9.2**	**+26.2**	**+6.8**	**+34.9**	**+9.6**	

Figure 4. "Buzău 1600" variety - fertilized with Rom-Agrobiofertil NP

Rom-Agrobiofertil NP effectiveness testing on peanut culture 2018

During vegetation, determinations were made on the plant's height, the number of shoots on the plant, the number of nodules per plant, and production determinations were made at harvesting (production was weighed on each variant and reported per unit area).

In the year 2018, the Rom-Agrobiofertil NP product was produced with 1402 kg/ha of pasta, surpassing the control variant with an significant production increase of 5.9% due to unfavorable weather conditions (abundant rainfall). It is recommended to resume the experiment so as to determine the action of Rom-Agrobiofertil NP biofertilizer on peanut culture (Table 5).

Table 5. Biometric determinations of peanuts under the influence of fertilization with Rom Agrobiofertil NP

Variant	Production of pods (kg/ha)	Relative production (%)	The difference from the control (kg/ha)
Control	1324	100	Ct.
Treated with Rom-Agrobiofertil NP	1402	105.9	+78

The plant's height was 15 cm in the untreated version and 19 cm in the variant treated with Rom-Agrobiofertil NP. The number of shoots per plant is the same in both treated and untreated versions. The number of nodosities is higher in the treated version (41 nodosities) compared to the untreated variant 37 (Table 6).

Table 6. Influence of fertilization with Rom-Agrobiofertil NP on peanut production

Variant	Plant height (cm)	Number of shoots/plant	Number of nodules/plant
Control	15	7	37
Treated with Rom-Agrobiofertil NP	19	7	41

Rom-Agrobiofertil NP effectiveness testing on potato culture 2018

Under the conditions of year 2018, the Rom-Agrobiofertil NP product was produced with a production of 1402 kg/ha of pasta, exceeding the control variant with an insignificant production increase of 5.9%.

The Rom-Agrobiofertil NP product was tested on three varieties: "Belarosa", "Riviera" and "Carera", among variants studied production differences were insignificant, statistically uninsured. Analyzing the influence of the treatment on the commercial production, the following results were obtained: 41.95 t/ha untreated and 45.81 t/ha in the plants treated with Rom-Agrobiofertil NP.

By analyzing the interaction between the variety and the treatment, an additional (commercial) production was obtained in the variants to which the product was applied:

+1.93 t/ha in the "Bellarosa" variety;
+8.21 t/ha in the "Riviera" variety;
+1.45 t/ha in the "Carera" variety.

CONCLUSIONS

All of the cultures responds positively to the action of bacteria in the bio-fertilizer content of Rom-Agrobiofertil NP.

The height of the plants showed an increase of approx. 1.4% against the unfertilized V_1 control. Increase of inflorescence in V_2 culture with approx. 6.8% vs. V_1. The increase in the number of fruit per plant on the V_2 crop with approx. 26.2%. Increase in the average weight of the group treated with Rom-Agrobiofertil NP (V_2) with approx. 6.8% vs. V_1 on the cabbage culture. It is recommended to resume the experiment for its long-term validation. It is necessary to repeat the experiments in order to validate the results obtained and to diversify them by introducing into study other vegetable species, in order to improve the applied crop technologies and to increase the economic efficiency of the crops.

Cabbage production

The recorded production (production increase) is very high (177.4%), from 422.5 kg/ha to 1171.8 kg/ha. The increase in length of silicve with approx. 24.8% against the non-fertilized V_1 control. Increasing the number of silicas per plant to V_2 by 102.9% compared to V_1. The increase in the number of seeds on silicone in variant V_2 by 20.8%.

Increase of the number of seeds per plant at the variant fertilized with Rom-Agrobiofertil NP by 224.79% against the control. Increase of the weight and average seed quantity per plant, from 8.3 g/plant to V_1 Control to 44.4 g/plant in variant fertilized with Rom-Agrobiofertil NP. To resume the experiment for its long-term validation.

"Buzău 1600" tomato culture on the trellis

It presents a positive response to Rom-Agrobiofertil NP biofertilizer, increase by approx. 1.4% against the control group of the crop plants, increase by approx. 6.8% against the control group, weight of the fruit on the plant with approx. 26.2%, the weight of the fruit in the variant fertilized with Rom-Agrobiofertil NP by approx. 6.8%.

Peanut production

Under the conditions of 2018 (a year full of precipitation in which the soil was saturated with water and the peanut culture was affected by the surplus water stored in the soil), the of Rom-Agrobiofertil NP product achieved a production of 1402 kg/ha, exceeding the control variant with an insignificant production increase of 5.9%.

Potato production

Potato culture responded well to the Rom-Agrobiofertil NP biofertilizer. The plants in the batch treated with this biofertilizer showed an increase in the waist, an increase in the vegetal mass, and an inflorescence richer than the control group. The recorded production was different because three potato varieties were planted.

REFERENCES

Attilla, H.I., Alhasin, A.M., Nasib, M.A., Ghazalim A.H., Zakaria, L., Jais, H.M., Balal, I.A.A, Salleh, B. (2010). Occurrence and microbiological caracteristics of *Azospirillum* strain associated with leguminous and non- leguminous plants in Al Jabal Al Akhdar Eco region, Lybia. *American-Eurasian J. Agric. & Environmental Sciences*, *8*(6), 617–625.

Levandovschi, N., Chiurciu, C., Chitonu, P. (2017). Rom-Agrobiofertil NP - biological fertilizer for a sustainable agriculture. Sao Paulo, Brazil: 2nd World Biotechnology Congress, December 04-06. *Journal of Biotechnology & Biomaterials*, *7*(6, Suppl), 69, ISSN 2155-952X, DOI: 10.4172/2155-952X-C1-086.

Patel, G., Singh, S., Saxena, S.K., Kaur, K.J. (2016). Isolation, biochemical characterization and production of biofertilizer from *Bacillus megaterium*. *International Journal of Life Sciences Scientific Research*, *2*(6), 749–752.

Sbîrciog, G., Șovărel, G., Costache, M. (2017). Stimulating effect of Rom-Agrobiofertil NP fertiliser on tomato, cucumber and cauliflower crops. *Agriculture & Food, Vol. 5*.

Steenhoudt, O., and Vanderleyden, J. (2000). *Azospirillum*, a free-living nitrogen-fixing bacterium closely associated with grasses: genetic, biochemical and ecological aspects. *FEMS Microbiology Reviews*, *24*. 487–506.

Sunita, G. (2013). *Pseudomonas striata* for Improving Phosphorus Availability in Soil under Pearl Millet Cultivation. *J. of Crop Improvement*, *27*(3), https://doi.org/10.1080/15427528.2012.760026.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

AMELIORATIVE, TECHNICAL AND OPERATIONAL SOLUTION OF VINEYARD IN THE CONDITIONS OF A VARIOUS SOIL COVER AND COMPLEX RELIEF

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Abstract

The aim of the study was to propose a method for systematization of a results of exploration of the relief, hydrology and soil cover for the development of a land-cultivating and ameliorative solution of vineyard terroir under conditions of complex erosion-accumulation landscape. A grouping of the relief and hydrology was carried out. The soil cover was investigated by the large-scale mapping method. The established soil differences were mapped and their distribution was compared with the data for the relief and the runoff. As a result, on the terrain were detached sections with different degrees of suitability for vineyards growth. In the terrains with limited suitability were detached zones, requiring a ameliorative effect on one or more of the three components of the landscape-relief, hydrology and soil.

Key words: *terroir, vineyards, water runoff, landscape, erosion.*

INTRODUCTION

The aim of the study was to investigate the relief, hydrology and soil cover of terrains located on the land of Goritsa village, belongs to the Continental-Mediterranean climatic region, Black-sea climate sub-region and the climatic region of Bourgas plain.

The presented study was an attempt to systematize the complex research of a complicated terrain in terms of its topographical and erosion conditions and the suitability for conversion into viticultural terroir.

RESULTS AND DISCUSSIONS

The identification of soil differences and the distribution boundaries of each of them within the object were carried out in a geographic network by drilling. Profiles were drilled in the top by squares with a side of 100 m and for each drilling point was determined soil difference. The results are shown in Figure 1. The distribution of the soil differences correlates with the development of the main catena, as in the direction north - south the soil profiles change from shallow and undeveloped to deep. In a direction approximately normal to that of the main catena, the soils change from carbonate to silicate. In the higher parts of the terrain, the main process of soil degradation was the erosion of the topsoil horizons of the profile, and in some of the low-lying areas there were zones in which the soils tend to rewetting. From a genetic point of view, erosion processes were diagnosed by the degree of reduction of the total depth of the soil profile and the taxonomic level through the boundaries of the distribution of deep and shallow (silicate and carbonate) soils, and the over-humidification - by the expression of the accumulative forms of the relief. In this sense, the soil map integrates the conditions of soil formation and the degree of soil degradation from the genetic plan, but had limited applicability as a prognostic model of soil degradation. Within the boundaries of the same soil diversity were established different topographical conditions for the course of the modern erosion process and over-wetting (Figure 1). For the aims of predicting the degradation processes, have been developed a spatial three-dimensional model of the terrain where the conditions for development of the two main degradation processes reduced to digital characteristics of height, slope and exposure (Gadjev, 2011).

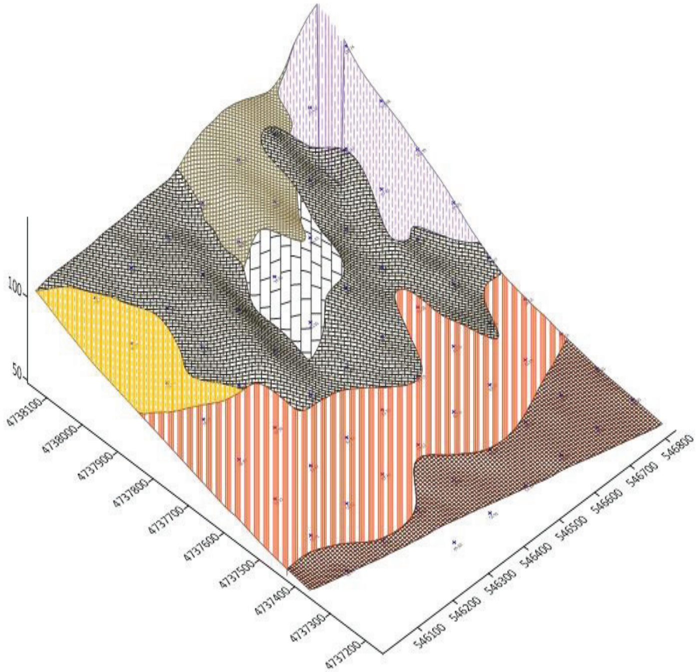

Figure 1. 3D model of the soil cover development, depending on the relief conditions

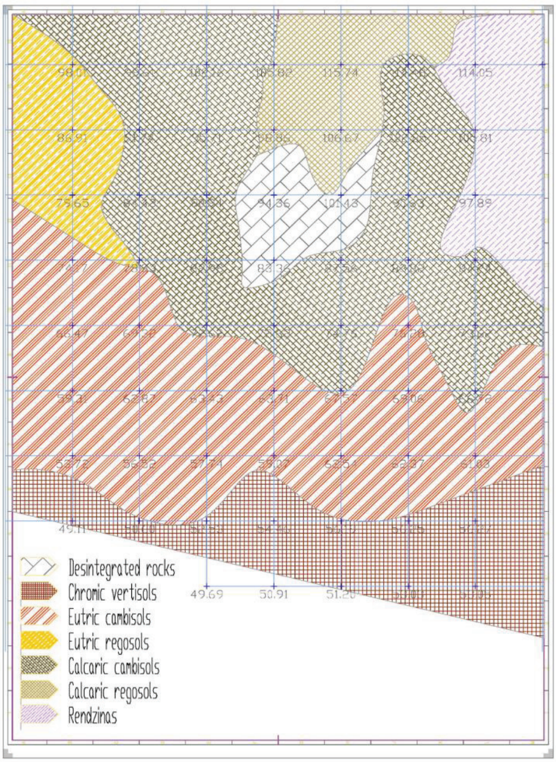

Figure 2. Soil map and location of the drilling profiles, on the basis of which were determined the distribution of the established soil differences in the terrain

Main statistical significance for the degree of erosion activity was the magnitude of the slope, the direction of exposure and some soil characteristics that determined the conditions for filtration or formation of surface runoff (Pannikov, 1983).

The data are shown on the vector mapping scheme in Figure 3.

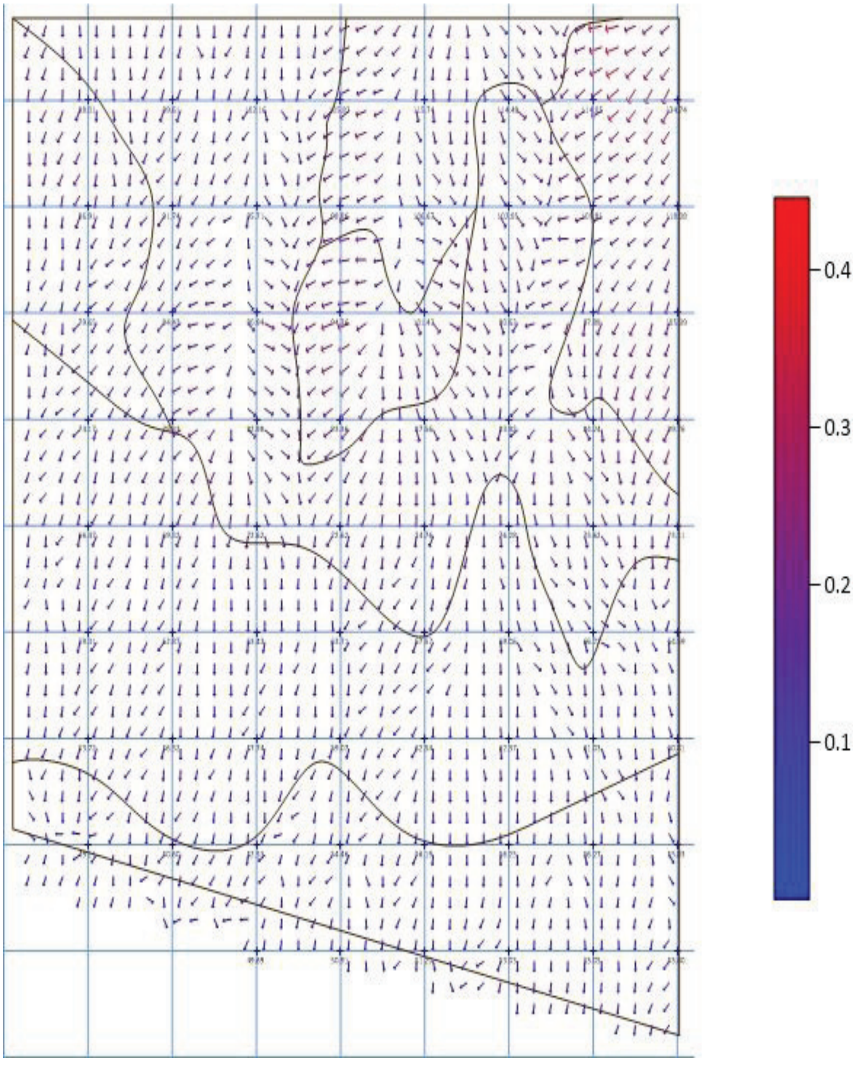

Figure 3. Vector mapping of the probability of forming a surface (red part of the scale) and an underground runoff (blue part of the scale)

The vector diagram on Figure 3 was an integral indicator of soil and topographic conditions. The presented mapping was a conditional visualization of the digital model, in which were integrated the conditions for formation of surface (erosion) and subsoil (in accumulated relief leading to periodic over-humidification) water runoff (Totsev, 2008).

The degree of probability for formation of surface component of the runoff was represented by the scale in the right part of Figure 3, and the dimension of the runoff - by the length of the vectors.

The vector mapping shows that the magnitude of the runoff did not change significantly. This is due to the almost complete coincidence of the flow direction with the general slope of the terrain and the direction of soil catena development.

Mathematically, this coincidence was illustrated by the variogram shown in Figure 4.

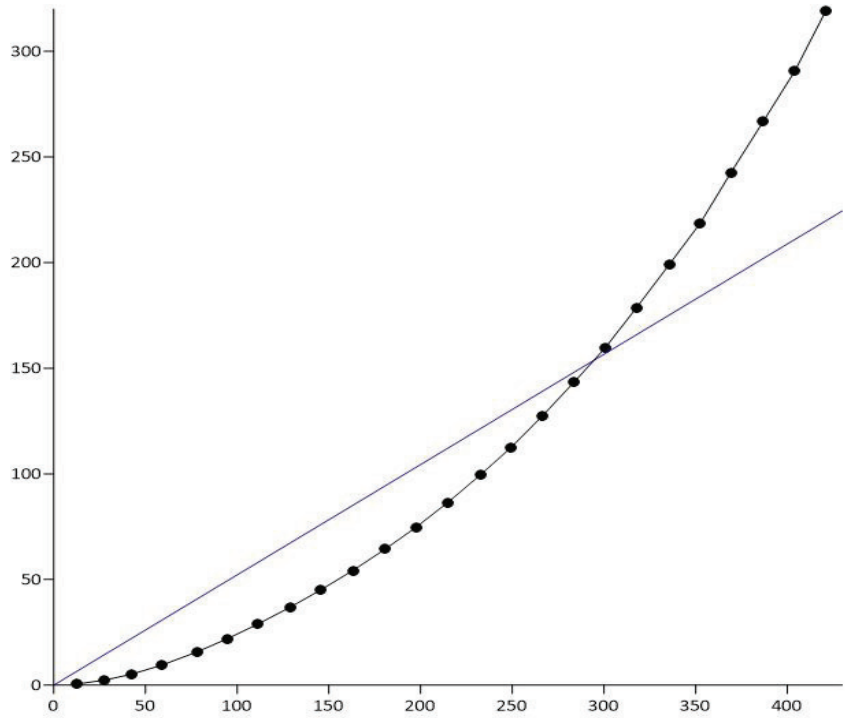

Figure 4. Variogram of terrain hypsometry

The variogram showed the correlation of the integral vector of the runoff with the topographic conditions.

The maximum adequacy of the variogram model was achieved at an angle of 15° NNE and 450 m length.

The probability with which the model presents the generalized conditions for the formation of runoff by dimension and type was 67%.

Based on the data from the digital model and the selected square grid, the topographical conditions were distributed, depending on the slope value and the terrain exposure.

The slope dimension was 3°, and the exposure was 1 rumb or 11.25 azimuth degrees. The distribution of the terrain, depending on these two criteria, is shown in Figures 5 and 6.

In the area where the deep soils were distributed, erosion-dangerous slope was not found. In the area with shallow soils, 60% of the terrain had a slope exceeding 6° therefore the erosion runoff can be predicted (Dimitrov & Konakchiev, 1981).

With regard to the exposure, two basic types of terrain were differentiated - with predominantly eastern and with predominantly western component.

However, the exposure did not directly affect the magnitude of the erosion outflow and had a role to characterize the actual terroir specificity that will form after the transformation of the terrain in vineyard, but in an ameliorative aspect to determine the predicted direction of the surface water runoff and the orientation of the hydroisohypses of the shallow subsoil runoff (Zaydelman, 1996).

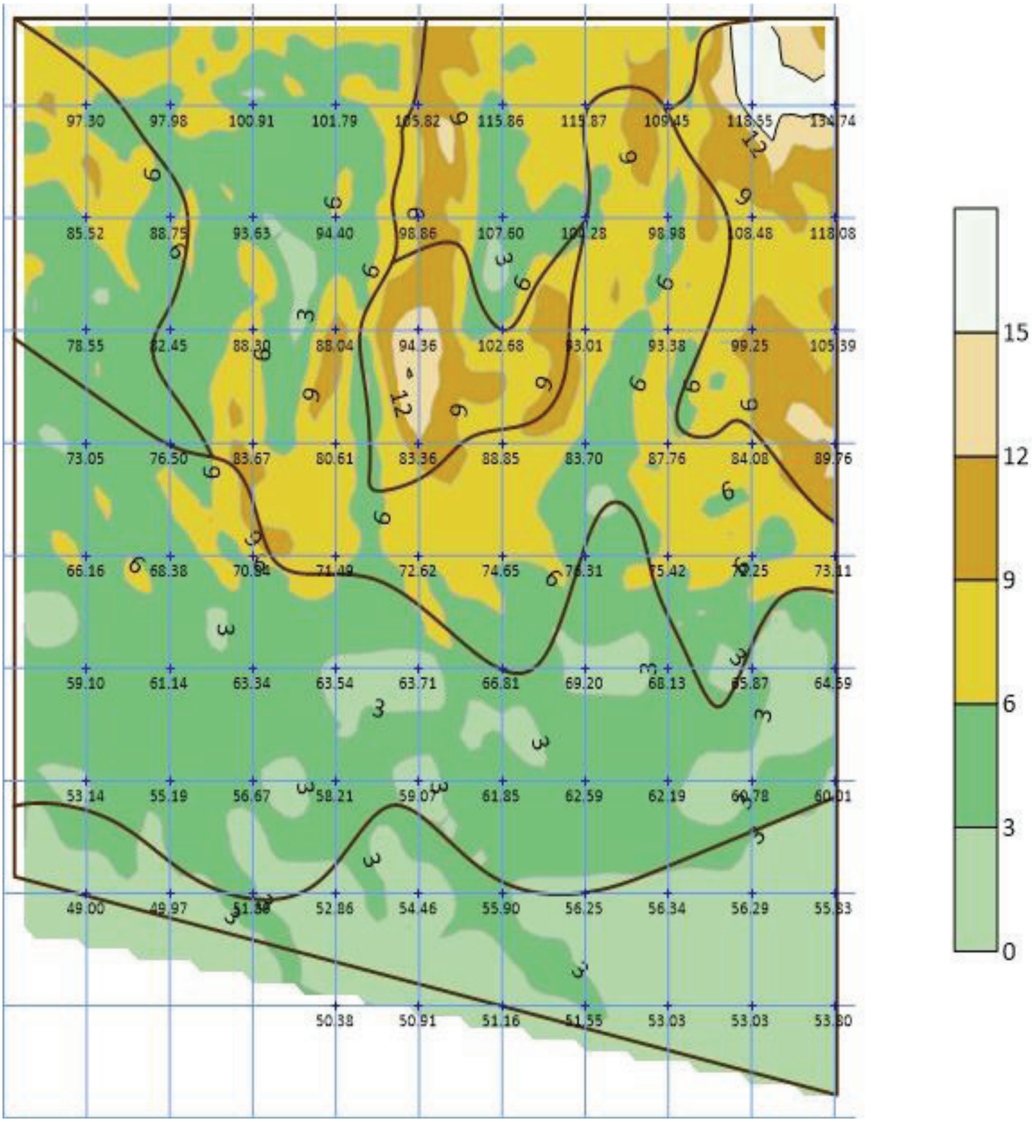

Figure 5. Cartographic of the zones according to the inclination in degrees relative to the horizontal plane

Analysis of the runoff magnitude was done, as to the data for the terrain in the digital model was added and data for the magnitude, the seasonal distribution and the probable extremes of the regional water runoff (Stanev, 1982). The terrain is adjacent to the southern slopes of the eastern parts of Old Mountain. This is supposed to predominate the outward runoff from north. Based on the data, a model for peak runoff was created, as depicted graphically in Figure 7 by the total length, direction and density of the midstream network.

The boundaries of soil differences in the cartographics are shown in contours that correspond to those given on the soil map in Figure 2.

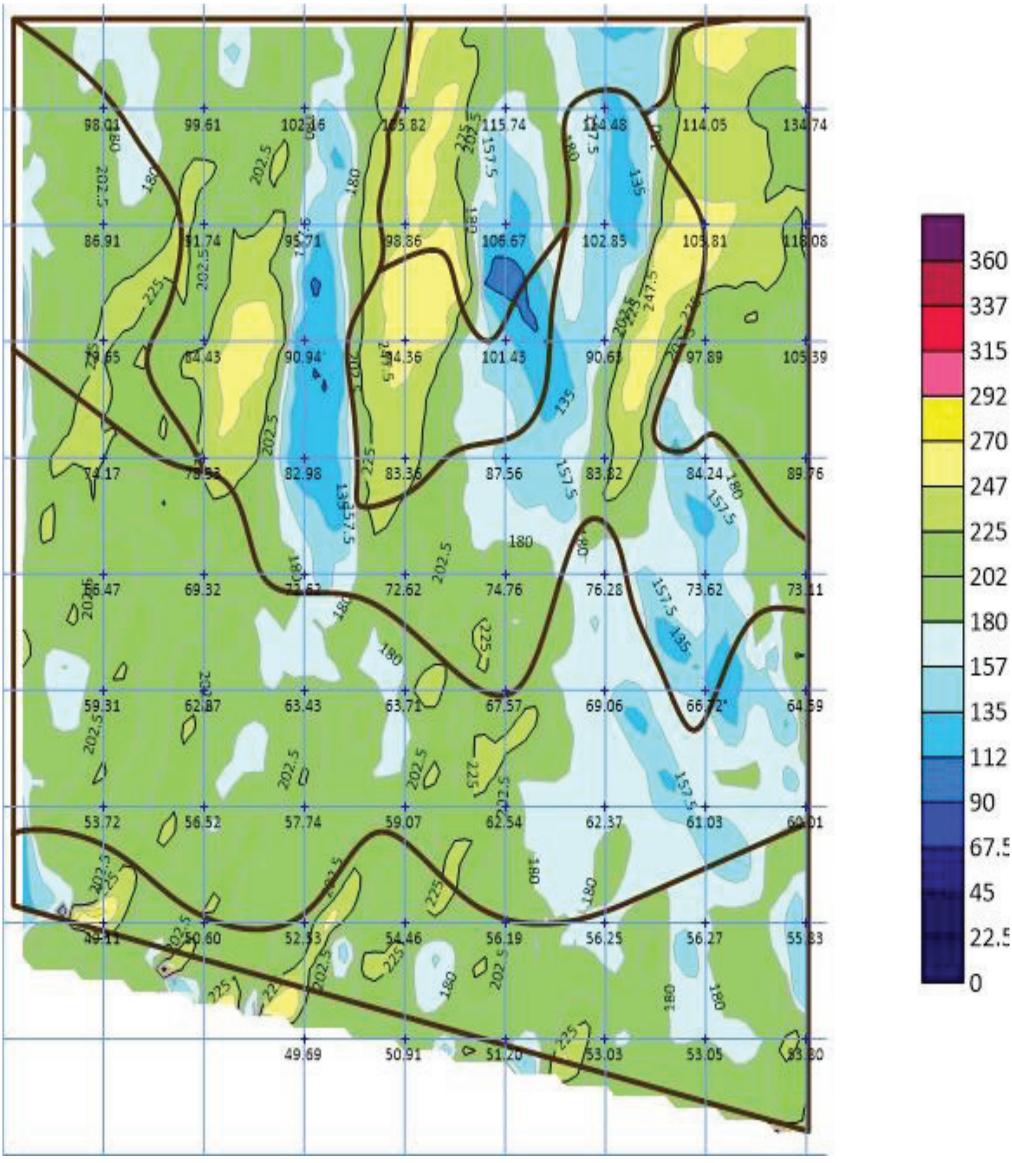

Figure 6. Cartographic of the zones according to the exposure

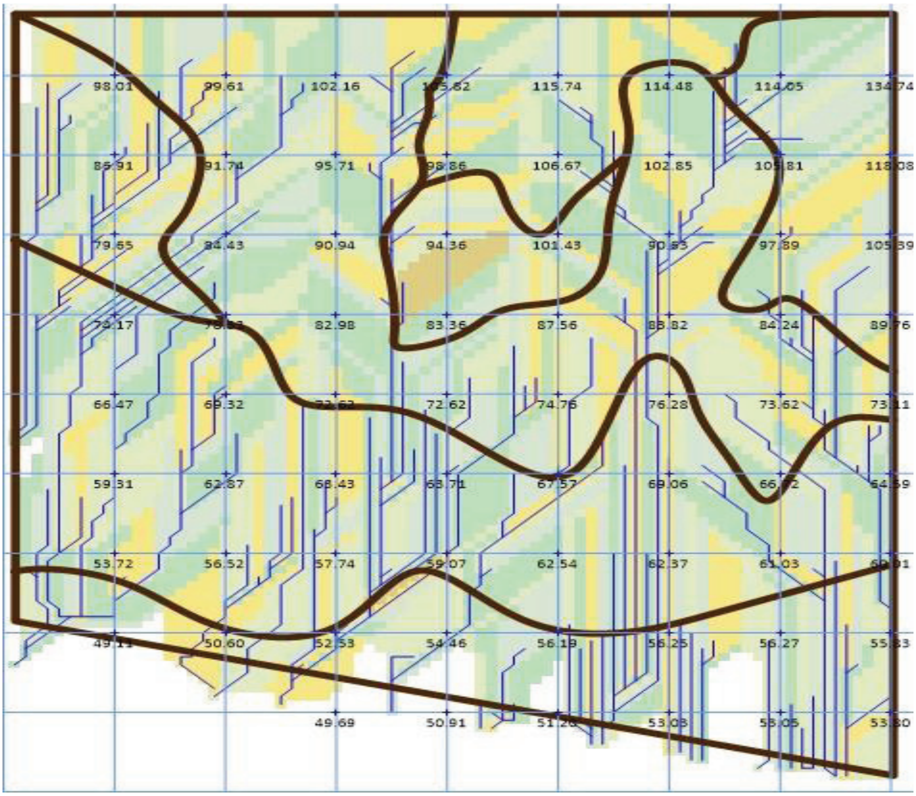

Figure 7. Cartographic of the zones according to the magnitude and the type of the water runoff

Figure 8. Final land regulation solution

CONCLUSIONS

The soil cover was studied, the established soil differences were mapped, and their distribution were compared with the data for the relief and the runoff. As a result, on the terrain were differentiated sections with different degrees of suitability for vineyards growth. In the terrains with limited suitability were differentiated zones requiring a ameliorative effect on one or more of the three components of the landscape - relief, hydrology and soil.

Based on the model that describes the studied terrain, the technological cartographic of the viticultural terroir has been created as follows:

- Determination of the degree of suitability for vineyard growth. Unsuitable terrains were excluded.
- Configuration of the production areas of the vine plantation in the suitable lands.
- Determination of the character and magnitude of the ameliorative restrictions for each of the separated plots.
- Detailed analysis of the soil and melioration conditions in each of the plots. On this basis was created a scheme for the distribution of the vine varieties, the direction and length of the rows, the type of the pad and the fertilization model.

REFERENCES

Dimitrov, S., and Konakchiev, S. (1981). *Erosion solution.* Zemizdat, Sofia.

Gadzhev, R. (2011). Prognosis of soil erosion in streams with variable water quantities. "100 Years of Soil Science in Bulgaria", 2, Sofia.

Ilov, D. (2004). Construction rock mechanics. IR ERA.

Pannikov, V.D. (1983). *Soil erosion and solution*. Zemizdat, Sofia.

Stanev, I.S. (1982). Engineering equipment for erosion solution of the tillable lands. Zemizdat, Sofia.

Totsev, A. (2008). Study of the sustainability on slopes and construction trenches on MKE by decreasing shear strength. 4.

Zaydelman, F.R. (1996). *Soil amelioration*. MGU, Moscow.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

ACCUMULATION AND DISTRIBUTION OF DRY MASS AND NITROGEN IN SORGHUM PLANTS GROWN AT DIFFERENT NUTRITIONAL LEVEL

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Abstract

A pot experiment was carried out to determine the accumulation and distribution of dry mass and nitrogen in sorghum depending on the nutritional level. The plants were grown on eight levels of nutrition – $N_0P_0K_0$, $N_0P_{200}K_{200}$, $N_{600}P_0K_0$, $N_{200}P_{200}K_{200}$, $N_{400}P_{200}K_{200}$, $N_{600}P_{200}K_{200}$, $N_{800}P_{200}K_{200}$, $N_{600}P_{400}K_{400}$. The different levels of nutrients were created by applying of NH_4NO_3, $Ca(H_2PO_4)_2$ and KCl dissolved in water. It was established that plants accumulated higher aboveground dry biomass and nitrogen at $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ levels. The fertilization 600 mg $N.kg^{-1}$ soil increased the grain nitrogen concentration both applied alone and in combination with two levels of phosphorus and potassium $P_{200}K_{200}$ and $P_{400}K_{400}$. The fertilization levels decreased the harvest index of sorghum compared to the control plants. The nitrogen harvest index changed from 47.8% at $N_{800}P_{200}K_{200}$ to 59.7% at $N_{200}P_{200}K_{200}$. Nitrogen harvest index at $N_{800}P_{200}K_{200}$ fertilization had the lowest value 47.8% of all studied levels.

***Key words**: dry mass, nitrogen, accumulation, sorghum.*

INTRODUCTION

Sorghum (*Sorghum bicolor* L. Moench) is one of the five major crops in the world (Shehzad et al., 2009). It can be used as food (grain), feed (grain and biomass), fuel (ethanol production), fiber (paper), fermentation (methane production) and organic by-products (Fernandes et al., 2013). In the European Union, the production is mainly concentrated in France, Italy and Spain and the total consumption of 850,000 tons in Europe exceeds production (Ivanov, 2006). In Bulgaria the production of grain sorghum has increased in recent years and sorghum is one of the top ten grown crops in the country (Bulgarian Ministry of Agriculture and Foods, Agrostatistics, 2016).

Sorghum is a multipurpose crop belonging to the Poaceae family, which are C_4 carbon cycle plants with high photosynthetic efficiency and productivity (Tari et al., 2012). The modern varieties are hybrids with high productivity potential appropriate for application of intensive forage grain production technologies (Kertikov, 2007; Kikindonov et al., 2008).

Nitrogen, phosphorus, potassium and water are considered as the major limiting factors in crop growth, development and finally economic yield (Enchev & Kikindonov, 2015). Proper nitrogen nutrition is critical to meet crop needs and indicate considerable opportunities for improving nitrogen use efficiency (Murrel, 2011). Many agricultural soils have a limited ability to supply available nitrogen for target yields and nitrogen is the most limiting nutrient for cereal crops, including sorghum production (Gerik et al., 2014).

Nutrient uptake of sorghum precedes dry matter accumulation because nutrients are required for growth and dry matter accumulation (Soleymani et al, 2011). The level of mineral nutrition has greater effect on growth and yield of cereals plants (Raun & Johnson, 1999).

The studies of sorghum genotypes in this connection are limited. For better fertilizer management the study about the effects of different levels of nitrogen, phosphorus and potassium on the growth and nutrient uptake of sorghum is very crucial.

The objective of this research was to investigate the effect of different nutritional levels on the accumulation and distribution of dry mass and nitrogen in sorghum plants.

MATERIALS AND METHODS

The effect of different levels of mineral nutrition on the accumulation and distribution of dry mass and nitrogen of sorghum plants was studied in a pot experiment under conditions of growing installation. The experimental design consisted of eight levels of mineral nutrition and four replications of each variant. The studied treatments of soil nutritional levels were: $N_0P_0K_0$, $N_0P_{200}K_{200}$, $N_{200}P_{200}K_{200}$, $N_{400}P_{200}K_{200}$, $N_{600}P_{200}K_{200}$, $N_{800}P_{200}K_{200}$, $N_{600}P_0K_0$ and $N_{600}P_{400}K_{400}$. The levels of nitrogen (N), phosphorus (P_2O_5) and potassium (K_2O) in the soil were created by adding of NH_4NO_3, $Ca(H_2PO_4)_2$ and KCl dissolved in a water.

Three plants per pot of hybrid EC Alize were grown under optimal water regime in plastic pots of 10 L. Each pot contained 15 kg air-dry soil. The main agrochemical characteristics of the soil before sowing of the sorghum were: pH_{H2O} = 7.80 (slightly alkaline reaction); content of mineral nitrogen 27.6 mg $Nmin.kg^{-1}$ soil; content of available phosphorus (Egner-Ream) 158 mg $P_2O_5.kg^{-1}$; and content of exchangeable potassium 210 mg $K_2O.kg^{-1}$.

These results showed low supply of the soil with mineral nitrogen and good soil supply with available phosphorus and potassium. A good water regime was maintained in the pots during the vegetation of the sorghum plants.

The aboveground biomass was collected from the plants under all the treatments in a full maturity phase. The harvested samples were separated in a grain and stover (stems + leaves + chaff). The dry weight of grain and stover was determined after drying for 48 hours at 60°C. The sub-samples of 0.5 g ground and dry plant material were mineralized using a wet digestion by H_2SO_4 and H_2O_2 as a catalyst (Mineev, 2001).

The concentration of nitrogen in plant samples were determined by colorimetric methods and potassium concentration was analyzed by the flame photometer model PFP-7 (Tomov et al, 2009). The content of accumulated nitrogen was obtained by multiplying the dry mass of sorghum grain and stover by the concentration of nitrogen in each plant part.

The differences in the accumulation and distribution of dry mass and nitrogen into sorghum plants among all levels of mineral nutritional were calculated by using the overall analysis of variance (ANOVA). Duncan's Multiple Range Test (Duncan, 1955) at $p < 0.05$ was used in order to determine the difference among the means.

RESULTS AND DISCUSSIONS

The average grain yield of all studied nutritional levels was 56.3 $g.pot^{-1}$ and the obtained average stover yield was 84.0 $g.pot^{-1}$ (Table 1). Unfertilized plants showed the lowest productivity of grain and stover in maturity. The grain and stover yields of sorghum significantly increased at all variants with applied nitrogen in rates from N_{200} to N_{800}, compared to the yield of the controlplants $N_0P_0K_0$. The increase was by 64.0% - 108.6% for the grain yield and by 82.0% - 144.0% for the stover, respectively.

Table 1. Productivity of grain and stover of sorghum plants depending on nutritional level, $g.pot^{-1}$

Nutritional level	Grain	% to $N_0P_0K_0$	Stover	% to $N_0P_0K_0$
$N_0P_0K_0$	35.0 ^{f*}	100.0	45.1 ^{e}	100.0
$N_0P_{200}K_{200}$	39.2 ^{f}	112.0	51.9 ^{e}	115.1
$N_{200}P_{200}K_{200}$	57.4 ^{d}	164.0	82.1 ^{d}	182.0
$N_{400}P_{200}K_{200}$	68.5 ^{bc}	195.7	102.7 ^{b}	227.6
$N_{600}P_{200}K_{200}$	73.0 ^{a}	208.6	110.0 ^{a}	244.0
$N_{800}P_{200}K_{200}$	47.0 ^{e}	134.3	80.1 ^{d}	177.6
$N_{600}P_0K_0$	61.1 ^{c}	174.6	93.1 ^{c}	206.5
$N_{600}P_{400}K_{400}$	69.1 ^{ab}	197.1	107.0 ^{ab}	237.3
Average	*56.3*		*84.0*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

Sorghum plants accumulated the highest quantity of dry above-ground mass when grown at $N_{600}P_{200}K_{200}$ level. The phosphorus-potassium-only fertilization $N_0P_{200}K_{200}$ had a slight effect on the grain and straw yields, and in this variant the sorghum productivity was not significantly different from that of the $N_0P_0K_0$control.The combination of applied phosphorus-potassium $P_{200}K_{200}$ with increasing nitrogen levels from N_{200} to N_{600} resulted in a proven increase of sorghum productivity along with an increase of nitrogen application up to N_{600} level. The exclusion of phosphorus and potassium fertilization and the cultivation of sorghum under N_{600} nitrogen fertilization had a negative effect on the accumulation of dry above-ground biomass at maturity. The productivity of plantsat this nutritional level $N_{600}P_0K_0$ decreased by 11.9 $g.pot^{-1}$ for grain and 16.9 $g.pot^{-1}$ for the stover, compared to the corresponding yields of the triple combination $N_{600}P_{200}K_{200}$ variant. Growing of sorghum at the elevated phosphorus-potassium level $P_{400}K_{400}$ combined with N_{600} did not affect the amount of dry above-ground biomass in maturity. The grain and stover yields of this variant $N_{600}P_{400}K_{400}$ were similar to those obtained with the $N_{600}P_{200}K_{200}$ fertilization.

Table 2. Total productivity of sorghum plants depending on nutritional level, $g.pot^{-1}$

Nutritional level	Grain+Stover	% to $N_0P_0K_0$
$N_0P_0K_0$	80.1 ^{g*}	100.0
$N_0P_{200}K_{200}$	91.1 ^{f}	113.7
$N_{200}P_{200}K_{200}$	139.5 ^{d}	174.2
$N_{400}P_{200}K_{200}$	171.2 ^{b}	213.7
$N_{600}P_{200}K_{200}$	183.0 ^{a}	228.5
$N_{800}P_{200}K_{200}$	127.1 ^{e}	158.7
$N_{600}P_0K_0$	154.2 ^{c}	192.5
$N_{600}P_{400}K_{400}$	176.0 ^{ab}	219.7
Average	*140.3*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

Hybrid EC Alize demonstrated higher productivity of above-ground dry mass in maturity when grown at levels N_{200} - N_{800} (Table 2). The increasing of grain and stover yields of these variants significantly increased by13.7% - 128.5% in comparison with the productivity of unfertilized control plants. Sorghum showed higher total productivity in maturity when grown at $N_{600}P_{200}K_{200}$, $N_{600}P_{400}K_{400}$ and $N_{400}P_{200}K_{200}$ fertilization levels. The high N_{800} level combined with $P_{200}K_{200}$ let to formation of 55.9 g less dry mass per pot, compared to total accumulated above-ground dry mass of plants grown at $N_{600}P_{200}K_{200}$ level.

Mineral nutrition had a significant effect on the percentage of nitrogen of the sorghum grain and stover in maturity (Table 3). The nitrogen concentration of grain was 1.88% N on average, and its values changed in a range 1.65% N - 2.14% N. The sorghum stover contained an average of 1.05% N and its values varied from 0.84% to 1.30% N depending on the fertilization level.

Growing the sorghum without addition of nitrogen ($N_0P_0K_0$ and $N_0P_{200}K_{200}$) resulted in obtaining a grain with the lowest nitrogen concentration of 1.65%-1.69%. Our results corresponded to studies on the effect of nitrogen on the concentration of nitrogen in sorghum grain of other authors (dos Santos et al., 2014). Low nitrogen rate N_{200} in combination with $P_{200}K_{200}$ and as well as application of only phosphorus-potassium fertilization $N_0P_{200}K_{200}$ demonstrated not significant effect on the nitrogen concentration of sorghum stover compared to the control. The nitrogen concentration of the stover of these three variants $N_0P_0K_0$, $N_0P_{200}K_{200}$ and $N_{200}P_{200}K_{200}$ showed lower values of 0.84%-0.90% nitrogen.

Grain nitrogen concentration of the sorghum plants proven increased in parallel with the applied nitrogen in amount of 200, 400, 600 и 800 mg $N.kg\ soil^{-1}$ when the nitrogen level was combined with $P_{200}K_{200}$. The effect of nitrogen fertilization alone $N_{600}P_0K_0$ on the percentage of grain nitrogen was unproven compared to the $N_{600}P_{200}K_{200}$ and $N_{800}P_{200}K_{200}$ variants. The highest grain nitrogen concentration (2.14% N) was established when sorghum grown at 600 mg $N.kg\ soil^{-1}$ combined with a higher $P_{400}K_{400}$ level. In this triple combination, nitrogen concentration of grain exceeded by 29.7% the grain nitrogen concentration of unfertilized plants.

Table 3. Concentration of nitrogen of grain and straw of sorghum plants depending on nutritional level, N %

Nutritional level	Grain, % N	% to $N_0P_0K_0$	Stover, % N	% to $N_0P_0K_0$
$N_0P_0K_0$	1.65 ^{f*}	100.0	0.90 ^{ef}	100.0
$N_0P_{200}K_{200}$	1.69 ^{f}	102.4	0.87 ^{f}	96.9
$N_{200}P_{200}K_{200}$	1.77 ^{e}	107.3	0.84 ^{f}	93.2
$N_{400}P_{200}K_{200}$	1.83 ^{d}	110.9	0.99 ^{de}	110.0
$N_{600}P_{200}K_{200}$	1.97 ^{c}	119.4	1.22 ^{ab}	135.7
$N_{800}P_{200}K_{200}$	2.03 ^{b}	123.0	1.30 ^{a}	144.6
$N_{600}P_0K_0$	1.99 ^{bc}	120.6	1.10 ^{cd}	122.1
$N_{600}P_{400}K_{400}$	2.14 ^{a}	129.7	1.18 ^{bc}	131.4
Average	*1.88*		*1.05*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

The nitrogen N_{400}, N_{600} and N_{800} levels positively affected the concentration of stover nitrogen, with an increase of 10.0%- 44.6%, compared to that of the untreated control (Table 3). The results showed higher nitrogen concentrations of stover (1.30%N) at a high nitrogen $N_{800}P_{200}K_{200}$ variant, but the difference with $N_{600}P_{200}K_{200}$fertilization was not mathematically proven. The doubled

$P_{400}K_{400}$level combined with N_{600} did not significantly change the percentage of stover nitrogen, compared to its valueof sorghum plants grown at $N_{600}P_{200}K_{200}$ level. The stover nitrogen concentration of only nitrogen fertilized plants $N_{600}P_0K_0$ was 1.10% N. The data showed similar values of the N percentage of stover at plants cultivated at $N_{600}P_0K_0$ and $N_{600}P_{400}K_{400}$ levels and no proven effect of $P_{400}K_{400}$ fertilization.

Table 4. Accumulated nitrogen of grain and stover of sorghum depending on nutritional level, g $N.pot^{-1}$

Nutritional level	Grain g $N.pot^{-1}$	% to $N_0P_0K_0$	Stover g $N.pot^{-1}$	% to $N_0P_0K_0$
$N_0P_0K_0$	0.58 ^{e*}	100.0	0.41 ^{e}	100.0
$N_0P_{200}K_{200}$	0.66 ^{e}	114.3	0.45 ^{e}	110.4
$N_{200}P_{200}K_{200}$	1.02 ^{c}	175.1	0.69 ^{d}	167.3
$N_{400}P_{200}K_{200}$	1.25 ^{b}	216.2	1.01 ^{c}	247.1
$N_{600}P_{200}K_{200}$	1.44 ^{a}	247.9	1.34 ^{a}	327.7
$N_{800}P_{200}K_{200}$	0.95 ^{d}	164.5	1.04 ^{c}	253.8
$N_{600}P_0K_0$	1.22 ^{b}	209.6	1.02 ^{c}	248.7
$N_{600}P_{400}K_{400}$	1.48 ^{a}	254.6	1.26 ^{b}	308.2
Average	*1.07*		*0.90*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

The accumulated nitrogen of the above-ground plant parts of sorghum in maturity widely varied depending on the level of mineral nutrition (Table 4). The average nitrogen uptake of sorghum grain was 1.07 g $N.pot^{-1}$ and the average stover nitrogen uptake was 0.90 g $N.pot^{-1}$. Plants with nitrogen fertilization accumulated more grain nitrogen and nitrogen of the vegetative above-ground dry mass, compared to control plants and only phosphorus-potassium fertilized plants. Sorghum accumulated the highest amount of grain nitrogen at N_{600} level combined with phosphorus-potassium $P_{200}K_{200}$ or $P_{400}K_{400}$ levels. The absorbed grain nitrogen of sorghum grown at $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ levels was 1.44 g $N.pot^{-1}$and 1.48 g $N.pot^{-1}$, respectively. These values exceeded the nitrogen taken up of control plants by 147.9% and 154.6%, respectively. Application of nitrogen alone in a rate N_{600} proven reduced the quantity of grain nitrogen in maturity compared to the $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ combinations. It was not established the significant effect of alone nitrogen fertilization $N_{600}P_0K_0$ and triple combination $N_{400}P_{200}K_{200}$ on the accumulated nitrogen of the grain. Sorghum plants grown at $N_{200}P_{200}K_{200}$ and $N_{800}P_{200}K_{200}$ levels distinguished by a lower grain yield and quantity of grain nitrogen among the nitrogen-fertilized variants. Applied mineral nitrogen in rates N_{200}, N_{400}, N_{600} and N_{800} significantly increased the amount of nitrogen accumulated in the sorghum straw. The increase was by 67.3%-208.2% in regard to the non-fertilized control. Plants grown at $N_{200}P_{200}K_{200}$ level accumulated the lowest stover nitrogen among all trails with nitrogen input. Sorghum stover contented more nitrogen (1.34 g $N.pot^{-1}$-1.26 g $N.pot^{-1}$) as a result of $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ fertilization levels. The accumulated nitrogen in the straw had similar values (1.01 g $N.pot^{-1}$-1.04 g $N.pot^{-1}$) when sorghum plants grown at $N_{400}P_{200}K_{200}$, $N_{800}P_{200}K_{200}$, $N_{600}P_0K_0$ levels.

Sorghum accumulated small quantity (0.98 g $N.pot^{-1}$-1.12 g $N.pot^{-1}$)of above-ground nitrogen at variants without added nitrogen (Table 5). Nitrogen fertilization significantly increased the total amount of nitrogen in maturity. The total uptake of nitrogen in above-ground plant parts was higher from 73.6% ($N_{200}P_{200}K_{200}$) to 183.8% ($N_{600}P_{200}K_{200}$) in comparison with unfertilized control plants. The results demonstrated higher accumulation of nitrogen in maturity when plants grown at the triple $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ combinations.

Table 5. Total accumulated nitrogen of sorghum grain + stover depending on nutritional level, g $N.pot^{-1}$

Nutritional level	Total N, g $N.pot^{-1}$	% to $N_0P_0K_0$
$N_0P_0K_0$	0.98 ^{e*}	100.0
$N_0P_{200}K_{200}$	1.12 ^{e}	113.8
$N_{200}P_{200}K_{200}$	1.70 ^{d}	173.6
$N_{400}P_{200}K_{200}$	2.27 ^{b}	231.3
$N_{600}P_{200}K_{200}$	2.78 ^{a}	283.8
$N_{800}P_{200}K_{200}$	2.00 ^{c}	203.6
$N_{600}P_0K_0$	2.24 ^{b}	228.1
$N_{600}P_{400}K_{400}$	2.74 ^{a}	279.6
Average	*1.98*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

The grain of the EU Alize sorghum hybrid represented 40.5% on average of the total above-ground biomass in maturity (Table 6).The portion of grain nitrogen from the total accumulated nitrogen of plants was 55.1%. The harvest index of sorghum plants varied from 47.8% to 59.7% and the nitrogen harvest index changed in range 47.8%-59.7% depending on the level of mineral nutrition. The highest values of harvest index were obtained at the unfertilized control plants and plants grown at lower levels of mineral nutrition. The harvest index of sorghum received elevated N_{600} and N_{800}levels proven decreased by 9.0%-15.5% compared to that at $N_0P_0K_0$ control. No significant differences were found in the harvest index of plants grown at $N_{600}P_{200}K_{200}$, $N_{800}P_{200}K_{200}$, $N_{600}P_0K_0$ and $N_{600}P_{400}K_{400}$ fertilization. The obtained harvest indexes of these levels were in a range 37.0%-39.9%.

Table 6. Harvest index HI and nitrogen harvest index NHI of sorghum depending on nutritional level, %

Nutritional level	HI	% to $N_0P_0K_0$	NHI	% to $N_0P_0K_0$
$N_0P_0K_0$	43.8 [a*]	100.0	58.7 [a]	100.0
$N_0P_{200}K_{200}$	43.0 [a]	98.1	59.4 [a]	101.1
$N_{200}P_{200}K_{200}$	41.2 [a]	94.0	59.7 [a]	101.7
$N_{400}P_{200}K_{200}$	40.1 [ab]	91.5	55.3 [ab]	94.2
$N_{600}P_{200}K_{200}$	39.9 [b]	91.1	51.7 [c]	88.1
$N_{800}P_{200}K_{200}$	37.0 [b]	84.5	47.8 [d]	81.5
$N_{600}P_0K_0$	39.6 [b]	90.5	54.4 [bc]	92.7
$N_{600}P_{400}K_{400}$	39.2 [b]	89.5	53.9 [c]	91.8
Average	*40.5*		*55.1*	

*Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.

The nitrogen harvest index showed the highest values when the sorghum plants grown without nitrogen fertilization and under low N_{200} level. The obtained result of NHI of variants $N_0P_0K_0$, $N_0P_{200}K_{200}$ and $N_{200}P_{200}K_{200}$ were similar 58.7%-59.7%. The differences in the nitrogen harvest index of these variants and the variants with nitrogen application N_{600} were mathematically proven. Our results showed strong negative effect of higher N_{800} level on the proportion of grain nitrogen from total above-ground nitrogen in maturity. The nitrogen harvest index of plants cultivated at $N_{800}P_{200}K_{200}$ was down up to 47.8% and NHI had the lowest value of all studied levels.

CONCLUSIONS

Sorghum hybrid EC Alize showed high response to the soil nitrogen level. The plants accumulated higher above-ground dry biomass and nitrogen at $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ levels. High N_{800} level combined with $P_{200}K_{200}$ significantly decreased the grain and stover yield compared to the productivity of above-ground dry mass of plants grown at lower N_{200}, N_{400} and N_{600} levels. The productivity of plants with applied N_{600}alone decreased grain and stover yields by 16.3% and 15.4%, respectively, compared to the corresponding yields of the triple $N_{600}P_{200}K_{200}$ combination.

Mineral nutrition had a significant effect on the nitrogen concentration of sorghum plants with changes in ranges 1.65% N - 2.14% N and 0.84% N - 1.30% N for grain and stover, respectively. The fertilization 600 mg $N.kg^{-1}$ soil increased the grain nitrogen concentration both applied alone and in combination with $P_{200}K_{200}$ and $P_{400}K_{400}$ levels. The highest grain nitrogen 2.14% was established in sorghum plants grown at 600 mg N.kg $soil^{-1}$ combined with a higher $P_{400}K_{400}$ level.

Sorghum accumulated the most grain nitrogen at N_{600} level combined with $P_{200}K_{200}$ or $P_{400}K_{400}$ levels. Application of N_{600} alone proven reduced the quantity of grain nitrogen in maturity compared to the $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ combinations. Higher accumulation of total nitrogen was established of plants grown at the triple $N_{600}P_{200}K_{200}$ and $N_{600}P_{400}K_{400}$ combinations.

The harvest index varied from 47.8 to 59.7% and the nitrogen harvest index changed in range 47.8 - 59.7% depending on the level of mineral nutrition. The fertilization levels decreased the harvest index of sorghum compared to the control plants. It was proven a strong negative effect of the higher nitrogen level on the nitrogen harvest index of sorghum. Nitrogen harvest index at $N_{800}P_{200}K_{200}$ fertilization had the lowest value 47.8% of all studied levels.

REFERENCES

Bulgarian Ministry of Agriculture and Foods, Agrostatistics. (2016). Retrieved http://www.mzh.government.bg/MZH/bg/ShortLinks/SelskaPolitika/Agrostatistics/Crop/Posts_copy3.aspx

Duncan, D. B. (1955). Multiple range and multiple F test. *Biometrics, 11*. 1–42.

dos Santos, G.R., Gama, F.R., Rodrigues, A.C., Bonifácio, A., Cardon, C.H., Mourão, D.S.C. (2014). Anthracnose severity and yield of grain sorghum in response to increasing nitrogen doses. [Severidade da antracnose e produtividade de sorgo granífero em resposta a doses crescentes de nitrogênio], *Bioscience Journal, 30*(4), 1070–1076.

Duncan, D.B. (1955). Multiple range and multiple F test. *Biometrics*, *11*. 1–42.

Enchev, S. & Kikindonov, G. (2015). Influence of mineral nitrogen and organic fertilization on the productivity of grain sorghum, *Agricultural Science and Technology, 7*(4), 441–443.

Fernandes, E., Pereira, W., Hackenhaar, L., Rodrigues, R. & Terra, R. (2013). The use of whole grain sorghum in broiler feeds. *Revista Brasileira de Ciência Avícola*, *15*(3), 217–222.

Gerik, T., Bean, B., Vanderlip, R. (2014). Sorghum Growth and Development. Texas Cooperative Extension. *Productivity Rotations on farms in Texas, 7*(3), 2–7. http://glasscock.agrilife.org/files/2015/05/Sorghum-Growth-and-Development.pdf.

Ivanov, P. (2006). Tendencies of cereal fodder crop production worldwide and in the European Union. *Field Crops Studies*, *3*(1), 89–98.

Kertikov, T. (2007). Study of productivity capacities for production of forage, crude protein and chemical composition in sorghum hybrids. *Bulgarian Journal of Agricultural Science, 13*(3), 281–289.

Kikindonov, Ts., Slanev, K., Kikindonov, G. (2008). Green mass productivity of sorghum origins. *Journal of Mountain Agriculture on the Balkans*, *11*(3), 503–511.

Mineev, V.G. (2001). *Manual of Agrochemistry*, Academic publisher of Moscow State University, Russia.

Murrell, T.S. (2011). Nutrient Use Efficiency: A Midwest Perspective, Paper presented at the Nutrient Efficiency and Management Conference. Rochester Minesota. International Plant Nutrition Institute, USA, http://mawrc.org/downloads/Nutrient_Use_Efficiency-Midwest_Perspective-Murrell.pdf.

Raun, W.R., and Johnson G.V. (1999). Improving Nitrogen Use Efficiency for Cereal Production. Agron. J. 91:357-363. doi:10.2134/agronj1999.00021962009100030001x

Shehzad, T., Okuizumi, H., Kawase, M. (2009). Genet Resour Crop Evol 56, 809. https://doi.org/10.1007/s10722-008-9403-1

Soleymani, A., Shahrajabian, M.H., Naranjani, L. (2011). The effect of plant density and nitrogen fertilization on yield, yield components and grain protein of grain sorghum. *Journal of Food, Agriculture and Environment, 9*(3-4), 244–246.

Tari, I., Laskay, G., Takács, Z., Poor, P. (2012). Responses of Sorghum to Abiotic Stresses: A Review. *Journal of Agronomy Crop Science*, *199*(4), 264–274.

Tomov, T., Rachovski, G., Kostadinova, S., Manolov, I. (2009). *Handbook of Agrochemistry*, Academic publisher of Agricultural University of Plovdiv, Bulgaria.

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Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

ANALYSIS OF AGROCLIMATIC RESOURCES IN ROMANIA IN THE CURRENT AND FORESEEABLE CLIMATE CHANGE - CONCEPT AND METHODOLOGY OF APPROACHING

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Abstract

Analyzes of the effects of climate change in agriculture is a priority in the strategic development actions in EU member countries and beyond. The interdisciplinary character of actions involves a comprehensive approach by identifying and linking activities to develop and implement measures intra and inter-sectoral those related to climate change in agriculture and related fields. Climate variability affects all sectors but agriculture remains the most vulnerable. Crop production varies year by year, it is significantly influenced by fluctuations in weather conditions and in particular the production of extreme weather events. In Romania, climate change has and will have a significant impact on the development of natural conditions, agriculture and biodiversity are the areas most vulnerable to climate change, given the dependence on climatic conditions and the negative ecological, economic and social. From an economic perspective, this phenomenon causes a reduction to total compromise agricultural production, with serious implications on food security of the population.

***Key words**: agriculture, agrometeorology, climate change, impact, vulnerability.*

INTRODUCTION

The climate is changing globally and in Europe. The Fifth Assessment Report of the Intergovernmental Panel on Climate Change (IPCC, 2013) concluded that the warming since the mid-20th century has predominantly been due to greenhouse gas emissions from human activities, in particular the combustion of fossil fuels, agriculture and other changes in land use.
The Paris Agreement adopted by 195 nations at the 21st Conference of the Parties to the UNFCCC in **December 2015** included the aim of strengthening the global response to the threat of climate change by “holding the increase in the global average temperature to well below 2°C above preindustrial levels and pursuing efforts to limit the temperature increase to 1.5°C above pre-industrial levels“.
There has been a steady increase over the last five years in national adaptation strategies and plans. By September 2016, 20 of EU Member States had adopted a national adaptation strategy and 12 (of which nine are Member States) had developed a **national adaptation plan**. Most progress regarding action plans has been reported for freshwater management, flood risk management, **agriculture** and forestry, with a focus on mainstreaming adaptation in these national sectoral policy areas.
On **8th October 2018**, at INCHEON, Republic of Korea, the IPCC approved *The Special Report (SR15)* where the new limiting global warming to 1.5°C would require rapid, far reaching and unprecedented changes in all aspects of society.
With clear benefits to people and natural ecosystems, limiting global warming to 1.5°C compared to 2°C could go hand in hand with ensuring a more sustainable and equitable society.
The report’s full name is *Global Warming of 1.5°C, an IPCC special report on the impacts of global warming of 1.5°C above pre-industrial levels and related global greenhouse gas emission pathways, in the context of strengthening the global response to the threat of climate change, sustainable development, and efforts to eradicate poverty* (IPCC, October 2018).

What is the IPCC?

The Intergovernmental Panel on Climate Change (IPCC) is the UN body for assessing the science related to climate change. It was established by the United Nations Environment Programme (UN Environment) and the World Meteorological Organization (WMO) in 1988 to provide policymakers with regular scientific assessments concerning climate change, its implications and potential future risks, as well as to put forward adaptation and mitigation strategies. It has 195 member states.

IPCC assessments provide governments, at all levels, with scientific information that they can use to develop climate policies. IPCC assessments are a key input into the international negotiations to tackle climate change. IPCC reports are drafted and reviewed in several stages, thus guaranteeing objectivity and transparency

Study of meteorological parameters of risk with impact on the vegetation and productivity of agricultural crops through different criteria of characterization - definition, main characteristics, agrometeorological risk aspects, as well as identification - spatio-temporal variability, frequency, intensity, succession, production duration, years extremes, extreme events etc., allows to know the vulnerability of agricultural species to the production of meteorological or climatic risk/stress phenomena and to determine the ways of using the agroclimatic potential of the areas of agricultural interest.

The degree of vulnerability of agricultural species to their production as well as of the agricultural territory as a whole is established through different reference thresholds/levels and risk classes in order to assess the agroclimatic degree of agroclimatic favorability of agricultural areas for varieties and agricultural species of varying degrees resistance to their production.

MATERIALS AND METHODS

In the current study, meteorological data and agrometeorological data from different periods/years (1981-2010, 2011-2018) were analyzed and interpreted.

The meteorological parameters (NMA database) used are the maximum and minimum daily temperatures, the average monthly air temperature, wind speed, sunshine duration, relative air humidity, monthly precipitation average, with the aim of highlighting a set of agrometeorological indicators used at European level in the field of agriculture.

The climatic parameters mentioned have been used to establish agrometeorological indicators, such as: scorching heat, precipitation during the agricultural year (September-August), soil moisture in the main agricultural crops.

The “scorching heat” phenomenon is characterized by the evolution in the dynamics of the maximum daily air temperatures exceeding the limiting biological parquets of the plants, expressed in the amount of the "heat units”, as well as in the intensity of the phenomenon during the active vegetation season (April-September) or characteristic intervals for the main field crops.

Precipitation is an agrometeorological reference indicator for an agricultural area, against which extreme years can be reported, considered agroclimatic risk. This value expresses the potential of rainfall resources useful in determining the degree of pluviometric favorability of an agricultural area for a species, namely variety or hybrid. In this way, a clear picture of the possibilities of expanding into the culture of those genotypes/varieties or hybrids, with economically efficient production on the surface unit, is obtained.

The soil moisture reserve (mc/ha) was analyzed on different profiles and calendar dates specific to field crops, correlated with the periods with maximum water requirements in summer.

In this analyze GIS techniques were used to graphically represent the dynamics of agro-meteorological parameters: scorching heat, rainfall and soil moisture.

RESULTS AND DISCUSSIONS

Almost all European countries have conducted national climate change vulnerability and risk assessments (Figure 1). In this context, the Southern Europe is more vulnerable to climate change several sectors will be affected such as: agriculture, forestry, energy, infrastructure, human health etc. Climate projections (JRC, Russo et al., 2014) show a marked increase in high temperature extremes, meteorological droughts, and heavy precipitation events with variations across Europe.

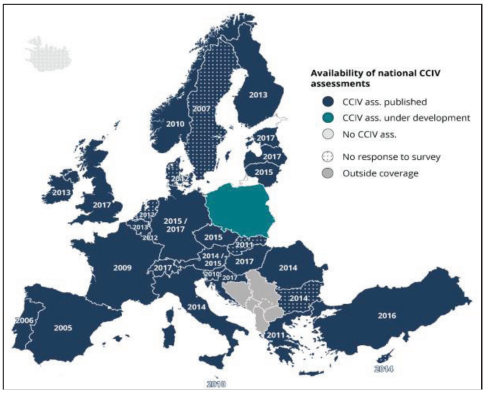

Figure 1. National climate change vulnerability and risk assessments in Europe (Source: EEA, 2018)

These scenarios show us that into the period 2020-2052, the number of projected extreme heatwaves will be highest in southern Europe with 3-6 extreme heatwaves over 33 years (i.e. one every 5-10 years) and for the future interval 2068-2100, will we have 12-15 extreme heat waves over 33 years (i.e. one every 2-3 years) in some parts of Southern Europe (Figure 2).

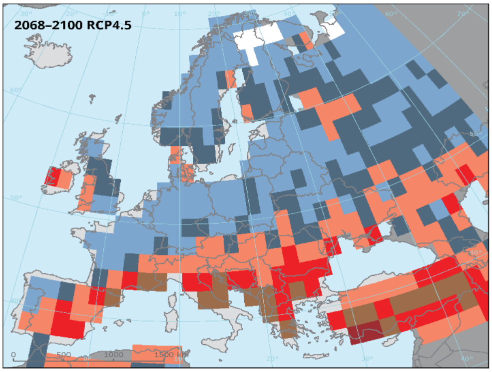

Figure 2. Heat Wave Magnitude Index (HWMI) is based on the magnitude and length of heat waves in a year, where heat waves are periods of at least 3 consecutive days with maximum temperature above the threshold for the reference period 1981-2010 (Source: JRC, Russo et al., 2014)

In this context, Romania is already facing the current environmental stresses, including increasing vulnerability in the intensity and frequency of climatic extremes (drought, floods, heat, frost, diseases and pests etc.), which cause significant losses in all economic sectors , but especially in agriculture, which is the most time-dependent sector. Every physical, chemical and biological process that determines the growth and development of agricultural crops is regulated by specific climatic requirements and any deviation from these requirements can result in great variability in the level of agricultural output, and implicitly, major negative consequences on food security. Agricultural production will be affected by predictable variability and climate change, especially in high-risk farming areas with low potential for adaptation.

In Romania, the changes in the climate regime are in the global context, but with the particularities of the geographical region in which our country is located. Thus, at the level of the period 1901-2018, the analysis of the average annual air temperature values from a number of 29 meteorological stations with a consecutive series of observations over 100 years (Figure 3) shows that the average annual temperature increased by 0.7°C in the period 2000-2018 (11.7°C) over the whole analyzed period (9.3°C).

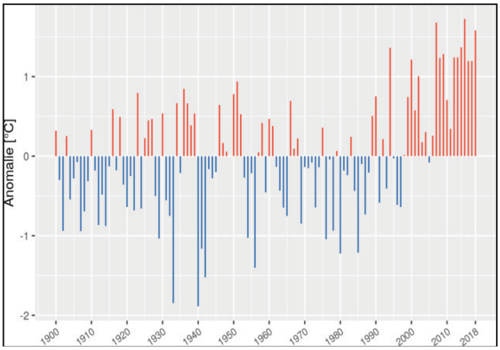

Figure 3. Deviations of the annual average temperature in Romania, 1901-2018 (NMA, 2018)

In the last 30 years, the warmest year was 2015 (11.7°C) and the coldest 1985 (8.4°C) (Table 1). Also, in Romania, the effects climate change has had and will have a significant impact on the evolution of natural conditions, agriculture and biodiversity are the areas most vulnerable to climate change, given dependend climatic conditions and the negative ecological, economic and social.

Table 1. The warmest years in Romania

No	Year	Mean annual air temperature (°C)	Deviation
1	2015	11.72	1.7371
2	2007	11.67	1.6916
3	**2018**	**11.57**	**1.5805**
4	2014	11.36	1.3808
5	1994	11.35	1.3748
6	2009	11.28	1.2972
7	2013	11.23	1.2546
8	2012	11.23	1.2538
9	2008	11.23	1.2466
10	2000	11.21	1.2270
11	2016	11.19	1.2095
12	2017	11.19	1.2094
13	2002	11.00	1.0175

These extreme weather events cause a significant economic loss in agriculture, transport, energy supply, water management, etc., and global climate models indicated that the frequency and intensity of these events can only be expected to increase.

Thus, may cause drought and reduce the potential for degradation of biological farmland soil. From an economic perspective, this phenomenon causes the decrease to total compromise agricultural production, with significant implications on food security of the population. In social, drought condition generates poverty, especially among the rural population, mainly dependent on agricultural activities.

Drought is a major natural hazard having different definitions depending on the type of impact or socio-economic activity which is affected. From the meteorological point of view, a drought period is defined by a significant deficit in the precipitation regime. Pedological drought refers to a significant deficit in the soil moisture. For agriculture, drought is defined by parameters affecting crops growth and production.

In Romania, approximately 14.7 million ha of agricultural land (of which 9.4 million hectares of arable land), soils are affected by drought for long periods and in consecutive years on an area of approx. 7 million hectares of arable land (48%) or excess moisture in the rainy years (about 4 million hectares). Drought is the limiting factor affecting crop production on the largest surface extension and intensity of this type of risk reduction causing fluid annual agricultural production by at least 30-50%.

In Romania, drought-affected areas have expanded over the past decades. The most affected areas are in the South and South-East of Romania, but the entire country has felt the effects of extensive pedological drought, especially in the last 30 years.

Since 1961 until now, Romania has seen in every decade one to four extremely droughty/rainy years, an increasing number of droughts being more and more apparent after 1981 (Table 2).

Table 2. Droughty/rainy years in Romania (1961-2020)

DECADE	XX-TH CENTURY	
	EXTREMELY DROUGHTY YEARS	EXTREMELY RAINY YEARS
1961-1970	1962-1963, 1964-1965	1969, 1970
1971-1980	1973-1974, 1975-1976	1972, 1974, 1975, 1976
1981-1990	1982-1983, 1985-1986, 1987-1988, 1989-1990	1981, 1990
1991-2000	1992-1993, 1999-2000	1991, 1997
	XXI-TH CENTURY	
2001-2010	2000-2001, 2001-2002, 2002-2003, 2006-2007, 2008-2009	2005, 2006, 2008, 2010
2011-2020	2011-2012, 2014-2015, 2015-2016, 2016-2017	2013

In the 21-th Century, 2006-2007, 2011-2012 and 2014-2015 were included in the list of the most droughty years, both through the intensity of the in-soil water deficits and through the duration of the scanty intervals and wideness of the surfaces affected by pedological drought (extreme, severe and moderate) in almost all areas of the country.

The analysis of the average monthly air temperature values in the 2006-2007 agricultural year shows that in 11 of the 12 months (February, March, April, May, June, August, September, October, November, December), average temperatures - above the climatological norms, the maximum deviation from the reference period was recorded in September, ie +3.8°C (Figure 4).

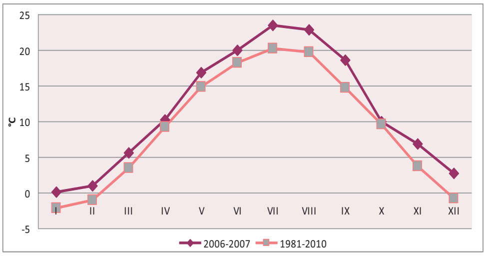

Figure 4. Monthly average air temperature variation in the agricultural year 2006-2007 as compared to multiannual averages (1981-2010)

In the agricultural year 2011-2012, the average monthly air temperature shows that only 3 of the 12 months, January, February and December respectively, show negative deviations from the reference period, the negative deviation being -5.4°C (Figure 5).

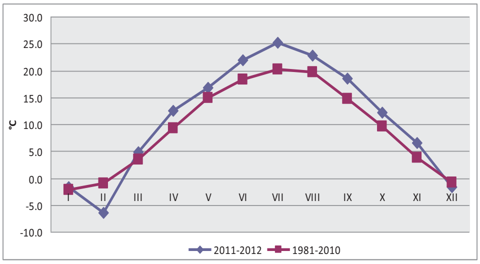

Figure 5. Monthly average air temperature variation in the agricultural year 2011-2012 as compared to multiannual averages (1981-2010)

The analysis of the average monthly air temperature values in the 2014-2015 agricultural year shows that during the longest part of the period the average temperatures were above the climatological norms, the maximum deviation from the reference period being recorded in July, respectively +3.8°C (Figure 6).

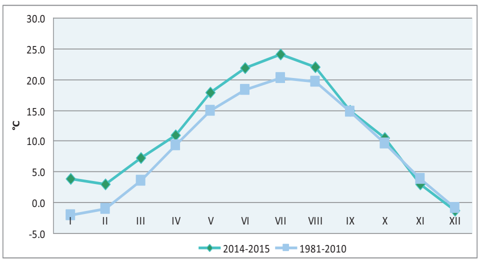

Figure 6. Monthly average air temperature variation in the agricultural year 2014-2015 as compared to multiannual averages (1981-2010)

In Romania, the NMA is covering the Agro-meteorology domain and processes the meteorological information as well as analyzes how weather conditions influence the processes of growing and developing crops/vines. The Department of Agrometeorology studying the agrometeorological parameters (humidity reserve, potential ETP-evapotranspiration, minimum, average and maximum air temperature, soil temperature, sunshine duration, wind speed, relative humidity, winter severity-, heat-units of heat/number of days, imprinting index) and establishes how to highlight and develop cultures through the development phases of both crops and fruit and wine species.

The ***scorching heat*** phenomenon is one of the most important agrometeorological risk factors that can have negative effects on plant growth and development processes. The evolution of scorching heat intensity in Romania from 1961 to 2018 shows an increasing trend especially after 1981 (Figure 7).

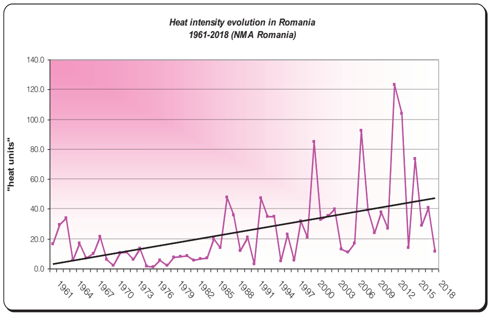

Figure 7. The evolution of scorching heat intensity in Romania, 1961-2018 (NMA Agrometeorology)

Given the multi-annual means of scorching heat intensity, phenomenon quantified by sums of air temperature highs equal to or above 32°C recorded during the summer months, it has become apparent a significantly higher thermal stress over the critical interval for crops (June-August), an increase from 13 units of scorching heat between 1961 and 1990 to 28 units over 1981-2010 (Figure 8).

In 2007, 2012 and 2015 the highest values of the heat scorching of the year were recorded on the agrometeorological stations: Giurgiu (2007-222.5 units, 2012-296.2 units) and Calafat (2015-182.0 units) (Table 3).

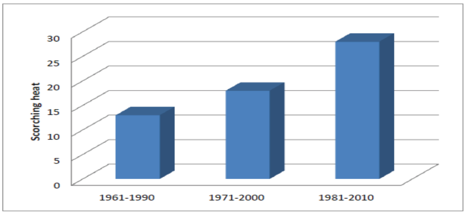

Figure 8. Intensity of scorching heat intensity in Romania, by 30 years interval, 1961-2010

Table 3. Maximum heat units in the most droughty years of the XXI Century in Romania

Year	Station	Heat units
2001	Băilești	127.0
2002	Băilești	102.9
2003	Calafat	141.8
2004	Calafat	70.1
2005	Calafat	38.0
2006	Turnu Măgurele	65.2
2007	**Giurgiu**	**222.5**
2008	Bechet	127.4
2009	Giurgiu	84.2
2010	Calafat	107.6
2011	Calafat	105.9
2012	**Giurgiu**	**296.2**
2013	Calafat	142.0
2014	Turnu Măgurele	53.9
2015	**Calafat**	**182.4**

Precipitation is the main source of water for the growth and development of agricultural crops, and the most significant elements of this meteorological parameter are the quantitative variability, the distribution and the spatio-temporal distribution.

Knowing the periods of deficiency/surplus in rainfall for each zone/agricultural region allows the choice of the crops/varieties that are most suitable for the area of interest, as well as the establishment of the pedoameliorative, agrofitotechnical and economic-organizational protection measures necessary for the production process in the agricultural field.

The precipitation regime, the 2011-2012 agricultural year was excessively droughty at the scale of the whole country's agricultural territory, with precipitation deficits even more severe than in the landmark droughty year 2006-2007.

The Figure 9 exemplifies the precipitation amounts' territorial distribution in the 2006-2007, 2011-2012 and 2015-2015 agricultural years, comparatively with 1981-2010.

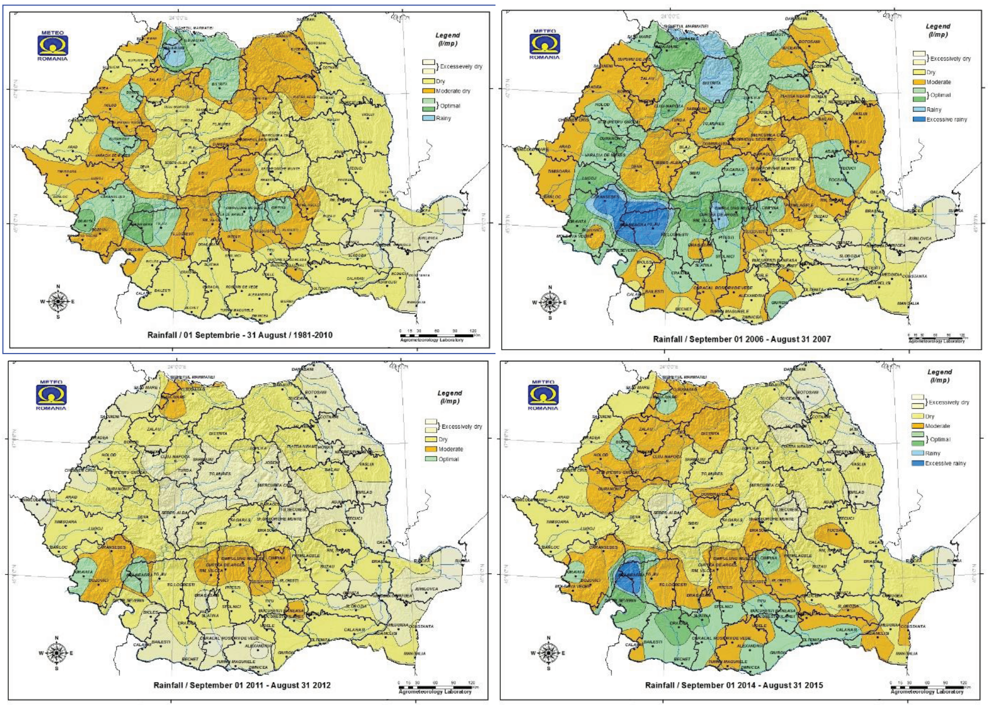

Figure 9. Rainfall amount in 2006-2007, 2011-2012 and 2014-2015 agricultural years compared with 1981-2010

Soil water reserve in soil layer 0-100 cm in non-irrigated corn crop on August 31, 2007, low values (moderate pedological drought) and particularly low (strong and extreme soil drought) on extended areas in the West, South, South-East and East of the country. Local to the West, center, Northeast and isolated in the East of the country, water supply to the soil ranged within satisfactory and close to optimal limits.

At the end of August 2012, in the non-irrigated corn crop, the water supply of the soil at the depth of the soil 0-100 cm present low and particularly low values, the pedological drought being moderate, strong and extreme on almost the entire agricultural territory of the country, with the exception of some areas in the Northwest of Moldova, where the supply of water to the soil was within satisfactory limits.

The moisture content in the 0-100 cm soil layer in non-irrigated maize culture on August 31, 2015 was low (moderate pedological drought) and particularly low (strong and extreme soil drought) for the most part cultural areas. On the agricultural areas in central and Western Transylvania, the center of Oltenia, isolated in the West, South and East of Muntenia, Southeastern Dobrogea and Northern Moldova, the water supply of the soil was within satisfactory and isolated limits close to the optimum (Figure 10).

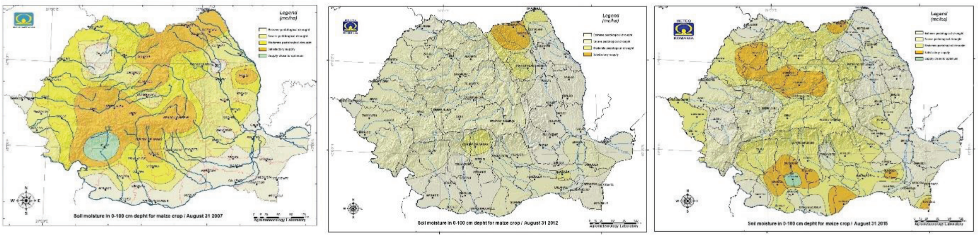

Figure 10. Moisture reserve in the soil layer 0-100 cm in corn crops on 31 August 2007, 2012 and 2015

Due to the maintenance of the water stress associated with the thermal in the air and the long-term soil, the non-irrigated pruning crops (corn and sunflower) maturation processes were hampered over all three analyzed years, with sub-dimensioning of the element elements small diameter head and dry sunflower seeds (Figure 11), as well as small sized oysters and shrimps on corn.

Figure 11. The state of vegetation (2007-2012) in Călărași/Muntenia and Adamclisi/Dobrogea in the sunflower and corn crops (NMA Agrometeorology Network)

Also, undersized and dehydrated fruits and their premature fall were reported in the tree species.

The effects of the phenomenon of pedological drought on the growth and development of agricultural plants are cumulative (Figure 12), depending on the intensity and duration, the consequences being accentuated when the action of thermal and water stress in the air and soil is combined (complex agricultural drought) and long lasting (>10-15 days consecutive, particularly in critical crop periods).

Figure 12. The state of vegetation-summer 2015 in Iasi/ Moldova in the sunflower and corn crops (NMA Agrometeorology Network)

The agricultural year 2006-2007 marked as the most droughty for the agriculture sector, with peak intensity especially in the south and south-east of the country, where the harvest was totally compromised. But, the 2011-2012 agricultural year can be considered excessively droughty, both through the intensity of the in-soil water deficits and through the duration of the scanty intervals and wideness of the surfaces affected by pedological drought (extreme, severe and moderate) in the almost all areas of the country.

The agroclimatic potential of a cropping area provides information on assessing the natural conditions of vegetation and the vulnerability of agricultural areas to the production of risk/stress phenomena that can cause significant annual deviations in terms of agricultural potential and economic development. The analysis of thermal and hydric resources involves identifying critical parameters and thresholds at specific time intervals that correspond to the process of growth and development of the main agricultural crops during plant vegetation, from sowing up to maximum water consumption period (June to August). Through continuous monitoring and surveillance of risk/stress phenomena, the most effective measures for prevention and mitigation regarding the effects on the obtained agricultural productions can be adopted.

CONCLUSIONS

Crop production varies year by year, being heavily influenced by fluctuations in climatic conditions and, in particular, by extreme meteorological events.

Climate variability influences all sectors of the economy, but agriculture remains the most vulnerable, and its impact is more acute at present, because climate change and variability is becoming more and more pronounced.

In Romania, the effect of climate change is felt, and these will be manifested by increasing temperatures, changing the rainfall regime, melting ice and snow, and raising sea levels.

Extreme weather phenomena leading to negative environmental impacts (floods and droughts) will become more frequent and intense in many regions. Effects on ecosystems, economic sectors, population health, and vulnerability vary from region to region.

Climate change is inevitable, which is why complementary action to adapt to its effects is needed.

The vulnerability degree of the cultivated species to the materialization of the thermal and hydric stress respectively is established on the grounds of the specific reference limits/hazard levels and classes, so as to assess the agroclimatic favourability degree of the agricultural surfaces for agricultural sorts and species with a different resilience to the occurrence of those hazards.

Thus, the analysis of the agroclimatic phenomena implying thermal and hydric stress involves identifying the critical parameters and thresholds over specific calendar intervals, corresponding to the undergoing of the growth and development processes in plants, as well as over the whole vegetation period, so that the favourability degree for agriculture from the agrometeorological standpoint is also established.

The agriculture will face more climate-related risks, the adaptation options being requiring continuing researches on the effect from irrigation and sustainability of yields under various water saving methods and irrigation technologies.

Policymakers need good information at different scales (local, regional, European, global) to identify priority issues, the most appropriate adaptation measures and economic activities.

REFERENCES

Bălteanu, D., Şerban, M. (2005). *Modificări globale ale mediului*. Editura Credis, Bucureşti.

Bîlteanu, Gh. (1998). *Fitotehnie*. Ediţia a doua. Editura Ceres, Bucureşti.

Berbecel, O. şi colab. (1970). *Agrometeorologia*. Editura Ceres, Bucureşti.

Mateescu, E., Povară, R., Oprişescu, R. (1999). Studiul parametrilor agrometeorologici de stres şi impactul acestora asupra grâului de toamnă în perioada înspicare - înflorire - umplerea bobului. *Lucrări Ştiinţifice USAMV Bucureşti, Seria A, XLII*, 133–149.

Povară, R., Mateescu, E., Oprişescu, R. (2003). *Estimarea influenţei deficitului de precipitaţii din lunile de toamnă asupra stării de vegetaţie şi a recoltei de grâu în judeţul Vâlcea, Geografia Judeţului Vâlcea - Teorie şi practică*. Vol. III, Editura Offsetcolor, Râmnicu Vâlcea, ISBN 973-86352-7-6, 90–98.

Russo et al. (2014). Magnitude of extreme heat waves in present climate and their projection in a warming world. *Journal of Geophysical Research Atmospheres, 19*(22), 12500–12512.

Sandu, I., Mateescu, E., Vătămanu, V. (2010). *Schimbările Climatice în România şi efectele asupra agriculturii*. Editura Sitech, Craiova.

***EEA information on climate change adaptation and disaster and risk reduction, Third Annual Scientific Seminar, Disaster Risk Management, Knowledge Centre, Bulgarian Academy of Sciences, Sofia, 26-27 April 2018.

***European Environment Agency https://www.eea.europa.eu/themes/agriculture.

***Eurostat, http://ec.europa.eu/eurostat/web/agriculture/data/database.

***Intergovernmental panel on climate change (IPCC) http://www.ipcc.ch/.

***IPCC Press Release (2018). Summary for Policymakers of IPCC Special Report on Global Warming of 1.5ºC approved by governments.

***National Strategy on Climate Change and Low-Carbon Economic Growth, Project co-financed from the European Regional Development Fund under the Operational Program Technical Assistance 2007-2013.

***National Research, Development and Innovation Strategy (2007-2013) http://old.uefiscdi.ro/userfiles/file/ROST/strategia20eng.pdf.

***National Action Plan on Climate Change 2016-2020 http://www.mmediu.gov.ro/categorie/planul-national-de-actiune-privind-schimbarile-climatice-2016-2020/122.

***Romanian Statistical Yearbook, National Institute of Statistics (2016) http://www.insse.ro/cms/sites/default/files/field/publicatii/anuar_statistic_al_romaniei_2016_format_carte.pdf.

***The National Research - Development and Innovation Plan III for 2015 -2020, approved by the Government, http://gov.ro/en/government/cabinet-meeting/the-national-research-development-and-innovation-plan-iii-for-2015-2020-approved-by-the-government.

***United Nations Framework Convention on Climate Change http://newsroom.unfccc.int/.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE MITODEPRESIVE AND GENOTOXIC EFFECT OF SOME FOOD COLORANTS ON THE MERISTEMATIC CELLS TO *Allium cepa*

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Abstract

Currently, most processed foods purchased from supermarkets contain one or more food colorants. One of the best-known colorants is the so-called red dye for eggs, used during the Easter religious holiday. The purpose of this paper was to highlight the mitodepresive and genotoxic effect of an assortment of red dye for eggs to meristematic cells of onion (Allium cepa). Three different concentrations (0.5, 1 and 1.5%) were used, the exposure time being 3 hours. The results obtained revealed a strong mitodepresive effect of food colorant to A. cepa, by decreasing of the mitotic index as the concentration of colorant increased. Also, a genotoxic effect has been observed by the occurrence of some chromosomal aberrations and nuclear alterations, such as sticky and vagrants chromosomes, multinucleated cells and cells with nuclear erosion. These results suggest prudence in using the red dye for eggs and finding other alternative and ecological solutions such as, for example, the use of some plant extracts for this purpose.

Key words: *Allium cepa, red dye, mitodepresive, genotoxic, meristematic cells.*

INTRODUCTION

Food colouring is used both in commercial food production and in domestic cooking. Either the synthetic food colours or natural food colours, the colour has always had an important implication on the minds of people as far as food is concerned. Cuisines prepared in attractive colours have immensely lured men folk in all the quarters of the world. It is therefore necessary either to preserve the natural or maintain the characteristic colour of a food product while it is manufactured or stored for future use. A non-attractive colour however makes the food look un- fresh and is likely to be rejected.

According to FDA, a food colorant is “any dye, pigment or substance which when added or applied to a food, drug or cosmetic, or to the human body, is capable (alone or through reactions with other substances) of imparting colour” (FDA, 2016). Synthetic food colorants were largely used, but have been progressively substituted by those obtained from natural origins. Despite natural pigments offer a strong advantage when compared to the synthetic ones, its safety and whole effects needs to be assessed, in order to conclusively demonstrate health improving effects (Martins et al., 2016). Numerous side effects and toxicity, at both medium and long-terms, allergic reactions, behavioural and neurocognitive effects have been related with their use. However, several food additives that were used over decades are no longer currently allowed, due to the real evidences of their side effects, toxicity at medium and long-terms and high frequency of health disturbance incidents.

At present, consumers are becoming more concerned about their health, meaning, firstly, adequate, high quality nutrition (Abebe et al., 2016; Ouis & Hariri, 2018; Righi et al., 2018).

Dyeing eggs for the Easter, a habit which we learned from parents and grandparents and we will pass on from generation to generation, may become dangerous under certain conditions. Since the commercial dye for eggs is obtained industrially, from artificial dyes in general, we considered this cytogenetic study to be appropriate for evaluating the cellular activity of meristematic plant tissues under action of the red dye (one of the most used), using the *A. cepa* species (onion) as the test plant. *A. cepa* has assayed to be best model plant for standard use in environmental monitoring and cytological analysis (Bonciu et al., 2018; Bonciu, 2018).

MATERIALS AND METHODS

The biological material used was represented by commercially onion bulbs, which were immersed in glasses with water for 72 hours, time required for the meristematic roots occurrence. Prior to initiating the test, the dry bottom plate of the bulbs was removed without destroying the root primordial. When the meristematic roots reached the length of 15-20 mm, they were immersed in dilutions of various concentrations of the red dye for eggs (0.5, 1.0 and 1.5%) for 3 hours, at room temperature. The red dye was purchased from a supermarket and the following ingredients were listed on the label: softened water, azorubine (E 122), glazing agent (glycerine and glucose), acidifier (citric acid E 330 and lactic acid E 270), preservative (sodium benzoate and potassium sorbate), and thickening agent (xanthan gum). A number of 5 onion bulbs were used for each treatment variant as well as an untreated control that was immersed in tap water.

The roots were processed according to the protocol of fixation, hydrolysis and staining to highlight the cytological activity and eventual presence of chromosomal aberrations.

In order to highlight chromosomes and chromosomal aberrations was used the Feulgen-Rossenbeck method and Schiff reagent. Feulgen stain is a staining technique discovered by Robert Feulgen (1924). Depend on acid hydrolysis therefore fixating agents using strong acids should be avoided. The necessity for hydrolysis of tissues with acid in order to obtain the specific reaction with Schiff reagent was demonstrated by Feulgen and Rossenbeck in their original description of the Feulgen stain in 1924. Schiff reagent is prepared by pouring 200 ml of boiling distilled water over 1-g basic fuchsin. Shake thoroughly, cool to 50°C, filter, and add 30 ml 1N HCl to the filtrate. Cool to room temperature and add 1 g potassium metabisulfite Allow the solution to stand overnight in the dark or until a light straw or faint pink colour develops.

The microscopic preparations were performed according to the squash method.

Statistical analysis was done using MS Excel 2007. The analysis of variance (ANOVA) was used to assess the significant differences between the control variant and each treatment. The differences between treatment means were compared using the LSD-test at a probability level of 0.05% subsequent to the ANOVA analysis.

The mitotic index was calculated using the following formula:

$$\text{MI (\%)} = \frac{\text{Total number of cells in division}}{\text{Total number of analysed cells}} \times 100$$

The index of the total abnormalities (TA) was also calculated:

$$\text{TA (\%)} = \frac{\text{Total number of aberrant cells}}{\text{Total number of cells in division}} \times 100$$

Photomicrographs of cells showing chromosomal aberrations and nuclear alterations as well as showing mitosis were taken using the Kruss microscope.

RESULTS AND DISCUSSIONS

The results are illustrated in Table 1. The cytotoxicity level can be determined by the decreased rate of mitotic index. It was found that red dye induced a strong mitodepresive effect in meristematic cells to *A. cepa*. The mitodepresive effect was enhanced as the concentration of red dye increased. Thus, compared to the Control variant, the mitotic index recorded a decrease of over 50%, from 36.42% (V1-Control) to 18.04% (V2), 1.0% (V3) and 9.03% (V4) in all treated variants.

Table 1. Effect of different concentrations of the commercial red dye on the cytological parameters to *A. cepa*

Variants/ Conc. (%)	MI ± SE %	Cells abnormalities frequency (%)				TA (%)
		S	V	MN	NE	
V1 (Control)	36.42±0.68	0	0	1.32	0	1.32
V2/0.5	18.04±0.42*	4.18	2.62	4.21	1.58	12.59*
V3/1.0	12.18±0.39*	6.45	4.01	5.98	2.08	18.52*
V4/1.5	9.03±0.34**	8.34	5.84	11.07	2.64	27.89**

MI = Mitotic index; SE = Standard error; S = Stickiness; V = Vagrants; MN = Multinucleated cells; NE = Cells with nuclear erosion; TA = Total abnormalities; *Significant at level 5% (p=0.05)

The decrease in the mitotic index was positively correlated with increasing concentration of the red dye solutions. According to Panda and Sahu (1985), a decrease of mitotic index below 50% usually has lethal effects. If mitotic index decreases below 22% of control, that it causes sub-lethal effects on test organism (Antonsie-Wiez, 1990). In our study, the mitodepressive effect of red dye may be due to its inhibitory effect on the mitotic cycle during interphase and delaying of spindle formation.

Synthetic colours are a major source of food intoxication and many surveys have been conducted to determine the presence of no permitted food colours in different food products (Vazhangat P. & Thoppil J.E., 2016). In the case of kids' foods, shines colours are also added to attract their attention and make the foods appear attractive and fun. But in most cases, if a food comes in a colour that is not found in nature, excessive consumption can cause health problems.

Food dyes are one of the most widely used and dangerous additives. While the European Union has recently placed regulations on labelling food dyes to inform consumers of the health risks, the United States has no such requirement. Every year, food manufacturers pour 15 million pounds of artificial food dyes into U.S. foods, according to the Center for Science in the Public Interest (CSPI). In the "Food Dyes: A Rainbow of Risks" report, CSPI revealed that nine of the food dyes currently approved for use in the United States are linked to health issues ranging from cancer and hyperactivity to allergy-like reactions and these results were from studies conducted by the chemical industry itself. For instance, Red 40, which is the most widely used dye, may accelerate the appearance of immune system tumours in mice. Almost all the toxicological studies on dyes were commissioned, conducted, and analysed by the chemical industry and academic consultants. Ideally, dyes (and other regulated chemicals) would be tested by independent researchers.

Several studies have been oriented to demonstrate the antimitotic and genotoxic activities of some food additives and pointed out their danger as carcinogens or mutagens. Many authors reported that the mitotic index of *A. cepa* root tips was successively decreased with the increase in different dye concentrations and duration of treatments (Vazhangat P. & Thoppil J.E., 2016).

In our study, all treatments with red dye resulted in a significant increase of the percentage of chromosomal aberrations and nuclear alterations (Figure 1).

Thus, the index of the total abnormalities (TA) recorded an increase in all treated variants, from 1.32% (Control) to 12.59% (V2), 18.52% (V3) and 27.89% (V4). The increase in the TA index was positively correlated with increasing concentration of the red dye solutions.

Multinucleated cells and stickiness were the dominant abnormality induced after treatment, especially at higher concentrations. Stickiness is an irreversible chromosomal aberration and reflects high toxicity of tested solutions.

Frequency of multinucleated cells recorded values between 1.32% (Control), 4.21% (V2), 5.98% (V3) and 11.07% (V4). Also, the frequency of cells with stickiness abnormalities was 4.18% (V2), 6.45% (V3) and 8.34% (V4). Regarding the cells with chromosomal aberrations type vagrant, their frequency ranged between 2.62% (V2) and 5.84% (V4). The frequency of nuclear erosion abnormalities was 1.58% (V2), 2.08% (V3) and 2.64% (V4) (Figure 2). In Figure 3 are shown some cytogenetic abnormalities identified in meristematic roots of *A. cepa* exposed to commercial red dye.

The results obtained indicated that red dye induced a strong mitodepresive and genotoxic effect in meristematic cells to *A. cepa*, by reduction of the mitotic index and occurrence a several cytological abnormalities.

These results suggest prudence in using the chemical red dye for eggs painting. Alternatively, natural extracts from various plants can be used (red beet juice, pomegranate juice, red onion peels, red peony petals etc.).

Natural food colours are preparations obtained from foods and other edible natural source materials obtained by physical and/or chemical extraction resulting in a selective extraction of the pigments relative to the nutritive or aromatic constituents. They come in many forms consisting of liquids, powders, gels and pastes.

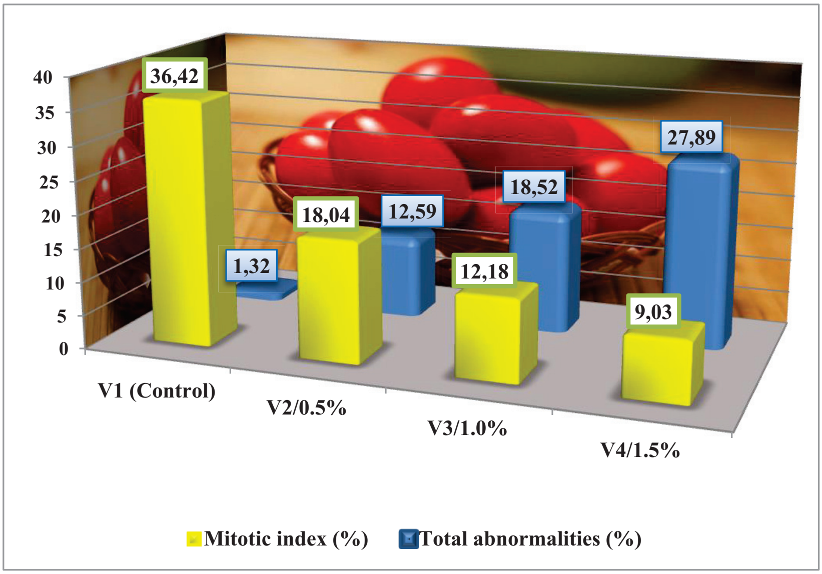

Figure 1. The mitodepresive and genotoxic effect of red dye on the meristematic cells to *A. cepa*

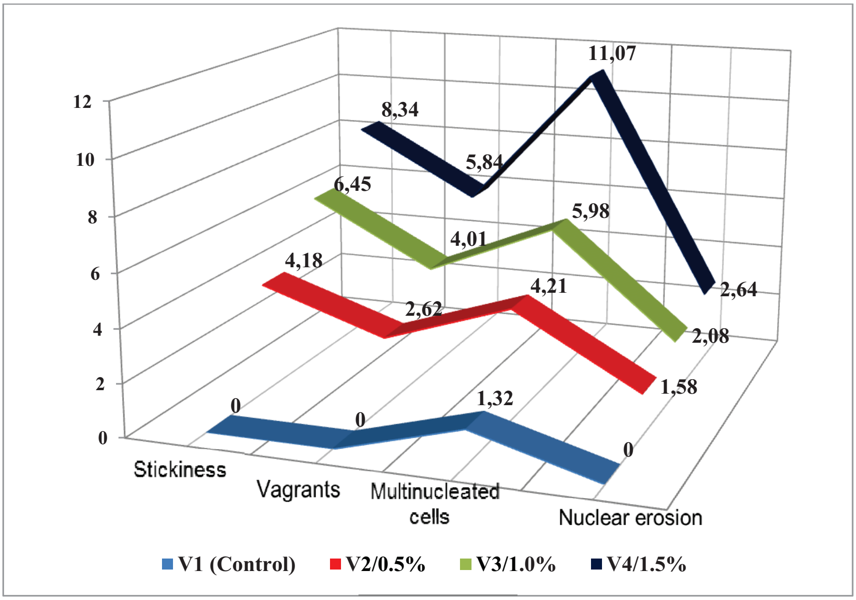

Figure 2. The frequency (%) of aberrant cells in meristematic tissues of *A. cepa* exposed to different concentrations of the commercial red dye

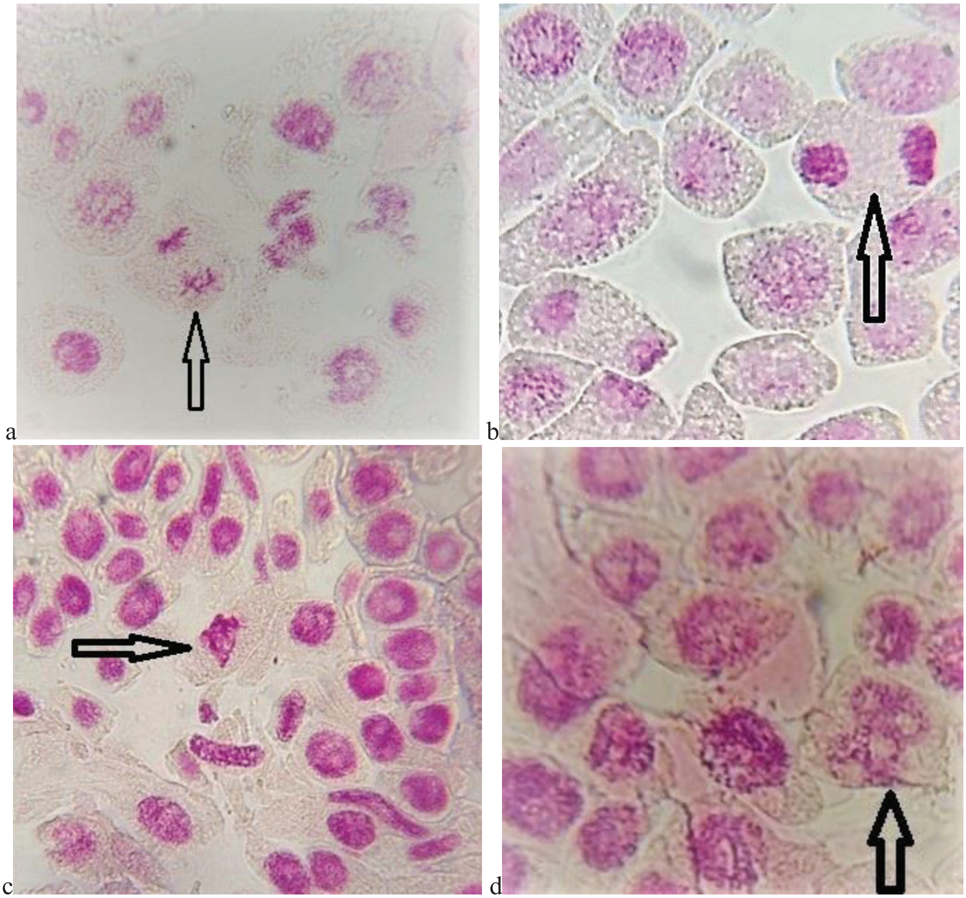

Figure 3. Some cytogenetic abnormalities in meristematic roots of *A. cepa* exposed to commercial red dye: disturbed anaphase with vagrants chromosomes (a); sticky telophase (b); sticky metaphase (c); multinucleated cell (d)

CONCLUSIONS

The cytological results observed in this study suggest that red dye induced a mitodepresive and genotoxic effects in *A. cepa* root tips, by reduction of the mitotic index and occurrence a several cytological abnormalities.

Multinucleated cells and stickiness were the dominant abnormality induced after treatment, especially at higher concentrations.

These results suggest prudence in using the red dye for eggs painting. An alternative solution to avoid the toxic effect of this chemical food colorant is the use of natural herbal extracts such as red beet juice, pomegranate juice, red onion peels, red peony petals etc.

REFERENCES

Abebe, A., Aklilu, M., Sisay, T. (2016). Assessment of knowledge and perception of health professionals towards toxoplasmosis in selected towns of Ethiopia. *Banat's Journal of Biotechnology*, *VII*(14), 113–119.

Antonsie-Wiez, D. (1990). Analysis of the cell in the root meristem of *Allium cepa* under the influence of Ledakrin. *Folia Histochemical Cytobiology*, *26*, 79–96.

Bonciu, E., Firbas, P., Fontanetti, C.S., Wusheng, J., Karaismailoğlu, M.C., Liu, D., Menicucci, F., Pesnya, D.S., Popescu, A., Romanovsky, A.V., Schiff, S., Ślusarczyk, J., De Souza, C.P., Srivastava, A., Sutan, A., Papini, A. (2018). An evaluation for the standardization of the *Allium cepa* test as cytotoxicity and genotoxicity assay. *Caryologia*, *71*(3), 191–209.

Bonciu, E. (2018). Evaluation of cytotoxicity of the herbicide Galigan 240 EC to plants. *Scientific Papers. Series A. Agronomy*, *LXI*(1), 175–178.

Center for Science in the Public Interest (CSPI). (2010). Food Dyes: A Rainbow of Risks. Available on https://cspinet.org/resource/food-dyes-rainbow-risks

FDA. (2016). Overview of food ingredients, additives & colors. Available on http://www.fda.gov/Food/IngredientsPackagingLabeling/FoodAdditivesIngredients/ucm094211.htm

Martins, N., Lobo Roriz, C., Morales, P., Barros, L., Ferreira, I.C.F.R. (2016). Food colorants: Challenges, opportunities and current desires of agro-industries to ensure consumer expectations and regulatory practices. *Trends in Food Science & Technology*, *52*, 1–15.

Ouis, N., Hariri, A. (2018). Antioxidant and antibacterial activities of the essential oils of *Ceratonia siliqua*. *Banat's Journal of Biotechnology*, *IX*(17), 13–23.

Panda, B.B., Sahu, U.K. (1985). Induction of abnormal spindle function and cytokinesis inhibition in mitotic cells of *Allium cepa* by the organophosphorus insecticide feusul fothion. *Cytobios*, *42*, 147–155.

Righi, K., Assia Righi, F., Boubkeur, A., Boungab, K., Elouissi, A., Djendara, A.C. (2018). Toxicity and repellency of three Algerian medicinal plants against pests of stored product: *Ryzopertha dominica* (Fabricius) (Coleoptera: *Bostrichidae*). *Banat's Journal of Biotechnology*, *IX*(17), 50–59.

Vazhangat, P., Thoppil, J.E. (2016). Apoptotic induction via membrane/DNA damage and metabolic inactivation by synthetic food colorants in *Allium cepa* root meristem. *Turk J. Biol.*, *4*, 922–933.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RELATIONSHIPS BETWEEN YIELD AND ASSOCIATED TRAITS OF MAIZE HYBRIDS UNDER DROUGHT STRESS AND NON-DROUGHT ENVIRONMENTS

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Abstract

Grain yield is one of the most important traits in maize breeding programs and depends on the association of different plant traits. Twenty maize hybrids were evaluated to explore the relationships between yield and associated traits, at Agricultural Research and Development Station Simnic. This experiment was conducted under two environmental conditions: 2017 (drought stress) and 2018 (non-drought) and was planted in a randomized block with three replications. Significant differences (p≤0.05) were found for all the studied traits excepting: the days to 75% anthesis, the days to physiological maturity and the protein content under drought stress conditions. In non-drought conditions were found significant differences for all the studied traits, excepting the days to physiological maturity and the shelling percentage. Correlation analysis revealed that grain yield exhibited a significant negative correlation only with hectolitre weight (r = -0.531^{0}) in drought stress conditions, and significant positive correlation only with thousand grains weight (r = 0.485) in non-drought conditions.*

***Key words**: correlation, drought, hectolitre weight, maize, thousand grains weight.*

INTRODUCTION

Maize is an important cereal crop of the world holding second position after rice in terms of production. In 2017, maize was produced in 2405 thousand hectare with the production of 14326 thousand tons with average yield of maize is 5.9 tons/ha in Romania (FAO, 2017).

Heat and drought are two factors that influence equally the maize crop evolution in the Romanian region of Oltenia. Water stress and heat lead always to different yield losses levels depending on the constrainer length, its intensity and crop stage (Bonea & Urechean, 2011; Pandia, 2006; Pandia et al., 2013; Urechean & Bonea, 2017). It is known that drought tolerance is a complex trait making the search for efficient selection traits, breeding and screening methods difficult (Petcu et al., 2018).

In cereal breeding programs, grain yield is one of the most important and complex traits and depends upon a combination of different plant traits (Mirosavljevic et al., 2015).

According to Georgieva and Kosev (2018), a new priority in the plant breeding is creating cultivars combining high potential productivity and resistance to stressful environmental factors.

Therefore, an effective breeding program requires a proper understanding of the relationships between grain yield and associated traits. According to Banziger et al. (2000) secondary traits that have a phenotypic correlation with yield under drought conditions can be used as selection traits of drought tolerant maize hybrids.

It is important to know that breeding progress using grain yield and a given secondary trait in selection is greater than progress using grain yield alone.

Correlation coefficient reveals the strength of relationship among the group of traits, thus is one of the important biometrical tools for formulating a selection index. This also helps to decide the dependability of the traits that have little or no importance (Jayakumar et al., 2007). Phenotypic correlation involves both genetic and environmental factors.

The present study was, therefore, conducted to assess the relationships among yield and associated traits, through association analysis for grain yield improvement in different environmental conditions.

MATERIALS AND METHODS

Plant materials

Twenty Romanian maize hybrids were used in this study.

Field evaluation and experimental design

The experiment was conducted under two environmental conditions: 2017 (drought stress) and 2018 (non-drought) and was planted at ARDS Simnic, in a randomized block with three replications. This Station is located at $44^{0}19'$ N, $23^{0}48'$ E, and 182 m altitude, characterized by preluvosoil. Fertilization was done with 250 kg/ha ($N_{20}P_{20}K_{0}$) complex fertilizers before sowing and in vegetation (phase 8-10 leaves) with ammonium nitrate 250 kg/ha. Planting was done on April 10th - 2017 and April 24th -2018, respectively. In order to prevent negative effects of weeds, DUAL GOLD 960 - 1.5 l/ha applied immediately after sowing and with EQUIP 1.5 l/ha + BUCTRIL 1.0 l/ha in the growing season (6-8 leaves).Two mechanical and two manuals weeding were applied.

Climatically data

The years of the investigation, were characterized as follows: 2017 was considered a dry year (drought stress conditions) and 2018 was considered favourable for the growth and development of maize (non-drought conditions). In 2017, the rainfall deficiency was largely pronounced in June and August. The mean monthly temperatures were over the multiannual average in June, July, and August.

For this area, the rainfall from August had a decisive role in defining the production capacity in the draughty years as well as in the favourable years (Urechean et al., 2010).

Field measurements

Grain yield and 10 secondary traits were measured or calculated.

Grain yield per hectare adjusted to 15.5% moisture. In each plot, days to anthesis (DA) and days to silking (DS) were recorded as the number of days from sowing to when 75% of the plants had shed pollen and emerged silks, respectively.

For measurement of physiological maturity, regular sampling of two cobs per plot was done to assess the presence of black layers at the base of the grains.

Plant and ear heights were measured in centimetre as the distance from the base of the plant to the height of the first tassel branch and the node bearing the upper ear, respectively.

Shelling percentage (SP) was calculated by using the following formula:

$$\mathrm{SP} = \frac{\text{Grains weight of 10 ears}}{\text{Total weight of 10 ears}} \text{x100}$$

Hectolitre weight (HW) was determined using Hectolitre Weight Apparatus.

Thousand grains weight (1000-grain weight/TGW) data was recorded by weighing thousand grains randomly taken, with the help of electronic balance, and then average weight was calculated.

The protein content (PC) and oil content (OC) of the maize grains was determined by Inframatic 9140 (Sweden or Foss Infratec 1241, Denmark).

Statistical analysis

The data collected were subjected to ANOVA: single factor and differences between mean values were tested by F-test and separated using the Least Significant Differences (LSD) at 5% level of probability. The relationships between the yield and associated traits were established using Pearson correlation coefficient (r). The variability presence in the hybrids was estimated by coefficient of variations (CV) using the procedure suggested by Săulescu and Săulescu (1967).

RESULTS AND DISCUSSIONS

Mean performance and analysis of variance

Mean values and significant levels of yield and yield associated traits of twenty maize hybrids were presented in Tables 1 and 2.

The results showed that under drought stress conditions, except for days to anthesis, days to maturity and protein content, and in non-drought conditions, except for days to maturity and shelling percentage, there were significant differences ($p \leq 0.05$) between studied hybrids (Tables 1 and 2). Variation in grain yield and associated traits was reported by numerous researchers for different environmental conditions. Raut el al. (2017) observed highly significant variation in grain yield, days to 50% tasseling, days to 50% silking, plant height, ear height and thousand grains weight, while comparing fourteen maize genotypes. Muchie and Fentie (2016) also found highly significant variation for days to 50% anthesis, plant height, ear height and grain yield.

In this study, the mean grain yield per hectare was 5.26 ± 0.13 tons/ha in drought stress and 9.22 ± 0.18 tons/ha in non-drought conditions. Significant differences (0.15 and 0.35) between studied hybrids were found for grain yield in both environmental conditions (Tables 1 and 2).

The mean value for day to 75% anthesis was 82.35 ± 0.36 days under drought stress, and 68.10 ± 0.59 days under non-stress conditions. ANOVA revealed significant difference only in non-drought conditions (2.90) for this trait.

The mean value for days to 75% silking among 20 hybrids was 85.20 ± 0.64 days in drought stress, and 69.95 ± 0.62 days in non-drought conditions. There were highly significant differences (3.56 and 2.95) among hybrids for this trait in both environmental conditions.

The mean value for days to maturity was 124.75 ± 0.73 days in drought stress and 123.45 ± 0.81 days in non-stress conditions. ANOVA revealed non-significant difference among hybrids for this trait in both conditions.

The mean value for plant height among experimented hybrids was 184.15 ± 4.04 cm in drought stress and 243.05 ± 4.46 cm under non-drought conditions. ANOVA revealed significant differences (5.72 and 8.50) among 20 hybrids, in both conditions.

The mean value for ear height was 73.65 ± 2.07 cm in drought stress and 96.95 ± 2.36 cm in non-drought conditions. ANOVA revealed significant difference among hybrids for this trait in both conditions (1.49 and 3.00).

The mean value for shelling percentage was 82.05 ± 0.69% in drought stress and 84.25 ± 0.42% in non-drought conditions. There were significant differences only in drought stress (3.41) among hybrids for this trait.

The mean value of hectolitre weight for the studied hybrids was 69.35 ± 0.46 kg/hl in drought stress and 71.15 ± 0.48 kg/hl in non-drought conditions, revealed significant differences were found among hybrids for this trait in both conditions (3.06 and 3.21).

The mean value of thousand grains weight for the experimented hybrids was 236.60 ± 6.42 g in drought stress and 319.80 ± 10.52 g in non-drought conditions, revealed significant differences were found among hybrids for this trait in both conditions (6.20 and 6.30).

The mean value for protein content of maize hybrids was 14.55 ± 0.15% in drought stress and 13.66 ± 0.18% in non-drought conditions. ANOVA revealed non-significant difference among 20 hybrids for this trait in drought stress and significant difference (0.54) in non-drought conditions.

For oil content, the mean value was 5.11 ± 0.09% in drought stress and 4.80 ± 0.1% in non-drought conditions. ANOVA revealed significant differences were found among hybrids for this trait in both conditions (0.19 and 0.13).

The differences in CV% values for different traits has been observed (Tables 1 and 2), indicating environmental conditions influence over these traits. Among the studied traits, moderate CV values (10-20%) were observed for grain yield, ear height and thousand grains weight in drought stress, and for ear height, thousand grains weight and oil content in non-drought conditions.

The other traits had low CV (0-10%) values in both conditions (Tables 1 and 2).

Correlation Coefficient Analysis

Analysis of correlation coefficient of yield related traits revealed some fundamental basis (Tables 2 and 4).

Under drought stress conditions, correlation coefficients of studied traits indicated that the grain yield has been correlated significant negative only with hectolitre weight ($r = -0.531^0$) (Table 3). The result is contradictory where Shiri et al. (2013) that found non-significant correlation of grain yield and hectolitre weight in thirty-six new late maturity maize hybrids under drought stress.

Thus, our findings suggested that the selection of hybrids having lower hectolitre weight should be the priority of breeders to achieve higher yield in drought stress conditions.

As well were significant negative correlations between days to 75% anthesis and oil content ($r = -0.530^0$), days to 75% silking and protein content ($r = -0.521^0$), plant height and oil content ($r = -0.501^0$), ear height and oil content ($r = -0.497^0$), shelling percentage and oil content ($r = -0.487^0$).

Table 1. Mean ± SEd, F test, CV% and LSD (at p≤0.05) of yield and associated traits at 20 maize hybrids under drought stress (2017)

Hybrid	GY	DA	DS	DM	PH	EH	SP	HW	TGW	PC	OC
H1	5.71	83	85	128	200	77	81	68	176	15.1	5.6
H2	4.47	82	84	128	188	75	85	69	190	15.4	5.0
H3	5.56	84	86	128	193	73	84	70	262	15.2	5.3
H4	6.20	84	86	128	210	78	82	67	254	14.6	4.6
H5	6.52	85	87	128	195	75	81	67	290	13.9	4.5
H6	5.87	83	95	128	183	70	83	68	210	12.9	4.9
H7	4.59	85	86	118	190	73	86	69	248	13.3	4.6
H8	5.52	81	83	125	190	70	80	67	254	14.4	4.9
H9	5.21	83	85	125	183	83	82	74	210	15.0	5.1
H10	5.81	82	83	118	175	78	86	66	212	13.6	4.9
H11	5.01	82	84	125	177	73	81	68	204	15.2	4.7
H12	4.40	82	84	125	169	75	85	69	246	14.4	4.9
H13	5.31	83	85	125	170	70	86	70	262	15.1	5.4
H14	5.10	81	83	125	200	65	78	69	258	15.1	5.5
H15	5.18	79	81	125	160	70	81	73	262	14.2	5.7
H16	4.66	81	85	125	185	75	83	70	252	14.7	5.1
H17	4.72	83	87	125	230	105	82	71	244	14.7	4.7
H18	4.77	83	87	125	160	63	82	72	224	14.3	5.4
H19	4.80	79	82	118	162	60	80	71	226	15.3	5.5
H20	4.97	82	86	123	163	65	73	69	248	14.6	5.9
Mean	5.26	82.35		124.75	184.15	73.65	82.05	69.35	236.60	14.55	5.11
± SEd	0.13	0.36	0.64	0.73	4.05	2.07	0.69	0.46	6.42	0.15	0.09
F test	*	ns	*	ns	*	*	*	*	*	ns	*
LSD5%	0.15		3.56		5.72	1.49	3.41	3.06	6.20	-	0.19
CV%	10.83	1.98	3.36	2.62	9.83	12.57	3.76	2.99	12.15	4.74	8.02

GY - Grain yield per hectare (t), DA- Days to 75% anthesis , DS - Days to 75% silking, DM -Days to maturity, PH - Plant height (cm), EH - Ear height (cm), SP - Shelling percentage (%), HW - Hectolitre weight (kg/hl), TGW - Thousand grains weight (g), PC - Protein content (%), OC - Oil content (%), SEd - Standard error of mean of differences; * = Significant at p≤0.05

Table 2. Mean ± SEd, F test, CV% and LSD (at p≤0.05) of yield and associated traits at 20 maize hybrids under non-drought conditions (2018)

Hybrid	GY	DA	DS	DM	PH	EH	SP	HW	TGW	PC	OC
H1	8.27	69	71	118	235	93	85	71	302	13.5	5.1
H2	8.08	71	73	126	218	85	84	68	217	14.6	4.9
H3	8.03	73	75	128	230	95	81	71	304	14.2	4.9
H4	9.89	69	70	126	240	98	85	69	321	12.8	4.6
H5	9.05	65	66	118	255	78	86	70	338	11.8	3.8
H6	9.81	66	67	127	213	85	84	72	374	12.1	3.6
H7	9.44	66	68	118	245	98	82	75	289	13.6	5.3
H8	10.16	67	68	125	230	103	84	70	303	13.8	4.8
H9	7.98	66	68	125	240	103	86	75	290	13.5	4.8
H10	8.85	64	66	118	215	95	86	71	362	12.4	4.8
H11	8.90	67	70	125	275	100	86	73	323	13.6	4.4
H12	10.43	66	68	122	235	80	86	69	325	13.5	4.8
H13	9.74	68	70	126	235	80	85	74	344	14.5	5.3
H14	8.30	72	74	118	250	108	84	69	268	14.6	4.7
H15	9.88	69	71	128	270	105	85	67	375	14.0	4.9
H16	9.78	69	71	126	265	110	81	72	258	13.8	5.9
H17	8.48	66	68	122	230	100	83	73	274	13.8	4.9
H18	9.91	70	72	126	285	115	80	71	410	14.6	4.8
H19	8.84	66	68	122	235	103	86	72	370	14.5	4.8
H20	10.60	73	75	125	260	105	86	71	349	14.0	5.0
Mean	9.22	68.10	69.95	123.45	243.05	96.95	84.25	71.15	319.80	13.66	4.80
± SEd	0.18	0.59	0.62	0.81	4.46	2.36	0.42	0.48	10.52	0.18	0.1
F test	*	*	*	ns	*	*	ns	*	*	*	*
LSD5%	0.35	2.90	2.95		8.50	3.00		3.21	6.30	0.54	0.13
CV%	9.16	3.89	3.97	2.94	8.22	10.89	2.27	3.07	14.71	6.04	10.17

* = Significant at p≤0.05

Table 3. Pearson's correlation coefficient among yield and yield associated traits of 20 hybrids of maize under drought stress

Traits	GY	DA	DS	DM	PH	EH	SP	HW	TGW	PC	OC
GY	1	0.371	0.275	0.375	0.213	0.032	0.007	-0.531^{0}	0.224	-0.323	-0.271
DA		1	0.571**	0.283	0.458*	0.343	0.320	-0.286	0.099	-0.291	-0.530^{0}
DS			1	0.371	0.233	0.142	0.082	-0.154	-0.085	-0.521^{0}	-0.287
DM				1	0.367	0.183	-0.061	-0.048	0.017	0.219	-0.061
PH					1	0.714**	0.073	-0.275	0.068	0.068	-0.501^{0}
EH						1	0.279	0.045	-0.073	0.001	-0.497^{0}
SP							1	-0.052	-0.125	-0.215	-0.487^{0}
HW								1	0.007	0.293	0.437*
TGW									1	0.224	-0.088
PC										1	0.376
PO											1

*;^{0} and **;^{00} - significant at 0.05 and 0.01 level of probability, respectively

Table 4. Pearson's correlation coefficient among yield and yield associated traits of 20 hybrids of maize under non-drought conditions

Traits	GY	DA	DS	DM	PH	EH	SP	HW	TGW	PC	OC
GY	1	-0.042	-0.093	0.274	0.302	0.041	0.018	-0.165	0.485*	-0.118	0.036
DA		1	0.985**	0.383	0.270	0.327	-0.367	-0.366	-0.214	0.595**	0.296
DS			1	0.362	0.320	0.360	-0.353	-0.302	-0.226	0.662**	0.361
DM				1	0.148	0.151	-0.251	-0.108	0.132	0.278	0.049
PH					1	0.549*	-0.207	-0.036	0.262	0.255	0.167
EH						1	-0.395	0.053	0.058	0.435	0.381
SP							1	-0.110	0.163	-0.343	-0.361
HW								1	-0.018	-0.032	0.182
TGW									1	-0.216	-0.369
PC										1	0.600**
OC											1

*;^{0} and **;^{00} - significant at 0.05 and 0.01 level of probability, respectively

In addition, there were significant and positive correlation between days to 75% anthesis and, days to 75% silking (r = 0.571**) and plant height (r = 0.458*).
The correlation between plant height and ear height in drought stress was significant positive (r = 0.714**), this finding being in agreement with the results of Beiragi et al. (2011).
In this study, simple correlation coefficients of studied traits in non-stress conditions indicated that grain yield was correlated significant positive only with 1000-grains weight (r = 0.485*) (Table 4).
The result is in agreement with findings of Rahman et al. (2017) that found positive significant correlation of grain yield with thousand grains weight in 15 maize genotypes experimented under normal conditions.
Thus, our findings suggested that the selection of hybrids having high thousand grains weight should be the priority of breeders to achieve higher yield in non-drought conditions.
There were positive and significant correlation between days to 75% anthesis and days to 75% silking (r = 0.985**), days to 75% anthesis and protein content (r = 0.595**), days to 75% silking and protein content (r = 0.662**), plant height and ear height (r = 0.549*), protein content and oil content (r = 0.600**) (Table 4).
The results are partial contradictory where Sharma et al. (2016) found significant negative correlation of protein content and oil content in 81 genotypes of maize.
Mehri (2015) found that in both environments (normal conditions and water scarcity during grain filling stage) there was a significant positive correlation between grain yield and, thousand grain weight and hectolitre weight.

CONCLUSIONS

The relationships between grain yield and associated traits are the primary important data in maize breeding programs. Selection for high yielding hybrids should focus on the strongly positively associated traits of maize plant with its yield.

The results of this study revealed that grain yield exhibited a significant negative correlation only with hectolitre weight (r = -0.531^0) in drought conditions, and a significant positive correlation only with thousand grains weight (r = 0.485*) in non-drought conditions. Therefore, for yield improvement in maize hybrids as per the findings of the plant traits association studied, it is necessary selection based on plant with lower hectolitre weight under drought stress, and high thousand grains weight in non-drought conditions.

The research emphasized that yield and the relationships between grain yield and other associated traits are influenced by genotypes and environmental conditions studied.

REFERENCES

Banziger, M., Edmeades, G.O., Beck, D., Bellon, M. (2000). *Breeding drought and nitrogen stress tolerance in maize: From Theory to Practice.* Mexico, DF CIMMYT.

Beiragi, M.A., Ebrahimi, M., Mostafavi, K., Golbashy, M.,Khorasani, S.K.. (2011). A study of morphological basis of corn (*Zea mays* L.) yield under drought stress condition using correlation and path coefficient analysis. *Journal of Cereals and Oil Seeds*, *2*(2), 32–37.

Bonea, D., Urechean, V. (2011). The evaluation water stress in maize (*Zea mays* L.) using selection indices. *Romanian Agricultural Research*, *28*, 80–86.

FAO. (2017). Statistical data base. *Food and Agriculture Organizations of the United Nations*. Rome, Italia. http://www.fao.org/faostat/en/#data/QC

Georgieva N., Kosev, V. (2018). Adaptability and stability of white lupin cultivars. *Banat's Journal Biotechnology, IX*(19), 65–76.

Jayakumar, J., Sundaram, T., Arun Prabu, D., Ragu Rama Rajan, A. (2007). Correlation studies in maize (*Zea mays* L.) evaluated for grain yield and other yield attributes. *International Journal of Agriculture Sciences, 3*(2), 57–60.

Mehri, S. (2015). Assessment of the performance correlation, agronomic characteristics, and drought tolerance indices in corn hybrids under late season moisture stress conditions. *Cumhuriyet Science Journal*, *36*(3), 586–594.

Mirosavljevic, M., Przulj, N., Canak, P., Momcilovic, V., Acin, V., Jockovic, B., Hristov, N., Mladenovet, N. (2015). Relationship between grain yield and agronomic traits in winter barley. *Ratarstvoi Povrtarstvo*, *52*(2), 74–79.

Muchie, A. Fentie, D. (2016). Performance evaluation of maize hybrids (*Zea mays* L.) in Bahir Dar Zuria District, North Western Ethiopia. *International Invention Journal of Agricultural and Soil Science*, *4*(3), 37–43.

Pandia, O.(2006). *Research regarding the effect of fertilizers upon maize production and quality*. Doctoral dissertation, Timişoara.

Pandia, O., Sărăcin, I., Bozgă, I., Marin, Gh. (2013). The modification of phisyological processes at the Partizan crop hybrid depending on the doses of nitrogen and phosphorus applied to the irrigated and un-irrigated system. *Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series*, *43*(1), 267–271

Petcu, E., Martura, T., Ciocăzanu, I., Iordan, H.L., Băduț, C., Urechean, V. (2018). The effect of water stress induced with PEG solution on maize seedlings. *Romanian Agricultural Research*, *35*, 21–28.

Rahman, M., Hoque, A., Hossain, Md. A., Al Bari, Md. A. (2017).Variability and traits association analyses in maize (*Zea mays* L.) genotypes**.** *The Agriculturists, 15*(2), 101–114.

Raut, S.K., Ghimire, S.K., Kharel, R., Kuwar, C.B., Sapkota, M., Kushwaha, U.K.S. (2017). Study of yield and yield attributing traits of maize. *American Journal of Food Science and Health, 3*(6), 123–129.

Săulescu, N.A., Săulescu, N.N., (1967). *Câmpul de experiență*. Editura Agro-Silvică, București.

Sharma, P., Punia, M.S., Kamboj, M.C. (2016). Association analysis of yield and quality attributes in maize. *Forage Research., 42*(3), 204–207.

Shiri, M., Momeni, H., Geranmayeh, B. (2013). The survey of the morphological and physiological basis of maize grain yield under drought stress condition through Path analysis. *Technical Journal of Engineering and Applied Sciences, 3*(24), 3647–3651.

Urechean, V., Bonea, D., Borleanu, I.C. (2010). The influence of climate on maize production in the centre of Oltenia. *Maize Genetics Cooperation Newsletter*, *84*, 14–15.

Urechean, V., Bonea, D., Constantinescu, E. (2012). Behaviour of some sunflower hybrids cultivated at ARDS Simnic, under climatic conditions of 2007 year. *Annals of the University of Craiova-Agriculture, Montanology, Cadastre Series*, *42*(2), 268–271.

Urechean, V., Bonea, D., (2017). Estimate of drought tolerance at some maize hybrids grown in the central Oltenia zone with using stress tolerance indices. *17th International Multidisciplinary Scientific GeoConference SGEM 2017, Conference Proceedings*, *17*(61), 681–688.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE INFLUENCE OF THE RESERVE LIPIDS CONTENT ON SEED GERMINATION AT SUNFLOWER HYBRIDS

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Abstract

The sunflower species Helianthus annuus L. belongs to the group of oil plants and it is considered to be one of the most important sources of vegetable oil used in human nutrition. The analyzed plant material was represented by the following hybrids: Armada Clearfield, Duet Clearfield, Meteor Clearfield, DT5234 Clearfield, P64LE25, DT6004, EXS7016 and Pokora. There have been made determinations regarding germination capacity, seed vigor expressed by electrolyte leakage, reserve lipids content, lipase activity and root length increase. Duet and Armada achieved the highest germination capacity, the leakage of electrolytes during imbibition directly influencing seed germination. The lipid content of the analyzed hybrids did not similarly influence germination and the radicle development took place 48 hours after the induction of the germination process, which can be explained by a quick mobilization and biodegradation of the organic substances under the influence of the lipase's increased activity.

Key words: *germination, electrolytes, lipase, lipids, sunflower seeds.*

INTRODUCTION

Sunflower belongs to the Asteraceae family and is one of the most important oleaginous seeds crops in the world. Achenia accumulates 49.9-55.5% lipids, 16.4-18.4% proteins and 5-14% carbohydrates. Sunflower achenia lipids contain 84% of unsaturated fatty acids and 18% of saturated fatty acids (Burzo, 2005).

Imbibition is the first process that occurs when seeds are placed in optimal germination conditions. The penetration of water into the seed and the increase in mass happen very fast. Thus, Levari (1960) stated that at a temperature of 28°C a very obvious increase of the seed mass was observed during the first 2 hours of imbibition. The free ion content of sunflower seeds decreases during the germination period of 2.13 times as a result of their diffusion into the water of imbibition. The diffusion of cellular electrolytes into the soaking water causes the conductivity to increase to 4.468 μS/g seeds. After 24 hours of imbibition, the permeability drops to 50% and after 3 days reaches 38.2% of the initial value, which indicates the restoration of the structure and their semipermeability (Burzo, 2005).

Seed germination is the process by which they initiate a new ontogenetic development cycle when favorable environmental conditions are ensured. Germination begins with water uptake by the seed (imbibition) and the emergence of embryonic axis, usually the radicle, through the structures surrounding it (Bewley et al., 2013; Erbas et al., 2016). Sucrose synthesized during maturation of oil seeds is the source of carbon for triacylglycerol (TAG) synthesis (Bewley & Black, 1994). TAGs are hydrolysed by lipases, enzymes catalyzing the hydrolytic cleavage of the fatty acid ester bonds to yield glycerol and free fatty acids (Theimer & Rosnitschek, 1978). Almost all TAGs present in oleiferous seeds are lost during seed germination and seedling development (Muto & Beevers, 1974; Yaniv et al., 1998; Rabiei et al., 2007; Kim et al., 2011; Tonguç et al., 2012).

The purpose of this paper was to investigate the germination capacity of sunflower hybrids, seed vigor expressed by electrolyte leakage, reserve lipid content, lipase activity and rootlet growth in lenght.

The results obtained are useful for agronomists for the initiation of culture, but also for breeders, in order to characterize and support for homologation the created hybrids.

MATERIALS AND METHODS

The homologated plant material is represented by Armada Clearfield, Duet Clearfield, Meteor Clearfield, P64LE25 and Pokora, while the hybrids proposed for approval are represented by: DT5234 Clearfield, DT6004 and EXS7016. Of the 8 analyzed hybrids, 7 come from Fetesti, a small town from the southeastern area of Ialomita County. The agricultural enterprise which has cultivated them has got an experience of over 14 years in the production of cereals and technical plants. The P64LE25 hybrid, belonging to DuPont Pioneer, was cultivated in the southern part of Teleorman County, in Beiu Village of Ştorobăneasa.

Determination of seed vigor

Determination of electrolyte leakage during the seeds imbibition period is an indicator in the assessment of germination capacity.

The method is based on the determination of the electrical conductivity of the water in which the seeds of known mass have been maintained for 24 hours.

20 seeds were weighed, immersed in 20 ml of distilled water and kept under these conditions for 24 hours. Using the automated conductivity meter, measurements of the conductivity of the solution were made 24 hours after the start of the experiment.

Determination of the lipid content

The lipid content of seed of sunflower hybrids was determined gravimetrically using the Soxhlet method.

The amount of lipids was calculated using the following ratio:

Lipids (%) = (a-b) * 100/m, where:

a is gross mass of the collector vessel;

b -collector vessel country;

m - mass of seed analyzed.

Determination of lipase activity from the studied sunflower hybrids

Lipase is part of the hydrolases class, the group of esterases, catalyzing the cleavage of the ester linkages between glycerol and lipid fatty acids. Lipase activity is determined by measuring the increase in acidity of some lipids as a result of the action of an enzyme preparation obtained from oilseeds. Free fatty acids resulting from hydrolysis of triglycerides under the action of lipase are titrated directly with sodium hydroxide solution. The ideal substrate for the study of lipase activity in some plant material are lipids from the same material.

0.2 g sunflower seeds were crashed and mixed with 3 ml sunflower oil. 2 ml 0.5 M acetate buffer (pH 4.7) was added and the pestle mill was placed in a thermostat at 30°C for 30 minutes. Subsequently, 15 ml 96% ethanol and 15 ml of petroleum ether were added. The fatty acids in the sample were titrated with 0.1 N NaOH in the presence of phenolphthalein as an indicator.

In parallel, a blank sample was made, which was immediately titrated with 0.1 N NaOH. The lipase activity was expressed in micromoles of oleic acid formed by the action of lipase per minute and grams of enzyme extract.

Observing rootlet growth

To observe the growth of the radicle measurements were made 24 to 24 hours after its occurrence for 3 days.

For this determination Petri dishes were taped with filter paper moistened with distilled water. A sample of 10 pre-germinated seeds was transferred to each flask. The measurements were performed for all analyzed hybrids and the data obtained is in the tables.

RESULTS AND DISCUSSIONS

Determination of imbibition and germination capacity of the analyzed hybrids

The research on the imbibition capacity of the 8 sunflower hybrids studied showed that the highest weighing pressure was achieved in the POKORA hybrid, where 70% of the seeds split the pericarp, followed by the DT6004 hybrid with 56% and the METEOR hybrid with 44%. The smallest values, of 1%, were recorded in the ARMADA and DUET hybrids. Intermediate values were recorded at the hybrids DT5234, EXS7016, P64LE25 at 1.9, 3.18 and 5.38 times smaller compared to the POKORA hybrid (Figure 1). It can be appreciated that the imbibition process was carried out at a different rhythm under the influence of the hybrid type.

The analysis performed on the hybrids proposed for approval, namely: DT6004, DT5235 and EXS7016 indicate a high rate of imbibition in the DT6004 hybrid, 1.51 times higher than that of the hybrid DT5234 CL and 2.54 times higher than the hybrid EXS7016.

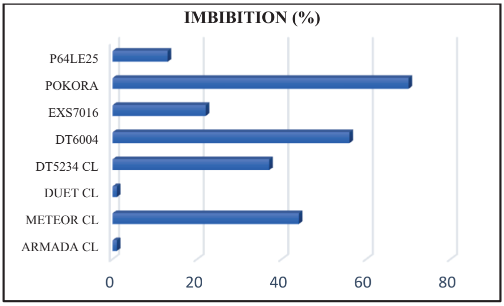

Figure 1. Imbibition capacity of the analyzed hybrids

In the next stages of the germination process, the germination was followed dynamically. After 48 hours from the start of the process, the following results were recorded: P64LE25 96%, EXS7016 79%, DT5234 75%, POKORA 93%, DT6004 88%, and METEOR CL 86%. Therefore, the germination capacity increased 7.38 times for P64LE25, 3.59 times for EXS7016, 2.02 times for DT5234 CL, 1.32 times for POKORA, 1.57 times for DT6004 and 1.95 times METEOR CL. However, a spectacular germination capacity can be noticed after 48 hours of optimum germination for DUET CL 47% and ARMADA CL 31%, given that during the imbibition period, in the two hybrids, a single seed (1%) presented the fragmented pericarp.

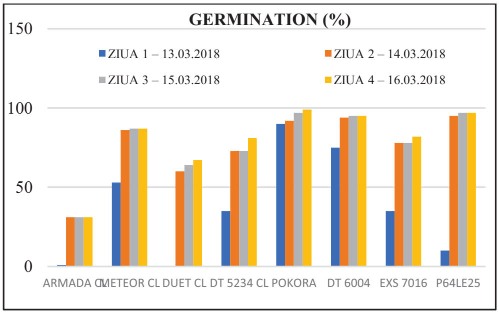

Figure 2. Germination capacity of the analyzed hybrids

Noteworthy is the fact that in the case of sunflower seeds, the formation of the rootlet takes place after 48 hours, which can be explained by rapid mobilization and biodegradation of the underlying organic substances under the influence of increased lipase activity. Thus, the optimal conditions for growth and development of the embryo are created, making possible the appearance of the first organ of the new plant. It is interesting to follow the germination process 72 hours after its initiation. Thus, the data presented in the graph indicates the completion of the germination, physiologically, of the analyzed hybrids.

The results obtained allow hybrids to be graded after the germination as follows:

- a first category is the P64LE25 and POKORA hybrids with over 95% germination;
- a second category is represented by hybrids whose germination capacity is between 75 and 94%, namely: DT5234, EXS7016, DT6004, hybrids proposed for approval, and METEOR CL, approved hybrid;
- in the third category is included the DUET hybrid, which is characterized by a germination capacity of 52%;
- the lowest germinating hybrid of 31% is ARMADA CL.

The determinations continued on the fourth day of germination, noticing that the hybrids ARMADA CL, METEOR CL, P64LE25 did not change their germline capacity compared to the previous day. With respect to hybrids DT5234 CL, DT6004, EXS7016, POKORA and DUET CL, the results indicated an increase of germination from 1.03 to 1.3 times (Figure 2).

At the end of the determinations, the non-homologated hybrids: DT5234 CL, DT6004, EXS7016 are considered to be of the germline capacity range of 80 to 95%.

Determination of the vigor of sunflower seeds expressed as electrolytes leakage

Dehydrated seeds have structurally damaged plasma membranes. Thus, in contact with water in the soil solution, some of the soluble substances dissipate by diffusion. The amount of soluble substances that diffuse into the soil solution is dependent on the rate of recovery of the structural integrity of the plasma membranes, being correlated with the germination capacity of the seeds (Figure 3).

Experimentally, the amount of ions that diffuse into the soil solution is conductively determined and constitutes an indicator in the assessment of germination capacity of the seeds.

Seed vigor was evaluated by conductometrically measuring the amount of ions that diffused in water after 24 hours of initiation of imbibition.

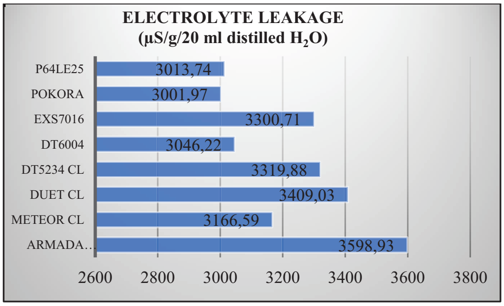

Figure 3. Values of electrolyte leakage of the sunflower seeds

The recorded data (Figure 3) indicates the highest values for the ARMADA hybrid, followed by DUET and EXS7016. The lowest values were determined at the POKORA and P64LE25 hybrids. Average values were obtained for hybrids METEOR, DT5234, DT6004. The results obtained and compared with the germination faculty indicated a direct influence between the two indicators, corresponding to the data from the specialty literature (Table 1).

Thus, in hybrids where the conductivity values were high, seed vigor by germinating capacity was reduced. Also, in the case of hybrids where the electrolyte leakage expressed by conductivity was low, the germination capacity was high.

Table 1. Relationship between germination and the electrolyte leakage

Hybrid	Germination (%)	Electrolyte leakage (µs/g/20 ml h_2o)
ARMADA CLEARFIELD	31	3598.93
METEOR CLEARFIELD	87	3166.59
DUET CLEARFIELD	67	3409.03
DT5234 CLEARFIELD	81	3319.88
EXS7016	82	3300.71
DT6004	95	3046.22
POKORA	99	3001.97
P64LE25	97	3013.74

Determination of the seed's reserve lipids content

During the germination, the mobilization of the organic substances, which requires an enzymatic biodegradation, takes place. Thus, it results in more simple intermediate products that can be transferred from the endosperm to the embryo, ensuring growth. Considering the importance of seed organic substances reflected in the formation of the new plant, during the study, the lipid content was determined for each of the analyzed hybrids.

The highest lipid content was calculated for the DT6004 hybrid, followed by the POKORA hybrid at the 7.28% difference. The lowest content was recorded in hybrids DT5234 CL 43.4068%, METEOR CL 45.7996%, P64LE25 45.8924%, DUET CL 47.2989%. As for the POKORA and ARMADA CL hybrids, the recorded values exceed 50% (Figure 4).

The lipid content determined in the 8 hybrids does not affect the germinative capacity similarly. Thus, it is noted that the DT6004 hybrid, whose seeds have the highest lipid content, also has the highest 95% germination. At P64LE25, the lipid content was relatively low, of 45.8924%, but the germination capacity was 97%. Regarding ARMADA CL, even though the lipid content was not reduced (50.1016%), the hybrid recorded the lowest germination capacity, of only 32%.

Also, the hybrids METEOR CL, DUET CL, DT5234 CL, EXS7016 and POKORA do not find that there is a direct relationship between lipid content and germinative faculty**.**

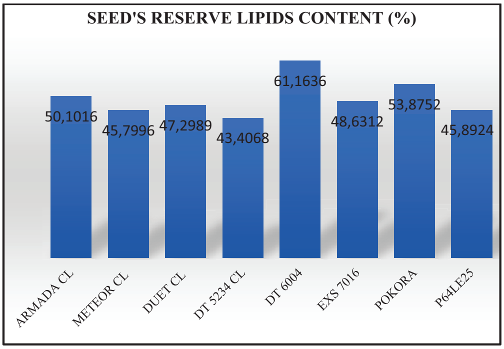

Figure 4. Values of the seed's content in lipids

Determination of lipase activity in sunflower seeds

Lipase is part of the hydrolases class, the group of esterases, catalysing the cleavage of the ester linkages between glycerol and lipid fatty acids. Regarding the activity of the lipase, the obtained results indicated that differences are caused by the characteristics of the hybrid and the physiological state of the seed (Table 2).

Table 2. Lipase activity

Hybrid	Lipase activity (µmole oleic acid/min/g seeds)		
	Non-germinated seeds	Seeds after a day of germination	Seeds after 3 days of germination
ARMADA CLEARFIELD	0.2	4.6	1.15
METEOR CLEARFIELD	0.26	2.9	0.7
DUET CLEARFIELD	0.065	0.9	0.2
DT5234 CLEARFIELD	0.05	0.5	0.12
DT6004	0.75	6.8	1.5
EXS7016	0.9	9.3	1.95
POKORA	0.45	2.1	0.68
P64LE25	0.47	1.9	0.62

Although the seeds were kept under the same temperature and humidity conditions, it can be observed that the non-germinated seeds of the DT5234 hybrid followed by the DUET hybrid show the lowest enzyme activity. The highest enzyme activity was calculated in the EXS7016 hybrid, its value being 0.9. For the other hybrids, values ranged between 0.2 and 0.7. With hydration of the seeds as a result of imbibition, the enzymatic activity intensifies at different rhythms, with high values being recorded in all studied hybrids.

The highest lipase activity was determined in the EXS7016 hybrid, followed by the DT6004 hybrid. It can be appreciated that, in sunflower seeds, the mobilization of reserve lipids is very rapid after only a day of germination, justifying the high activity of the lipase that caused the appearance of the rootlet.

Three days after germination, the enzymatic activity decreases remarkably, approximately 4 times, in all hybrids. The exceptions is the P64LE25 hybrid in which the enzyme activity decreases 3 times.

Measurement of the rootlet length in sunflower seeds

Of the total amount of hybrids analyzed for the rootlet extension, the hybrids DT6004 and POKORA have been noted (Table 3). In these, the growth of the rootlet on day one is 8.6 and 8.3 mm, respectively. There is a direct relationship between the lipid content and the roolet growth. Thus, the high lipid content has positively influenced the prolongation of the rootlet in the DT6004 and POKORA hybrids.

Noteworthy is that the DT6004 hybrid maintains its high growth rate on days 2 and 3 when it records the longest length of the rootlet compared to the other hybrids. It is estimated that the growth rate is almost constant from day to day, being 2.1 and 2.2 times higher respectively.

Table 3. Rootlet length (mm)

Hybrid	Day 1 14.03.2018	Day 2 15.03.2018	Day 3 16.03.2018
ARMADA CLEARFIELD	3.8	9.9	26.1
METEOR CLEARFIELD	7	13.8	24.1
DUET CLEARFIELD	3	9.3	26
DT5234 CLEARFIELD	5.7	13.1	32.5
DT6004	8.6	17.9	39.3
EXS7016	7	14.2	39.2
POKORA	8.3	14.3	30.4
P64LE25	4.8	10.6	26

Regarding the POKORA hybrid, the growth rate is not constant. Thus, on the first day, the extension of the rootlet is comparable to that of the DT6004 hybrid, the growth rate decreases on day two and registers a new increase on the third day. From the analysis of the data obtained from the 8 studied sunflower hybrids, the DT6004 hybrid shows a positive relationship between lipid content and lipase activity, these indicators favorably influencing rootlet growth throughout the research period.

It is also interesting to look at the EXS7016 and DT6004 hybrids, at the relationship between the lipid content, the lipase activity and the rootlet length, as it is found that in the EXS7016 hybrid, although the lipid content is lower,the lipase activity is the highest, causing a rapid mobilization of the reserve lipids. Thus, the rootlet growth was favored. This explains the similar value of the rootlet length to the two hybrids. Within 48 hours, the length of the EXS7016 hybrid rose 5.6 times, from 7 mm to 39.2 mm.

Regarding the characterization of the ARMADA CL, DUET CL and P64LE25 hybrids, it was found that the rootlet length did not exceed 4.8 mm on the first day. On day two, although the rhythm of growth is high, the rootlet length of these hybrids remains the smallest. The next day, the growth rate remains constant, the rootlet stretching to 26 mm in all 3 hybrids studied. Following the rhythm growth rate of all the analyzed hybrids, it is noted that on day two, the highest growth rate is recorded for DUET and the lowest for POKORA and METEOR hybrids. On the third day, in all analyzed hybrids, the growth rate was between 2.2 and 2.6 times, except for the METEOR hybrid at which the growth rate was the lowest, 1.75 times. Indeed, throughout the determinations, at the METEOR hybrid, the growth rate was the lowest, 1.86 times.

Also, in the analyzed timeframe, in the case of the DT5234 hybrid, there is an increase of the radicle from 5.7 to 32.5 mm, the elongation being 5.7 times. Thus, it is considered that the rhythm of growth was the highest of all the analyzed hybrids.

CONCLUSIONS

The properties of the hybrids have influenced the imbibition process. Thus, the highest imbibition pressure was achieved at the POKORA hybrid, and the lowest pressure was recorded at the DUET and ARMADA hybrids. The dynamics of the germinating faculty 48 hours after the start of the process indicated an increased germination capacity of 7.38 times for the P64LE25 hybrid, 3.59 times for the EXS7016 hybrid, 2.02 for the DT5234 hybrid, 1.32 times for the the POKORA hybrid, 1.57 times for the DT6004 hybrid and 1.95 times for the METEOR hybrid. At the analyzed hybrids, the electrolyte leakage during imbibition directly influences the seeds germination. The lipid content determined in the 8 hybrids does not affect the germination degree similarly, and the rootlet formation in sunflower seeds takes place 48 hours after the germination process is induced, which can be explained by a rapid mobilization and biodegradation of organic back-up substances under the influence of the increased lipase activity.

REFERENCES

Bewley, J.D., Black, M. (1994). Seeds: physiology of development and germination. *Plenum Press*, New York, USA.

Bewley, J.D., Bradford, K.J., Hilhorst, H.W.M., Nonogaki, H. (2013). *Seeds physiology of development, germination and dormancy*. Springer Science+Business Media, LLC, 3rd Edition, ISBN 978-1-4614-4692-7.

Burzo, I., Dobrescu, A. (2005). *Fiziologia culturilor de câmp, Vol II*, Editura Elisavaros.

Erbaş, S, Tonguç, M., Karakurt, Y., Şanli, A (2016). Mobilization of seed reserves during germination and early seedling growth of two sunflower cultivars, *Journal of Applied Botany and Food Quality, 89*, 217–222.

Kim, H.T., Choi, U.K., Ryu, H.S., Lee, S.J., Kwon, O.S. (2011). Mobilization of storage proteins in soybean seed (*Glycine max* L.) during germination and seedling growth. *Biochim. Biophys. Acta*, 1814, 1178–1187.

Levari, R. (1960). Imbibition by Various Seeds During Germination, Ph.D. thesis, Hebrew University, Jerusalem, Israel,

Muto, S., Beevers, H. (1974). Lipase activities in castor bean endosperm during germination. *Plant Physiol.*, *54*, 23–28.

Rabiei, Z., Tahmasebi Enferadi, S., Vannozzi, G.P. (2007). Regulation of polyunsaturated fatty acids accumulation and characterization of linolenic acid after germination of sunflower seed. *Helia*, *30*, 175–182.

Theimer, R.R., Rosnitschek, I. (1978). Development and intracellular localization of lipase activity in rapeseed (*Brassica napus* L.) cotyledons. *Planta, 139*, 249–256.

Tonguç, M., Elkoyunu, R., Erbaş, S., Karakurt, Y. (2012). Changes in seed reserve composition during germination and initial seedling development of safflower (*Carthamus tinctorius* L.). *Turk. J. Biol.*, *36*, 107–112.

Yaniv, Z., Shabelsky, E., Schafferman, D., Granot, I., Kipnis, T. (1998). Oil and fatty acid changes in *Sinapis* and *Crambe* seeds during germination and early development. *Industrial Crops and Products* *9*(1), 1–8.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

ANALYSIS OF YIELD AND YIELD RELATED TRAITS IN SOME SUNFLOWER (*Helianthus annuus* L.) HYBRIDS UNDER CONDITIONS OF THE REPUBLIC OF MOLDOVA

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Abstract

The growth and productivity traits of twenty local sunflower hybrids have been evaluated, during two years, under Moldovan environmental conditions. It has been established that yield and yield related traits of analysed combinations depended both on the year of observation and the genotype. Environmental conditions affected especially the number of leaves, number and weight of seeds per head and 1000 seeds weight. Determination of relationships between seed yield and some morpho-physiological parameters, such as plant height, head diameter, leaf number, number and weight of total and full seeds per head and 1000 seeds showed high positive correlations between some traits. The analyses of Pearson correlation coefficients showed that the number and weight of total and full seeds per head and the weight of 1000 seeds had a high positive correlation with seed yield per surface unite. Also, a positive but low correlation with seed yield was found related the head diameter. In both years a significant positive correlation was established between plant height and number of leaves.

***Key words**: plant height, head diameter, seed weight, number of achenes per head, sunflower hybrids.*

INTRODUCTION

Helianthus annuus L. is a valuable source of edible vegetal oil due to its high content of oil (40-52%) rich in unsaturated fatty acids (83-91%), vitamins A, D, E, K, tocopherols and phytosterols (Khalifa & Awad, 1997; Velasco et al., 2014). Consumption of sunflower oil leads to the reduction of LDL cholesterol in humans and decreases the risks of heart attack, having positive effects on the health (Hu et al., 2001). Furthermore, sunflower seed cakes contain high concentration of proteins (35%), carbohydrates (18-20%) and could be used for animals feed (Ibrahim, 2012). According to Zagoul'ko et al. (2011) from one hectare of sunflower with a yield of 2.5 tons per hectare 1200 kg of oil, 800 tons of cakes (300 kg of protein), 500 kg of husk (70 kg of yeast), 1500 kg of sunflower heads (1000 kg of qualitative fodder), 25-30 kg of honey, and other useful bio-products could be obtained.

Considering its economic importance and role in human nutrition, the sunflower has become the third most important oilseed crop in the world following soybean and rape, with the average of 18 million hectares sown land and 40.0 million metric tons yield (FAOSTAT, 2015).

A similar trend has been revealed in the Republic of Moldova, where sunflower is grown almost on 25% of the total arable land, after wheat and corn, ranging from 225,000 to 400,000 ha per year (Moroz et al., 2015). In recent years, the area of sunflower in Moldova tripled, while yields have doubled (Sunflowers Excel in Ukraine, Romania, Bulgaria and Moldova in 2017). Sunflower yields varied between 1.5 and 2.1 tons per hectare with an average yield of 1.8 t/ha. The yield is affected by a complex of factors, such as the aggravation of the phytosanitary state due to failure of crop rotation, simplified cultivation technologies, lands degradation, low quality of seeds, as well as unfavorable environmental conditions, including drought, salinity and low temperature stress (Kandakov et al., 2012; Statistical databank). The Republic of Moldova is ranked 15-16th in the World Sunflower Seed Producers Rating (FAO, 2010).

In order to face increased consumer demand for high-quality sunflower seeds the effort of breeders are focused on obtaining of new highly productive hybrids, resistant to biotic

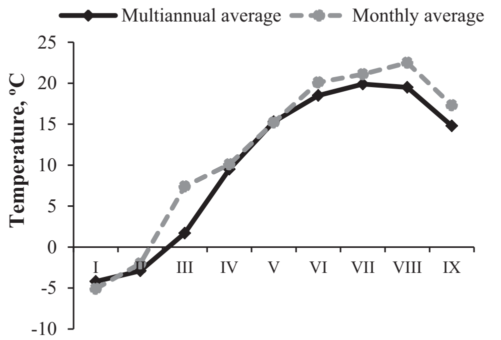
and abiotic factors, characterized by stable yield across different environments affected by climatic change (Mrdja et al., 2012). The main goals of sunflower breeding programs are: the yield over 4 t/ha, the husk share lower than 25%, more than 50% fat content in the seed, higher contents of healthy fatty acids, mostly oleic acid (Škorić, 1989; Kocjan Acko, 2008). Considering that seeds productivity and quality are greatly influenced by genotype, environment and their interaction the creation of new sunflower hybrids requires testing of breeding materials (Mrdja et al., 2012; Cerny et al., 2013). Plant phenology, crop growth period, as well as yield, oil and fatty acid accumulation is influenced by temperature, humidity, light levels and other environmental factors (Kaleem et al., 2010). There are many studies consisting in the evaluation of agronomic traits of different commercial or experimental sunflower hybrids in field conditions (Ullah et al., 2018; Ruzdik et al., 2015). Plant height, head diameter, number of seeds per plant and 1000 seeds weight are the main important parameters that determine yield improvement in sunflower (Kaya, 2015).
It is of great importance to know the relationships between yield contributing characters. Yield components, total leaf area, plant height, total seed per head and 1000 seeds weight were found to correlate with seed yield per plant (Hladni et al., 2004; 2010). Behradfar et al. (2009) reported that seed yield was positively associated with 1000 seeds weight and total seed number per head. According to Škorić (2012) the head diameter influences the number of seeds per head and represents a very important trait in the sunflower seed yield structure. A positive and important interdependence was determined among morpho-physiological traits like total leaf area, plant height, head diameter, mass of 1000 seeds and total number of seeds per head with seed yield and oil yield (Hladni et al., 2008; 2010)
The aim of this study was to evaluate, in two years, the growth and productivity of new sunflower hybrids grown under Moldovan environmental conditions in order to provide an insight into the effects of some weather parameters on productivity and identify the more stable genotypes.

MATERIALS AND METHODS

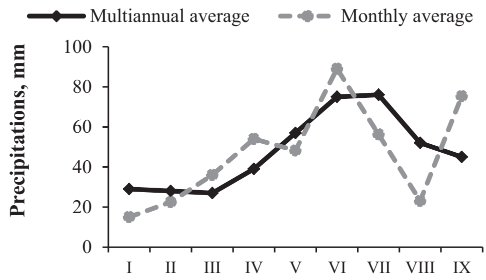
For the study were used 20 experimental sunflower hybrids created by the Company AMG-Agroselect Comert, Republic of Moldova (noted conventionally HM1-HM20). The experiment was performed during the growing seasons 2017 and 2018 in the field of AMG-Agroselect Comert Company, in Soroca district, Republic of Moldova, using the same growing technology in both years. The experimental field is located on the first terraces of the right bank of the Nistru River, at an altitude ranging from 53 to 77 m above sea level.
The trial design was randomized blocks with three replications. Each experimental plot consisted of 6 rows. The seeds were sown manually at a spacing of 0.35 m between plants and 0.70 m between the rows.
Plant height, head diameter, leaf number, number and weight of total and full seeds per head and 1000 seeds weight were established. At the stage of physiological maturity, ten plants were randomly selected from each replication and their height was measured from ground level to the top edge of the head and then their average was calculated. The head diameter was determined by measurements made for 10 plants in each block and repetitions in the field and calculation of average. Number of seeds per head was recorded from ten plants taken randomly from each replication and then their average was calculated.
To obtain thousand seed weight, 100 seeds taken randomly from each plot and replications were weighted using an electronic balance with an accuracy of 0.001 g and then multiplied by 10. Seed yield was measured separately from each plot and expressed as kg per hectare.
The meteorological data, monthly precipitation and daily temperature, as well as multiannual monthly average, were obtained from State Hydrometeorological Service of the Republic of Moldova.
To determine the relationships among the analysed traits the Pearson correlation coefficient between morphological characters (plant height, number of leafs, head diameter) and yield were obtained.

RESULTS AND DISCUSSIONS

The effect of the year on the sunflower yield differed due to different environmental conditions, such as soil or/and weather. According to Mijić et al. (2012) and Liović et al. (2006) rainfall and temperature are both important before and during the vegetation period, contributing to the replenishing of water reserves of soils or directly influencing the vegetative and generative development and, respectively, yield in sunflower. In order to study the influence of the year on sunflower yield and yield related parameters, the temperatures and rainfall amounts in 2017 and 2018 growing seasons were evaluated (Figures 1 and 2). The average annual temperature in 2017 growing season was 11.8°C, with lows of -5.1°C in January and highs of 22.5°C in August. In 2018 the average temperature was 11.5°C, with minimal value of -2.7°C in February and maximum 21.6°C in August (Figures 1a, 2a).



a.



b.

Figure 1. Average monthly temperatures (a) and amount of precipitations (b) for the growing season 2017 and multianual data

Generally, in both growing seasons the average temperature was higher by 1.3-1.6ºC compared to previous years, with increased temperatures, especially in summer months throughout the generative phase of sunflower. The average temperature in winter was -2.6ºC analogic with the multiannual data of -2.8ºC in 2017 and higher (-0.7ºC) in 2018.

Spring was characterised as warm with an average temperature of 11.0°C in both analyzed years exceeding the multiannual average by 2.2°C. In the summer, temperature was higher compared to previous years, with average values of 21.2ºC and 20.9ºC per season, respectively in 2017 and 2018, by 1.6-1.9ºC higher than the multiannual average. As described above, temperature values in both agricultural years are similar.

Most often precipitation comes in the form of rain, with an average of about 419.4 mm in 2017 and 307.5 mm in 2018 gowing season, being 8.6 mm and, respectively, 120.5 mm lower than multiannual data (Figures 1b, 2b).

Spring (April, May) and summer (June, August) months were characterized by a humidity deficiency, lower by 25-51.77 then the multiannual monthly average.

The analysis of some agro-morphological traits of new sunflower hybrids developed by the local company AMG-Agroselect Comert, in the experimental field, during 2017 and 2018 seasons, revealed that growth and yield parameters varied significantly. Since the genotypes were grown in the same field environment, applying similar agronomic practices, the difference in plant height, number of leaves, head diameter, seed number and seed weight per head, 1000 seeds weight, as well as seed yield of analyzed sunflower hybrids, in one year of investigation, could be explain by their different genetic potential and the differences in both years are additionally determined by deferentiated weather conditions.

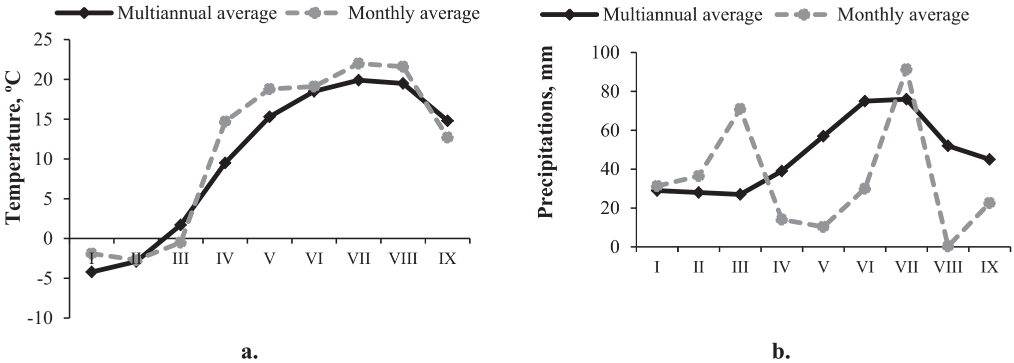


Figure 2. Average monthly temperatures (a) and amount of precipitations (b) for the growing season 2018 and multianual data

As shown in figures 3-9, for the majority of hybrids the values of yield and yield related traits were lower in 2018 compared to 2017. Considering that plants have been cultivated in relatively similar conditions, excepting the quantity of precipitation received, which was lower during 2018 vegetation period (146.1 mm) comparative to 2017 (270.6 mm), the decrease of analysed parameters could be mainly attributed to the inadequate water supply.

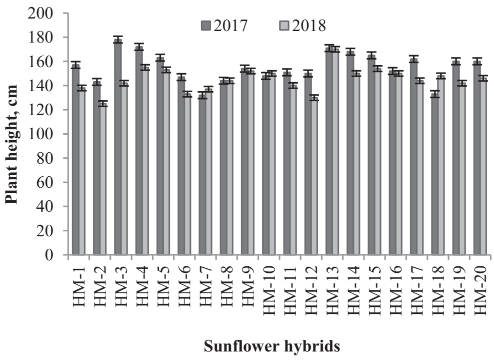


Figure 3. Plant height (cm) of twenty sunflower hybrids during two experimental periods (2017 and 2018)

The same findings were obtained by Yegappan et al. (1982) who reported that hydric stress significantly reduced number of leaf, head diameter, leaf area, weight of 1000 grains and grain yield. Đukić et al. (2011) stated that amount of precipitation and temperature conditions during the growing season are important limiting factors of yield levels and 1000 seeds weight.

Regarding the plant height, the values ranged between 132-178 cm in 2017 and 125-170 cm in 2018. The hybrids HM3, HM4 and HM13 were the tallest among tested genotypes, with the height of 171 to 178 cm in 2017, excepting HM3, the same hybrids were the tallest in 2018.

Compared to the tallest genotypes, the hybrids HM2, HM7 and HM18 were the lowest in 2017 with average height of 132 cm to 143 cm and HM2, HM6 and HM12 presented the minimal values (125-133 cm) in 2018. Plant height of a crop is the function of the combined effects of genetic makeup and the environment (Gvozdenović et al., 2005). Thus, different values in sunflower hybrids growing in identical field environments could be explained by their different genetic makeup. Marinkovic (1992) reported that plant height had a positive effect on seed yield. In our studies, no significant ($p<0.01$ or 0.05) correlations between plant height and seeds yield have been established (Table 1), the data being in accordance with those obtained by Papatheohari et al. (2016).

As shown in table 1, in both years a significant positive correlation, 0.5637 and 0.7074, in 2017 and 2018, respectively, were found between plant height and number of leaves. Divergent to the results of Hladni et al. (2010) and Khan et al. (2018), who reported a strong positive correlation of total leaf number per plant with the sunflower yield and seed oil content, in the present study, no significant correlations between the number of leaves and yield traits have been observed. According to the data, the highest leaves number (35 in 2017) was observed in HM4, one of the tallest hybrids and the least number of leaves (25 in

2017 and 18 in 2018) was sighted in HM18 and HM6, respectively, which were the smallest hybrids (Figure 4).

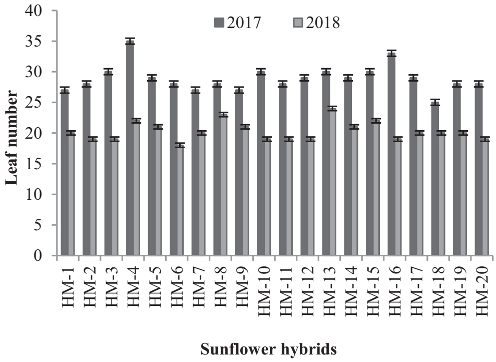


Figure 4. Number of leaves of twenty sunflower hybrids during two experimental periods (2017 and 2018)

The number of leaves per plant was significantly lower (by around 20-40%) in 2018 comparative to 2017 and may be due to a low quantity of rainfall during vegetation period. Obtained data are in agreement with the fact that plants under water deficit have reduced leaf growth, by the diminution of leaf number and/or reduction of area of individual leaves (Pereyra-Irujo et al., 2008).

Another important trait in the sunflower seed yield structure is the size of the head diameter, which influences the number of flowers and seeds per head and directly affects the seed yield per plant (Balalic et al., 2016). Data shown in Figure 5 revealed significant differences among head diameter of various sunflower hybrids. In the first analysed year, the largest head 24 cm was recorded in HM7, followed by HM1, HM4 and HM10.

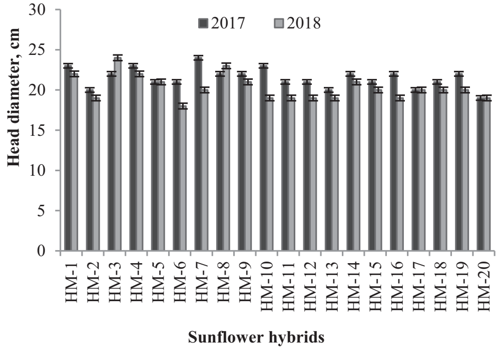


Figure 5. Head diameter (cm) of twenty sunflower hybrids during two experimental periods (2017 and 2018)

Table 1. Pearson correlation coefficients between some morpho-physiological traits and yield parameters in twenty sunflower hybrids

Traits	X2	X3	X4	X5	X6	X7	X8	X9
2017								
X1	0.5637*	-0.1539^{ns}	-0.1336^{ns}	-0.1055^{ns}	-0.3263^{ns}	-0.3414^{ns}	-0.1617^{ns}	-0.0762^{ns}
X2	-	0.1568^{ns}	-0.0147^{ns}	-0.0196^{ns}	-0.1963^{ns}	-0.2029^{ns}	-0.1112^{ns}	-0.1175^{ns}
X3	-	-	0.4871**	0.5415*	0.7290*	0.7188*	0.6350*	0.4546**
X4	-	-	-	0.9719*	0.5058**	0.5074**	0.3305^{ns}	0.5708*
X5	-	-	-	-	0.5905**	0.5532*	0.4180^{ns}	0.6145*
X6	-	-	-	-	-	0.9978*	0.6414*	0.5387**
X7	-	-	-	-	-	-	0.6340*	0.5285**
X8	-	-	-	-	-	-	-	0.7482*
2018								
X1	0.7074*	0.1287^{ns}	0.1513^{ns}	0.2748^{ns}	-0.0043^{ns}	-0.0276^{ns}	-0.0728^{ns}	-0.0498^{ns}
X2	-	0.3661^{ns}	0.3845^{ns}	0.3881^{ns}	0.1803^{ns}	0.1824^{ns}	0.0410^{ns}	0.3243^{ns}
X3	-	-	0.5135**	0.5620*	0.8338*	0.7931*	0.6473**	0.3822**
X4	-	-	-	0.8257*	0.4642**	0.4231**	-0.0467^{ns}	0.2262^{ns}
X5	-	-	-	-	0.6596*	0.6886*	0.1330^{ns}	0.5397*
X6	-	-	-	-	-	0.9884*	0.7915*	0.6117*
X7	-	-	-	-	-	-	0.7675*	0.6685*
X8	-	-	-	-	-	-	-	0.4997**

X1 - Plant height (cm)
X2 - Total leaf number per plant
X3 - Head diameter (cm)
X4 - Total seed number per head (g)
X5 - Full seed number per head (g)
X6 - Weight of total seed per head (g)
X7 - Weight of full seed per head (g)
X8 - Weight of 1000 seeds (g)
X9 - Yield (kg/ha)

* F test for significance at level P<0.01; ** F test for significance at level P<0.05 ; ns - not significantly different

The shortest head diameter (19 cm) was observed in hybrid HM20. In second year the maximal value (24 cm) has been sighted at HM3, followed by HM8 and HM1 and the lowest at HM6 (18 cm). Obtained data confirms the findings that sunflower head diameter depend on genotype and environmental conditions, as well as interaction between these factors (Hladni et al., 2014; Moisa & Smit, 2014). Diameter values are higher comparative to those reported by other authors (Iqrasan et al., 2017; Khan et al., 2018; Balalic et al., 2016) and could be considered as intermediate (ranged between 20-25 cm).

Similar with the results reported by many authors (Hladni et al., 2004; 2008; 2010; Kaya et al., 2009; Killi & Tekeli, 2016; Gvozdenović, 2005), significant positive correlations between head diameter and seed yield related traits have been found (Table 1).

Thus, head diameter highly correlates with the number of total and full seeds per head (*r* = 0,4871** and 0,5415* in 2017, respectively, 0,5135** and 0,5620* in 2018), weight of total and full seeds per head (*r* = 0,7290* and 0,7188* in first analysed year and *r* = 0,8338*; *r* = 0,7931* in second year), as well as 1000-seed weight (*r* = 0,6350* in 2017 and *r* = 0,6473** in 2018).

The highest number of total and full seeds per head in both analysed years was showed by the hybrid HM14, followed by HM3, HM4, HM1, HM9 and HM18, the last three genotypes also indicating the higher value of weight of seeds per head. In 2018 growing season, the maximum was found in HM15 (Figure 6).

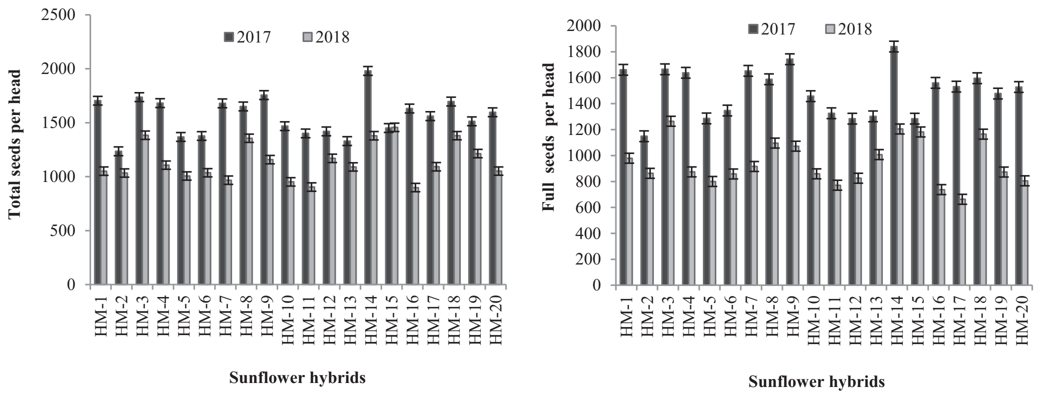


Figure 6. Number of total (a) and full (b) seeds per head of twenty sunflower hybrids during the two experimental periods (2017 and 2018)

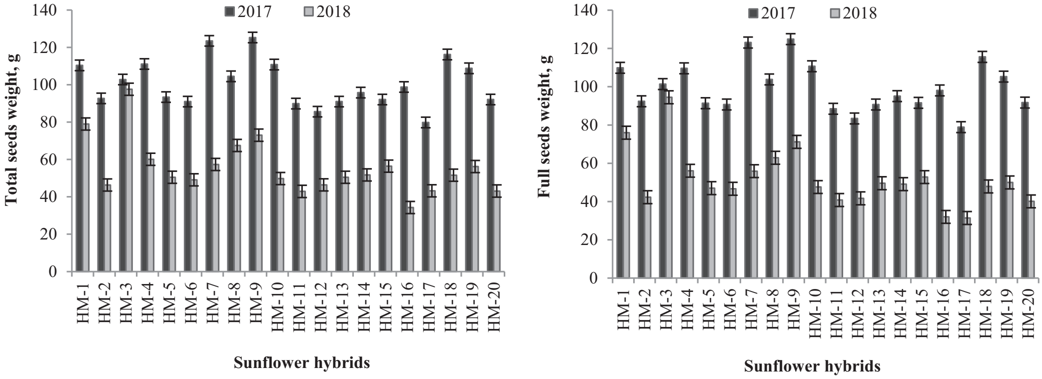
The lowest number of achenes per head and one of the minimal values of seeds weight were characteristic for HM2 in 2017 and HM16 in 2018 (Figure 7).

Number and weight of seeds per head were significantly lower in 2018 comparative to 2017. This fact may be due to lower water supply and correspond with the findings of Stanojević and Dragović (1988) which observed significant reductions in flower number and, respectively, seeds number in the presence of water deficit at budding or flowering stage.

In both experimental years, the analysis of the Pearson correlation coefficients has indicated very high positive correlations between these traits and seed yield (kg/ha). Thus, high correlations existed between the number of full seeds (0.6145* in 2017 and 0.5397* in 2018, respectively), as well as weight of total (0.5387** and 0.6117*) and full seeds (0.5285** and 0.6685*) and yield. Similarly, high positive correlations among mentioned traits and yield per plant have been reported by Hladni et al. (2010). Goksoy and Turan (2007) observed a very high positive correlation (*r* = 0.890**) between seed yield and number of seeds per head.

According to Škorić (1989) 1000-seed weight has an indirect effect on the quality of the seeds to be produced as well seed yield per unit area. Obtained data has shown that the average value of 1000 seeds weight of all hybrids was 69.12 g in 2017 and 52.4 g in 2018, being by 24%

lower. High number of 1000 seeds weight in both years was noticed in HM7 (84.9 g and 59.48 g, respectively), HM1 (83.7 g and 76.45 g, respectively), HM3 (78.5 g and 70.04 g, respectively) and HM4 (77.6 g and 63.21 g, respectively), while the lowest value of 1000 seeds weight (50.7 g) was noticed in HM11 in 2017 and HM18 (0.39 g) in 2018 (Figure 8).
The results are in agreement with those reported by Ibrahim (2012).



a. b.

Figure 7. Weight of total (a) and full (b) seeds per head of twenty sunflower hybrids during the two experimental periods (2017 and 2018)

The value of 1000 seeds weight varied significantly among the hybrids and years of study, being greatly influenced by the last factor. Thus, in the second year, this parameter decreased by 38-42% in the case of HM16 and HM18, significantly affecting the seed yields (by 46.4% and 21.5%, respectively). Data is in agreement with the findings of Radic et al. (2013), according to which 1000-seed weight depends first and foremost on the year of study and the hybrid.

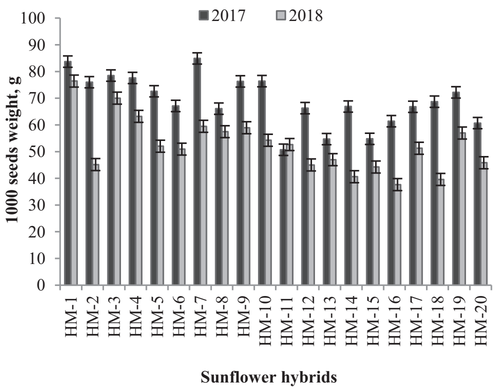


Figure 8. 1000 seeds weight (g) of twenty sunflower hybrids during two experimental periods (2017 and 2018)

Similar with the results reported by numerous authors (Hladni et al., 2004; 2010; Behradfar et al., 2009; Kaya, 2015) this parameter highly correlates with seed yield. Thus, the Pearson correlation analysis showed significant ($p<0.01$ or 0.05) positive correlation (r=0.7482* in 2017 and 0.4997* in 2018) of 1000 seeds weight with seeds yield.
The seed yield in 2017 growing season varied between 2139.1-3152.9 kg/ha, with an average value of 2743.5 kg/ha (Figure 9).

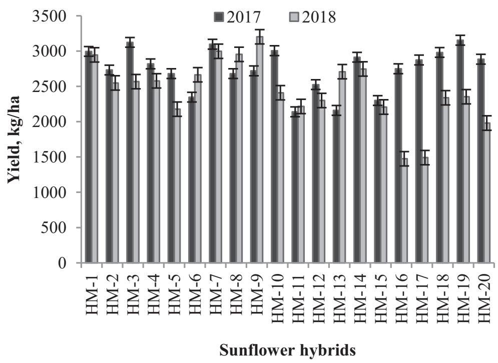


Figure 9. Seeds yield (kg/ha) of twenty sunflower hybrids during two experimental periods (2017 and 2018)

The highest value for seed yield was established in HM19 (3152.9 kg/ha) followed by HM3 (3122.7 kg/ha) and other hybrids,

such as HM7, HM10, HM14 and HM1 with the value ranged among 2912.5-3096.9 kg/ha.
The minimal yield observed at hybrid HM11 (2139.1 kg/ha).
In 2018 seed yield was lower for the majority of hybrids, with values between 1474-3202 kg/ha, with average value of 2442.5 kg/ha. The sunflower hybrid HM9 showed the maximum yield, even 17% higher than in previous year. The most affected hybrids were HM16 and HM1, which produced the minimal yield values, by around 50% lower than in 2017 growing season. In the majority of cases the diminution of seeds yield was insignificant. Thus, the hybrid combinations HM1, HM2, HM4, HM6-HM9 and HM11-HM15 showed stable yield values.

CONCLUSIONS

The results of the study of twenty sunflower hybrids grown under Moldovan environmental conditions, during two years, indicated that the growth and productivity of combinations depended both on the year of observation and the genotype. A stable seeds yield was obtained in HM1, HM2, HM4, HM6-HM9 and HM11-HM15 sunflower hybrids. Mentioned hybrids shown similar values in both years, even the 2018 growing season is characterized by a significant lower rainfall quantity. The number of leaves per plant was the most affected by environmental conditions trait, this finding being in agreement with the fact that plants under water deficit reduce leaf growth. Also, yield related traits, such as number and weight of seeds per head and 1000 seeds weight, were significantly influenced by the unfavourable environmental conditions. Thus, in the second year, these parameters have been decreased by 21-45% for all analysed hybrids.
Determination of relationships between seed yield and some morpho-physiological parameters, such as plant height, head diameter, leaf number, number and weight of total and full seeds per head and 1000 seeds showed high positive correlations between some of traits. Analyses of Pearson correlation coefficients indicated that the number and weight of total and full seeds per head and the weight of 1000 seeds had a high positive correlation with seed yield per surface unite. Also, a positive but low correlation with seed yield was found related the head diameter. In both years a significant positive correlation were established between plant height and number of leaves.

ACKNOWLEDGEMENTS

This study was supported by the research project of bilateral cooperation from Moldova and Romania *"Evaluation of some sunflower hybrids to water and heat stress resistance in Romania and Republic of Moldova"*.

REFERENCES

Balalić, I., Crnobarac, J., Joci,ć S., Miklič, V., Radić, V., Dušanić, N. (2016). Variability of head diameter in sunflower hybrids depending on planting date. *Genetika*, *48*(3), 983–990.

Behradfar, A., Gorttapeh, H., Zardashty, M.R., Talat, F. (2009). Evaluation correlated traits for seed and oil yielded in sunflower (*Helianthus annuus* L.) through Path analysis in under conditions relay cropping. *Research Journal of Biological Sciences, 4*(1), 82–85.

Černý, I., Veverková, A., Kovár, M., Mátyá, M. (2013). The variability of sunflower (*Helianthus annuus* L.) yield and quality influenced by the weather conditions. *Acta Universitatis Agriculturae et Silviculturae, Mendelianae Brunensis*, *LX*I(3), 595–600.

Đukić, V., Balešević-Tubić, S., Đorđević, V., Tatić, M., Dozet, G., Jaćimović, G. and Petrović, K. (2011). Yield and quality of soybean seeds as affected by growing conditions. *Journal on Field and Vegetable Crops Research*, *48*(1), 137–142.

FAO (2010). Agribusiness handbook. Sunflower Crude and Refined Oil, www.fao.org, Accessed on 11.02.2019

FAOSTAT database (2015). http://www.fao.org/faostat, Accessed on 29.10.2017.

Goksoy Abdurrahim, T., Turan, Z.M. (2007) Correlations and path analysis of yield components in synthetic varieties of sunflower (*Helianthus annuus* L.). *Acta Agronomica Hungarica*, *55*(3), 339–345.

Gvozdenović, S., Joksimović, J., Škorić, D. (2005). Gene effect and combining abilities for plant height and head diameter in sunflower. *Genetika-Belgrade*, *37*(1), 57–64.

Hladni, N., Jocić, S., Miklič, V., Mijić, A., Saftić-Panković, D. (2008). Direct and indirect effects of morphophysiological traits on seed yield of sunflower (*Helianthus annuus* L.). *Proc. 17th Int. Sunfl. Conf.. Cordoba. Spain, 1,* 393–397.

Hladni, N., Škorić, D., Kraljević-Balalić, M., Ivanović, M., Sakač, Z., and Jovanović, D. (2004). Correlation of yield components and seed yield per plant in sunflower (*Helianthus annuus* L.). *Proc. 16th Intl. Sunflower Conf., Fargo, ND, USA, 29 August-4*

September 004. Intl. Sunflower Assoc., Paris, France. II. 491–496.

Hladni, N., Miklič, V., Jocić, S., Kraljević-Balalić, M., Škorić, D. (2014). Mode of inheritance and combining ability for plant height and head diameter in sunflower (*Helianthus annuus* L.). *Genetika*, *46*(1), 159–168.

Hladni, N., Jocić, S., Miklič, V., Mijić, A., Saftić-Panković, D., Škorić. D. (2010). Effect of morphological and physiological traits on seed yield and oil content in sunflower. *Helia*, *33*(53), 101–116.

Hu, F.B., Manson, J.E., Willett, W.C. (2001). Types of dietary fat and risk of coronary heart disease: A critical review. *Journal of the American College of Nutrition, 20*, 5–19.

Ibrahim, H.M. (2012). Response of some sunflower hybrids to different levels of plant density. *APCBEE Procedia*, *4*, 175–182.

Iqrasan, A., Qayyum, S.U., Khan, S.A., Khan, A., Mehmood, Y., Bibi, A., Sher, Khan, H., Jenks, M.A. (2017). Sunflower (*Helianthus annuus*) hybrids evaluation for oil quality and yield attributes under spring planting conditions of Haripur. Pakistan. *Planta daninha, 35*. e017161596.

Kaleem, S., Fayyaz-ul-Hassan, Muhammad, F., Muhammad, R., Munir, A. (2010). Physio-morphic traits as influenced by seasonal variation in sunflower: A review. *International Journal of Agriculture and Biology*, *12*(3), 468–473.

Kandakov, A., Havrland, B., Ojog, C., Ivanova, T. (2012). Sunflower market assessment in the Republic of Moldova. *Engineering for Rural Development Jelgava,* 128–133.

Kaya, Y., Evci, G., Durak, S., Pekcan, V., Gucer, T. (2009). Yield components affecting seed yield and their relationships in sunflower (*Helianthus annuus* L.). *Pakistan Journal of Botany*, *41*(5), 2261–2269.

Kaya, Y. (2015). *Sunflower.* In S.K. Gupta (Ed.), *Breeding oilseed crops for sustainable production: Opportunities and constraints,* San Diego, CA: Academic Press, 55–88.

Khalifa, H.E., Awad, H.A., (1997). Sunflower yield and water consumptive use as affected by planting methods and skipping irrigation at different growth stages. *Journal of Agricultural Science, Mansoura University*, *22*, 2101–2107.

Khan, H., Safdar, A., Ijaz, A., Ihsanullah, K., Shujaat, H. Bashir, A.K., Muhammad, S. (2018). Agronomic and qualitative evaluation of different local sunflower hybrids. *Pakistan Journal of Agricultural Research*, *31*(1), 69–78.

Killi, F., Tekeli, F. (2016). Seed yield and some yield components of sunflower (*Helianthus annuus* L.) genotypes in Kahramanmaras (Turkey) conditions. *Journal of Scientific and Engineering Research, 3*(4), 346–349.

Kocjan Ačko, D. (2008). Some economically important properties of sunflower cultivars (*Helianthus annuus* L.) in the field trials performed at Biotechnical Faculty. *Acta Agriculturae Slovenica*, *91*, 47–58.

Liović, I., Kovačević, V., Krizmanić, M., Mijić, A., Šimić, B. (2006). Precipitation influence on edible oil production from sunflower crop in Croatia. *Cereal Research Communications*, *34*, 573–576.

Marinkovic, R. (1992). Path coefficient analysis of some yield components of sunflower (*Helianthus annuus* L.). *Euphytica, 60*, 201–205.

Mijić, A., Liović, I., Kovačević, V., Pepó, P. (2012). Impact of weather conditions on variability in sunflower yield over years in eastern parts of Croatia and Hungary, *Acta Agronomica Hungarica*, *60*(4), 397–405.

Moisa, F., Smit, G.A. (2014). Performance of the sunflower hybrids in the north-west of Romania, *Analele INCDA Fundulea*, *82*, 149–154.

Moroz, V., Stratan, A., Ignat, An., Lucasenco, E. (2015). Country Report: Republic of Moldova, developed in the framework of the FP7 International Project AGRICISTRADE, http://www.agricistrade.eu, Accessed on 29.01.2019

Mrdja, J., Crnobarac, J., Radić, V., Miklič, V. (2012). Sunflower seed quality and yield in relation to environmental conditions of production region. *Helia, 35*(57), 123–134.

Papatheohari, Y., Travlos, I.S., Papastylianou, P., Argyrokastritis, I.G., Dimitrios, B.J. (2016). Growth and yield of three sunflower hybrids cultivated for two years under Mediterranean conditions. *Emirates Journal of Food and Agriculture*, *28*(2), 136–142.

Pereyra-Irujo, G.A., Velázquez, L., Lechner, L., Aguirrezábal, L.A.N. (2008). Genetic variability for leaf growth rate and duration under water deficit in sunflower: analysis of responses at cell, organ, and plant level. *Journal of Experimental Botany*, *59*(8), 2221–2232.

Radić, V., Mrđa, J., Jocković, M., Čanak, P., Dimitrijević, A., Jocić, S. (2013). Sunflower 1000-seed weight as affected by year and genotype, *Journal on Field and Vegetable Crops Research*, *50*(1), 1–7.

Ruzdik, N.M., Karov, I., Mitrev, S., Gjorgjieva, B., Kovacevik, B., Kostadinovska, E. (2015). Evaluation of sunflower (*Helianthus annuus* L.) hybrids using multivariate statistical analysis. *Helia, 38*(63), 175–187.

Škorić, D. (2012). Sunflower breeding. *Sunflower Genetics and Breeding.* (Eds Škorić, D., Seiler, G. J., Zhao, L., Jan, C. C., Miller, J. F. & Charlet, L. D.), Novi Sad, Serbia: Serbian Academy of Science and Arts., 165–354.

Škorić, D. (1989). *Suncokret*. Beograd. Nolit: 636.

Stanojević, D., Dragović, S. (1988). Water balance in vertisol at different stages of sunflower development. *Proc. 12th Int. Sunflower Conference, Novi Sad, Yugoslavia, 1*, 451.

Statistical databank, http://statbank.statistica.md, Accessed on 23.01.2019.

Sunflowers Excel in Ukraine, Romania, Bulgaria and Moldova in 2017, http://www.nuseed.com, Accessed on 10.03.2019.

Ullah, S., Ahmad, S., Khan, R., Sohail, A. (2018). Assessment of different sunflower genotypes under agro-climatic conditions of district Malakand Khyber-Pakhtunkhwa. *Current Trends in Biomedical Engineering & Biosciences, 11*(2), 1–6.

Velasco, L., Fernández-Cuesta, Á., Fernández-Martínez, J.M. (2014). New sunflower seeds with high contents of phytosterols. *OCL, 21*(6), D604.

Yegappan, T.M., Paton, D., Gates, C.T., Muller, W. (1982). Water stress in sunflower (response of cyptla size). *Annuals of Botany*, *49*, 63–68.

Zagoul'ko, A.V., Kvashin, A.A., Malyuga, N.G. (2011). *Sunflower. Biology and agrotechnology of cultivation in the south of Russia*, Krasnodar. 291 pp.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EFFICACY OF HERBICIDES AND HERBICIDE COMBINATIONS AT MAIZE (*Zea mays* L.)

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Abstract

The research was conducted during 2012 - 2014. Under investigation was cycloxydim tolerant maize hybrid Ultrafox duo (Zea mays L.). Factor A included no treated check and 3 soil-applied herbicides - Adengo 465 SC, Wing P and Lumax 538 SC. Factor B included no treated check and 5 foliar-applied herbicides - Stellar 210 SL, Principal plus, Ventum WG, Monsun active and Laudis OD. In addition to these variants by conventional technology for maize growing, in the experience is included one variant by Duo system technology. It includes soil-applied herbicide Merlin flex 480 SC and tank mixture of antigraminaceous herbicide Focus ultra + antibroadleaved herbicide Kalam. Combinations between soil-applied herbicides Adengo, Wing and Lumax and vegetation-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis by conventional technology, have very high efficacy against annual graminaceous weeds and against Sorghum halepense Pers. from seeds and rhizomes, medium efficacy against Agropyrum repens L. and not efficacy against Cynodon dactylon Pers. Herbicide combination between soil-applied herbicide Merlin flex and tank herbicide mixture Focus ultra + Kalam by Duo system technology have very high efficacy against perennial weeds Cynodon dactylon Pers. and Agropyrum repens L. Herbicide Wing is inefficacy against of ClearField and ExpressSun sunflower self-sown plants in maize crops. Herbicide combination of soil-applied herbicide Merlin flex with tank mixture Focus ultra + Kalam by Duo system technology lead to obtaining of high grain yield. High yields of maize grain also are obtained by herbicide combinations Lumax + Principal plus, Lumax + Laudis and Wing + Principal plus. Alone application of herbicides leads to lower yields due to must to combine soil-applied with foliar-applied herbicides for full control of weeds in maize crops.

Key words*: grain maize, herbicides, herbicide combinations, efficacy, selectivity, grain yield.*

INTRODUCTION

Weeds are one of the main limiting factors for maize production (Vancetovic et al., 2010; Dragičević et al., 2012). The main species weeding maize crops are the group of late spring weeds. When growing cool-resisting maize hybrids weed infestation is changed and serious competitors and also early spring weeds that are typical for sunflower fields (Sinzar et al., 1998; Michel, 2001; Delchev, 2018).

From an economic and ecological view point, the combination of the chemical and mechanical controls of weeds is very positive. Limiting soil tillage on mechanical weed control reduces the risk of soil erosion, especially in hilly areas and decreasing herbicide treatments reduces the risk of contamination of soil and water (Dawoud et al., 2006; Moteva & Stoyanova, 2008; Pajić et al., 2009; Stoyanova & Gospodinov, 2009; Korpanov et al., 2010).

The use of herbicides in the early stages of maize development is essential for high yield production (Kees & Lutz, 1991). In conventional technology for grain maize growing there are unsolved problems of control of some perennial graminaceous weeds such as *Cynodon dactylon* Pers. and *Agropyron repens* L., which necessitated the introduction of the new Duo system technology for maize growing (Jovovic et al., 1999; Asadi et al., 2009).

The purpose of this investigation was to establish the efficacy and selectivity of some herbicides and herbicide combinations on the grain maize by influence of different meteorological conditions.

MATERIALS AND METHODS

The research was conducted during 2012-2014 with the cycloxydim tolerant maize hybrid Ultrafox duo (*Zea mays* L.). It was carried out a two factors field experiment as a block method in 4 repetitions, on a 20 m^2 harvesting area, on pellic vertisol soil type, after durum wheat predecessor. Factor A included no treated check and 3 soil-applied herbicides - Adengo 465 SC, Wing P and Lumax. Factor B included no treated check and 5 foliar-applied herbicides - Stellar 210 SL, Principal plus, Ventum WG (foramsulfuron + iodosulfuron) - 150 g/ha, Monsun active and Laudis OD. In addition to these variants by conventional technology for maize growing, in the experience is included one variant by Duo system technology. It includes soil-applied herbicide Merlin flex 480 SC and tank mixture of antigraminaceous herbicide Focus ultra + anti-broadleaved herbicide Kalam. The active substances and doses of the investigated herbicides are given in Table 1.

Table 1. Investigated variants

№	Variants	Active substance	Doses
		Conventional technology	
		After sowing, before emergence	
1	Check	-	-
2	Adengo 465 SC	isoxaflutol + tiencarbazon	440 ml/ha
3	Wing P	pendimethalin + dimethenamid	4 l/ha
4	Lumax 538 SC	S-metolachlor + terbuthylazine + mesotrione	4 l/ha
		5 - 7 leaf stage	
1	Check	-	-
2	Stellar 210 SL	topramezon + dicamba	1 l/ha
3	Principal plus	nicosulfuron + rimsulfuron + dicamba	380 g/ha
4	Ventum WG	foramsulfuron + iodosulfuron	150 g/ha
5	Monsun active OD	foramsulfuron + tiencarbazon	1.5 l/ha
6	Laudis OD	Tembotrione	2 l/ha
		Duo system technology	
		After sowing, before emergence	
1	Merlin flex 480 SC	Isoxaflutole	420 g/ha
		5 - 7 leaf stage	
2	Focus ultra + Kalam	cycloxydim tritosulfuron + dicamba	2 l/ha 300 g/ha

Herbicides Stellar 210 SL and Kalam they were used in addition with adjuvant Dash HC - 1 l/ha, herbicide Principal plus - with adjuvant Trend - 0.2% and herbicide Ventum WG - with adjuvant Mero 80 EC - 2 l/ha.

Soil-applied herbicides were treated during the period after sowing before emergence. Foliar-applied herbicides were treated during 5-7 leaf stage of the maize. All of herbicides and herbicide combinations were applied in a working solution of 200 l/ha. Due to of low adhesion of the herbicides Stellar 210 SL and Kalam they were used in addition with adjuvant Dash HC - 1 l/ha, herbicide Principal plus - with adjuvant Trend - 0.2% and herbicide Ventum WG - with adjuvant Mero 80 EC - 2 l/ha.

It was investigated efficacy and selectivity of herbicides and their tank mixtures. It was established their influence on grain yield. Efficacy of herbicides against weeds and self-sown plants of sunflower was appointed according to 100% scale of EWRS (European

Weed Research Society). Selectivity of herbicides to maize plants was followed according to the 9-rate scale of EWRS (rating 1 - without damages, rating 9 - crop is completely destroyed). The mathematical processing is done with analysis of variance method.

RESULTS AND DISCUSSIONS

Dominant weeds that determine secondary weed infestation in the experiment field are late spring annual broadleaved species *Xanthium strumarium* L., *Amaranthus retroflexus* L., *Amaranthus albus* L., *Chenopodium album* L., *Solanum nigrum* L., *Datura stramonium* L., *Polygonum aviculare* L., *Abutilon teophrasti* Medic., *Portulaca oleracea* L., in a small amount *Amaranthus blifoides* W., *Polygonum aviculare* L., *Hibiscum trionum* L., *Tribulus terrestris* L. Early spring annual broadleaved species are mainly *Sinapis arvensis* L. and *Falopia convolvulus* Leve.

Annual graminaceous weeds are represented by *Echinochloa crus-galli* L., *Panicum sanguinale* L., *Setaria viridis* Beauv., *Setaria glauca* Beauv. As single plants is established *Echinochloa coarctata* Vas. and *Setaria verticilata* Beauv.

Perennial species in experiment are broadleaved weeds, *Cirsium arvense* Scop. and *Convolvulus arvensis* L., and graminaceous weeds, *Sorghum halepense* Pers., *Cynodon dactylon* Pers. and less frequently *Agropyrum repens* L.

Self-sown plants of sunflower (*Helianthus annuus* L.) are from sunflower was grown two years ago as predecessor. In the previous year, before the maize was grown durum wheat (*Triticum durum* Desf.).

Soil-applied herbicides Adengo, Wing and Lumax applied during the period after sowing before germination are inefficacy against perennial broadleaved weeds, *Cirsium arvense* Scop. and *Convolvulus arvensis* L. (Table 2). These herbicides have very high efficacy against late spring annual broadleaved weeds - *Amaranthus retroflexus* L., *Amaranthus albus* L., *Chenopodium album* L., *Solanum nigrum* L., *Datura stramonium* L., *Abutilon teophrasti* Medic., *Portulaca oleracea* L., *Polygonum aviculare* L., *Hibiscum trionum* L. et al. Adengo and Wing are inefficacy against *Xanthium strumarium* L. only and Lumax control this weed of 80%. Wing is less effective against *Sinapis arvensis* L. and *Chenopodium album* L. - controls them, respective of 90% and 93%.

Foliar-applied herbicides Principal plus and Laudis, applied during 4-8 leaf of maize have very high efficacy against perennial broadleaved weeds *Cirsium arvense* Scop. and *Convolvulus arvensis* L. Herbicides Ventum and Monsun active control of 100% *Cirsium arvense* Scop. but their efficacy against *Convolvulus arvensis* L. is weaker - 95%. It may be explained by the later emergence of *Convolvulus arvensis* L. in which a part of its later germinated shoots remain unsprayed with herbicide solution. Herbicide Stellar have weaker efficacy against perennial broadleaved weeds - 95% against *Cirsium arvense* Scop. and 90% against *Convolvulus arvensis* L.

Foliar-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis and their combinations control successfully all annual broadleaved weeds.

Self-sown plants of ClearField and ExpressSun sunflower are controlled successfully by soil-applied herbicides Adengo and Lumax and foliar-applied herbicides Stellar, Ventum, Monsun active and Laudis. Foliar-applied herbicide Principal plus has weaker efficacy against these self-sown plant - control them of 97%, but this percent is sufficient for their efficacy control. Soil-applied herbicide Wing is inefficacy against these self-sown plants.

The initial effect of herbicide Monsun active against self-sown plants of ClearField and ExpressSun sunflower is weaker. They die slower compared with other herbicides and herbicide combinations. Part sunflower self-sown plants not die immediately after treatment with this herbicide. Usually these are those volunteer who are in rows between the maize plants. They absorb less herbicide solution because they are partially covered by leaves of maize. These self-sown plants in all cases are highly depressed in their development. They remain under the maize plants, some of them later died, while other

Table 2. Efficacy of some herbicides and herbicide combinations against broadleaved weeds and self-sown plants at maize according to the 100% visual scale of EWRS (mean 2012-2014)

Herbicides		Weeds							
Soil-applied	Foliar-applied	*Cirsium arvense*	*Convolvulus arvensis*	*Xanthium strumarium*	*Amaranthus retroflexus*	*Chenopodium album*	*Solanum nigrum*	*Sinapis arvense*	*Helianthus annuus* *
Conventional technology									
-	-	0	0	0	0	0	0	0	0
	Stellar – 1 l/ha	95	90	100	100	100	100	100	100
	Principal plus – 380 g/ha	100	100	100	100	100	100	100	97
	Ventum – 150 g/ha	100	95	100	100	100	100	100	100
	Monsun active – 1.5 l/ha	100	95	100	100	100	100	100	100
	Laudis – 2 l/ha	100	100	100	100	100	100	100	100
Adengo – 440 ml/ha	-	0	0	0	100	100	100	100	100
	Stellar – 1 l/ha	95	90	100	100	100	100	100	100
	Principal plus – 380 g/ha	100	100	100	100	100	100	100	100
	Ventum – 150 g/ha	100	95	100	100	100	100	100	100
	Monsun active – 1.5 l/ha	100	95	100	100	100	100	100	100
	Laudis – 2 l/ha	100	100	100	100	100	100	100	100
Wing – 4 l/ha	-	0	0	0	100	93	100	90	0
	Stellar – 1 l/ha	95	90	100	100	100	100	100	100
	Principal plus – 380 g/ha	100	100	100	100	100	100	100	100
	Ventum – 150 g/ha	100	95	100	100	100	100	100	100
	Monsun active – 1.5 l/ha	100	95	100	100	100	100	100	100
	Laudis – 2 l/ha	100	100	100	100	100	100	100	100
Lumax – 4 l/ha	-	0	0	80	100	100	100	100	100
	Stellar – 1 l/ha	95	90	100	100	100	100	100	100
	Principal plus – 380 g/ha	100	100	100	100	100	100	100	100
	Ventum – 150 g/ha	100	95	100	100	100	100	100	100
	Monsun active – 1.5 l/ha	100	95	100	100	100	100	100	100
	Laudis – 2 l/ha	100	100	100	100	100	100	100	100
Duo system technology									
Merlin flex – 420 g/ha	Focus ultra – 2 l/ha + Kalam – 300 g/ha	100	100	100	100	100	100	100	100

* - self-sown plants of ClearField and ExpressSun sunflower

ones remained alive, but poorly developed and practically no influenced on the value of maize grain yield

Herbicide combination by Duo system technology between soil-applied herbicide Merlin flex applied during after sowing before emergence, and tank herbicide mixture Focus ultra + Kalam applied during 5-7 leaf stage, control successfully of 100% all annual and perennial broadleaved weeds in fields of cycloxydim tolerant maize. This combination has very high efficacy against self-sown plants of ClearField and ExpressSun sunflower.

Soil-applied herbicides Adengo, Wing and Lumax have very high efficacy against all existing in the trial annual graminaceous weeds - *Echinochloa crus-galli* L., *Panicum sanguinale* L., *Setaria viridis* Beauv., *Setaria glauca* Beauv. (Table 3).

Table 3. Efficacy of some herbicides and herbicide combinations against graminaceous weeds at maize according to the 100% visual scale of EWRS and selectivity according to the 9-rate scale of EWRS (mean 2012-2014)

Herbicides		Weeds							
Soil-applied	Foliar-applied	*Sorgum halepense*	*Cynodon dactylon*	*Agropirum repens*	*Echinochloa crus-gali*	*Setaria viridis*	*Setaria glauca*	*Digitaria sangvinale*	Selectivity
Conventional technology									
-	-	0	0	0	0	0	0	0	1
-	Stellar – 1 l/ha	100*	0	0	100	100	100	100	1
-	Principal plus – 380 g/ha	100	0	85	100	100	100	100	1
-	Ventum – 150 g/ha	100	0	85	90	98	100	90	1
-	Monsun active – 1.5 l/ha	100	0	75	95	98	100	95	1
-	Laudis – 2 l/ha	82	0	70	100	100	100	100	1
Adengo – 440 ml/ha	-	100*	0	0	100	100	100	100	1
Adengo – 440 ml/ha	Stellar – 1 l/ha	100*	0	0	100	100	100	100	1
Adengo – 440 ml/ha	Principal plus – 380 g/ha	100	0	85	100	100	100	100	1
Adengo – 440 ml/ha	Ventum – 150 g/ha	100	0	85	100	100	100	100	1
Adengo – 440 ml/ha	Monsun active – 1.5 l/ha	100	0	75	100	100	100	100	1
Adengo – 440 ml/ha	Laudis – 2 l/ha	82	0	70	100	100	100	100	1
Wing – 4 l/ha	-	96*	0	0	100	100	100	100	1
Wing – 4 l/ha	Stellar – 1 l/ha	100*	0	0	100	100	100	100	1
Wing – 4 l/ha	Principal plus – 380 g/ha	100	0	85	100	100	100	100	1
Wing – 4 l/ha	Ventum – 150 g/ha	100	0	85	100	100	100	100	1
Wing – 4 l/ha	Monsun active – 1.5 l/ha	100	0	75	100	100	100	100	1
Wing – 4 l/ha	Laudis – 2 l/ha	82	0	70	100	100	100	100	1
Lumax – 4 l/ha	-	80*	0	0	100	100	100	100	1
Lumax – 4 l/ha	Stellar – 1 l/ha	100*	0	0	100	100	100	100	1
Lumax – 4 l/ha	Principal plus – 380 g/ha	100	0	85	100	100	100	100	1
Lumax – 4 l/ha	Ventum – 150 g/ha	100	0	85	100	100	100	100	1
Lumax – 4 l/ha	Monsun active – 1.5 l/ha	100	0	75	100	100	100	100	1
Lumax – 4 l/ha	Laudis – 2 l/ha	82	0	70	100	100	100	100	1
Duo system technology									
Merlin flex – 420 g/ha	Focus ultra – 2 l/ha + Kalam – 300 g/ha	100	100	100	100	100	100	100	1

* - against *Sorgum helepense* Pers. from seeds only

These three herbicides are inefficacy against perennial graminaceous weeds - *Sorghum helepense* Pers., *Cynodon dactylon* Pers. and *Agropyrum repens* L. Adengo and Wing control only *Sorghum halepense* Pers. by seeds, respective of 100% and 96%. Lumax has weaker efficacy against this weed - 80%.

Foliar-applied herbicides Ventum and Monsun active have weaker efficacy against annual graminaceous weeds - *Echinochloa crus-galli* L. and *Panicum sanguinale* L. Ventum controls these two weeds of 90% and Monsun active - 95%. Herbicides Stellar, Principal plus and Laudis have very high efficacy against the annual graminaceous weeds. Herbicide Stellar controls only *Sorghum halepense* Pers. by seeds, but does not control perennial graminaceous weeds.

Combinations of foliar-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis with soil-applied herbicides Adengo, Wing and Lumax control successfully all annual graminaceous weeds.

Herbicide combinations of Principal plus, Ventum and Monsun active with soil-applied herbicides Adengo, Wing and Lumax have very high efficacy against *Sorghum halepense* Pers. by seeds and rhizomes, middle efficacy against *Agropyrum repens* L. and inefficacy *Cynodon dactylon* Pers. Combinations of Laudis with Adengo, Wing and Lumax are weaker efficacy also against *Sorghum halepense* Pers. by rhizomes. Combinations of Stellar with the three soil-applied herbicides control only *Sorghum halepense* Pers. by seeds, but do not control *Sorghum halepense* Pers. by rhizomes; *Cynodon dactylon* Pers. and *Agropyrum repens* L.

Herbicide combination of soil-applied herbicide Merlin flex with tank herbicide mixture Focus ultra + Kalam by Duo system technology has 100% efficacy against *Sorghum halepense* Pers. by seeds and rhizomes, as well as against all annual graminaceous weeds - *Echinochloa crus-galli* L., *Panicum sanguinale* L., *Setaria viridis* Beauv., *Setaria glauca* Beauv. This combination has also very high efficacy against perennial graminaceous weeds *Cynodon dactylon* Pers. and *Agropyrum repens* L. This is due to the participation of herbicide Focus ultra in this combination.

Perennial graminaceous weeds *Cynodon dactylon* Pers. and *Agropyrum repens* L. practically cannot be controlled at the conventional technology. Duo system technology is the only way to chemical control of *Cynodon dactylon* Pers. and *Agropyrum repens* L. in maize crops.

Soil-applied herbicides Adengo, Wing and Lumax, foliar-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis by conventional technology, as well as their combinations have very high selectivity for maize - rating 1 by scale of EWRS (Table 3).

Herbicide combination of soil-applied herbicide Merlin flex with tank herbicide mixture Focus ultra + Kalam has also very high selectivity for cycloxydim tolerant maize (CTM) which are grown by Duo system technology - rating 1 by scale of EWRS.

Data about the influence of the investigated herbicides and herbicide combinations on maize grain yields (Table 4) show that the lowest yield was obtained by the untreated check. The alone use of soil-applied herbicides Adengo, Wing and Lumax increases grain yield average for the period from 114.5% to 116.2%.

Alone use of vegetation-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis increased grain yield from 116.1% to 118.2%.

The herbicide combinations between soil-applied herbicides Adengo, Wing and Lumax by the one hand and vegetation-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis by the other hand, lead to a bigger increase in yield compared with alone use of these herbicides during the three years the investigation. Herbicide combinations provide full control of all annual and perennial weeds in maize crops except *Cynodon dactylon* Pers. and *Agropyron repens* L. The highest grain yield by conventional technology is obtained by the herbicide combination Lumax + Principal plus - 125.7% over the no treated check, followed by Lumax + Laudis - 124.9% and Wing + Principal plus - 124.6%.

Table 4. Influence of some herbicide combinations on maize grain yield (2012-2014)

Herbicides		2012		2013		2014		Mean	
Soil-applied	Vegetation-applied	kg/ha	%	kg/ha	%	kg/ha	%	kg/ha	%
Conventional technology									
-	-	3510	100	4688	100	5555	100	4584	100
	Stellar – 1 l/ha	4037	115.0	5429	115.8	6499	117.0	5322	116.1
	Principal plus – 380 g/ha	4114	117.2	5532	118.0	6605	118.9	5417	118.2
	Ventum – 150 g/ha	4103	116.9	5485	117.0	6583	118.5	5390	117.6
	Monsun active – 1.5 l/ha	4082	116.3	5466	116.6	6555	118.0	5368	117.1
	Laudis – 2 l/ha	4068	115.9	5466	116.6	6583	118.5	5372	117.2
Adengo – 440 ml/ha	-	4012	114.3	5335	113.8	6499	117.0	5282	115.4
	Stellar – 1 l/ha	4233	120.6	5626	120.0	6788	122.2	5549	121.1
	Principal plus – 380 g/ha	4342	123.7	5757	122.8	6960	125.3	5686	124.0
	Ventum – 150 g/ha	4258	121.3	5644	120.4	6855	123.4	5586	121.9
	Monsun active – 1.5 l/ha	4282	122.0	5682	121.2	6888	124.0	5617	122.5
	Laudis – 2 l/ha	4310	122.8	5719	122.0	6927	124.7	5652	123.3
Wing – 4 l/ha	-	3994	113.8	5307	113.2	6444	116.0	5248	114.5
	Stellar – 1 l/ha	4247	121.0	5644	120.4	6760	121.7	5550	121.1
	Principal plus – 380 g/ha	4370	124.5	5785	123.4	6977	125.6	5711	124.6
	Ventum – 150 g/ha	4324	123.2	5766	123.0	6888	124.0	5659	123.5
	Monsun active – 1.5 l/ha	4293	122.3	5696	121.5	6833	123.0	5607	122.3
	Laudis – 2 l/ha	4352	124.0	5719	122.0	6922	124.6	5664	123.6
Lumax – 4 l/ha	-	4037	115.0	5419	115.6	6527	117.5	5328	116.2
	Stellar – 1 l/ha	4324	123.2	5715	121.9	6855	123.4	5631	122.8
	Principal plus – 380 g/ha	4419	125.9	5827	124.3	7038	126.7	5761	125.7
	Ventum – 150 g/ha	4370	124.5	5766	123.0	6949	125.1	5695	124.2
	Monsun active – 1.5 l/ha	4352	124.0	5752	122.7	6899	124.2	5668	123.6
	Laudis – 2 l/ha	4388	125.0	5785	123.4	7000	126.0	5724	124.9
Duo system technology									
Merlin flex – 420 g/ha	Focus ultra – 2 l/ha + Kalam – 300 g/ha	4461	127.1	5893	125.7	4461	127.1	5822	127.0
	LSD 5 %	221	6.2	236	5.0	240	4.3		
	LSD 1 %	260	7.4	271	5.8	289	5.2		
	LSD 0.1 %	286	8.1	305	6.5	333	6.0		

Combination of soil-applied herbicide Merlin flex with tank herbicide mixture Focus ultra + Kalam by Duo system technology, increases the most grain yield - 127.0% compared no treated check. This is due to the complete control of all broadleaved and graminaceous weeds, including *Cynodon dactylon* Pers. and *Agropyron repens* L.

CONCLUSIONS

Combinations between soil-applied herbicides Adengo, Wing and Lumax and vegetation-applied herbicides Stellar, Principal plus, Ventum, Monsun active and Laudis by conventional technology, have very high efficacy against annual graminaceous weeds and against *Sorghum helepense* Pers. from seeds and rhizomes, medium efficacy against *Agropyrum repens* L. and not efficacy against *Cynodon dactylon* Pers.

Herbicide combination between soil-applied herbicide Merlin flex and tank herbicide mixture Focus ultra + Kalam by Duo system technology have very high efficacy against perennial weeds *Cynodon dactylon* Pers. and *Agropyrum repens* L.

Herbicide Wing is inefficacy against of ClearField and ExpressSun sunflower self-sown plants in maize crops.

Herbicide combination of soil-applied herbicide Merlin flex with tank mixture Focus ultra + Kalam by Duo system technology lead to obtaining of high grain yield.

High yields of maize grain also are obtained by herbicide combinations Lumax + Principal plus, Lumax + Laudis and Wing + Principal plus.

Alone application of herbicides leads to lower yields due to must to combine soil-applied with foliar-applied herbicides for full control of weeds in maize crops.

REFERENCES

Asadi, H., Abbasi, A., Seyyedi N., Heydari, N., Abbasi, F., Moayyeri, M., Lotf A., Mazraeh, S., Absalan, Sh., Shadid, K., Shomu, F., Farahani, H., Nuri, Q., Qorbani, M. (2009). Study of effects of economic factors on water use efficiency in irrigated cereals under farmers' condition in areas of lower KRB in Khuzestan province. *Seed and Plant Improvement,* 60–69.

Dawoud, D.A., Mubarak, H.A., Abdel-Aaziz, E., Hamada, A.A., Babiker, A.E. (2006). Evaluation of guardian 84% EC (acetochlor) and atrazine for preemergence weed control in maize. *Proceedings: The 74th meeting of the National Pests and Diseases Committee*, *Wad Medani (Sudan)*, 143–153.

Delchev, Gr. (2018). *Chemical control of weeds and self-sown plants in eight field crops.* Monograph, ISBN: 978-613-7-43367-6, LAP LAMBERT Academic Publishing, Saarbrücken, Germany, 397.

Dragičević, V., Simić, M., Sečanski, M., Cvijanović, G., Nišavić, A. (2012). Study of the susceptibility of maize lines to some sulfonylurea herbicides. *Genetika*, *44*(2), 355–366.

Jovovic, Z., Mitrovic, D., Biberdzic, M. (1999). The influence of applied herbicides on the yield of maize grain (ZPTC 109). *Poljoprivreda i Sumarstvo*, *45*(1-2), 91–99.

Kees, H., Lutz, A. (1991). About the problems of triazine resistant weeds in maize and horticultural crops. *Gesunde Pflanzen*, *43*(7), 216–220.

Korpanov, R., Tereshchuk, V., Lapkovskaya, T., Soroka, L., Soroka, S., Kolesnik, S., Stashkevich, A. (2010). Herbicide Ltd. 'Frandesa' in maize, spring barley and after harvest predecessor. *Agriculture Plant Protection: Scientific Journal*, *3*, 47–50.

Michel, P. (2001). What's new at COLUMA (2001). Herbicides for cereals, corn, sorghum, sunflower and soya, reduced doses, additives and compounds. *Phytoma*. *La Défense des Végétaux*, *544*, 20–22.

Moteva, M., Stoyanova, A. (2008). FAO"s Ks coefficients for grain maize in two regions of southern Bulgaria. *Plant Science*, *4*, 328–334.

Pajić, M., Ercegović, Đ., Raičević, D., Oljača, M., Vukić, Đ., Gligorević, K., Radojević, R., Dumanović, Z., Dragičević, V. (2009). Effects of using different soil tillage systems and treatments of herbicides in production of maize. *Traktori i Pogonske Mašine, 14*(4), 70–76.

Sinzar, B., Stefanovic, L., Stanojevic, M. (1998). Changes in weed flora and maize vegetation after several years of herbicide use. *Pesticidi*, *13*(2), 119–130.

Stoyanova A., Gospodinov, I. (2009). Biophysical coefficients K and Z of grain maize. *Proceeding of Scientific Papers at the International Scientific Conference, 4-5 June, Stara Zagora*, *2*, 1–4.

Vancetovic, J., Mladenovic-Drinic, S., Babic, M., Ignjatovic-Micic, D., Andjelkovic, V. (2010). Maize gene bank Scollections as potentially valuable breeding materials. *Genetika*, *42*(1), 9–21.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESEARCH REGARDING INFLUENCE OF FERTILIZATION ON SUNFLOWER CROPS IN THE SOUTHERN PART OF ROMANIA

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Abstract

Sunflower (Helianthus annuus L.) is the most important oil crop in Romania. Fertilization of sunflower crops has as a system a complex basic fertilization (NPK or NPK+S+Mg), used in preparing the seedbed and planting, who supports the normal coverage of the critical period of nutrition and laying the foundation of total yield.
The aim of this research is to analyse the behaviour of sunflower under the influence of the degree of fertilization and also of the pedoclimatic conditions in the Oltenia Plain.
Research was performed in the year 2018, in the southern part of Romania, within the Agricultural Research and Development Station Caracal (44°06' N latitude and 24°21' E longitude) in Olt County. For setting up the experience, the NEOMA hybrid was used in 6 test variants as follows: V1 - $N_0P_0K_0$ (control); V2 - $N_{90}P_0K_0$; V3 - $N_{90}P_{60}K_0$; V4 - $N_{90}P_{60}K_{60}$; V5 - $N_{90}P_{60}K_{60}$ plus 1.5 liters/hectare of Fertigrain foliar fertilizer; V6 - $N_{120}P_{60}K_{60}$. The doses used are in kg/ha of active substance. Plant population was 57.000 plants per hectare and the determinations were performed throughout the entire vegetation period of the plant, including the harvest period where the yield value was between 2.234 kg/hectare and 4.035 kg/hectare. Climatic conditions during the vegetation period were favorable, as the average temperatures have registered values between 16.3 and 24.3°C. The total rainfalls was 378.8 mm, those been sufficient for the favorable development of the sunflower crop.

***Key words**: sunflower, fertilization, hybrid, climatic condition, yield.*

INTRODUCTION

In Romania, sunflower was introduced for oil production, around the mid-19th century in Moldavia, being the main vegetable oil producing plant (Panaitescu et al., 2010).
Compared to other crop plants, the sunflower is a big consumer of nutrients and when it comes to provisioning the soil it is very demanding in potassium, demanding in nitrogen and moderately demanding in phosphorus. Therefore, the degree and the type of fertilization have a special importance in achieving a significant but also a qualitative production.
Sunflower recovered less fertilizer than other crops because its root system with a high capacity to extract nutrients and soluble harder combinations mineral fertilizers are less valued and that seed has a small share of the plant biomass is formed (sixth or seventh of biomass epigenous), and in some areas and in some years hydric soil improperly makes mineral fertilizers to be less valued by short seeds. (Moisii et al., 2012).
Sunflower has well-developed and branched root system, and maintains open stomata under condition of high evaporation demands that usually prevail in hot arid and semi-arid regions (Abubaker et al., 2014).
Judicious and timely application of fertilizers at critical growth stages of sunflower can increase its yield considerably.
Several studies showed the positive response of sunflower to fertilization under irrigation and rain fed conditions in the central clay plains of Sudan (Elhassan et al., 2006; Lotfie & Salah, 2013).
Other studies indicated that increase in growth and yield of sunflower and other crops is dependent upon the adequate supply of nitrogen (Ali et al., 2004) and phosphorus (Weiss, 1983) and their ratio (Bi et al., 2013).
Malligawad et al. (2004) had reported that 100 kg N ha^{-1} was suitable for nitrogen fertilization of sunflower.

Nitrogen (N) and Phosphorus (P) are mineral nutrients often limiting plant growth because they are required in large amounts in relation to their availability in soil (Harpole et al., 2011, quoted by Ekwe, 2015).
However, nitrogen fertilization is very variable and it depends on the amounts of the element already present in the soil and the potential yield of the environment (Lauretti et al., 2007).
The first phase of vegetation is a critical period for nutrition with N, P and K, and the negative impact of their failure cannot be corrected later, even if it provides the best nutritional conditions.
Therefore, ensuring a good supply of sunflower plants with all the nutrients from the emergence is one of the main conditions for obtaining high yields.
The average yield per hectare at national level depends very much on the evolution of the climatic conditions and soil natural fertility (Bîlteanu, 2003).
Organic substances, such as: protein hydrolysates of animal origin (composed of peptides, ureide and amino acids - glycine, alanine, phenylalanine, proline, asparagine, glutamine, arginine, histidine, lysine, serine, threonine, valine) and protein hydrolysates of vegetal origin (algae extracts) can be successfully used to obtain new fertilizers formulas (Mihalache et al., 2014).
The relative efficacy of these fertilizers is determined by the rate and size of nutrient transport in plant (Cioroianu et al., 2009, quoted by Mihalache & Stanescu, 2017).
Agriculture of the future does not only have to be sustainable, but also performant and this is achieved only through the correct application of all the technological links specific to agricultural crops from various ecological areas of the country.
The specific consumption of sunflower for 100 kg of seeds, plus the production of roots, leaves, stems and inflorescences is 1.8-3.5 kg Nitrogen, 0.29-0.7 kg Phosphorus, 0.38-1.65 kg Potassium, 0.11 kg Calcium, 0.18-0.23 kg Magnesium (Hera et al., 1998).
Soils recommended for sunflower crops are those in clay or sandy-clay ones, averagely aerated, deep rich in humus and nutrients with a high usable water-retention capacity (Bîlteanu et al., 1988).

MATERIALS AND METHODS

The experiment was set up on illuvial- clay soil, decarbonated soil, typical, with a well-defined profile and insignificant differences regarding the physical, hydric and chemical attributes.
In its arable layer, soil has a cleyey texture, an apparently normal density (1.32 g/cubic centimetres) total satisfying porosity (51%) and a low ram degree (penetration resistance 28 kgf/mc) (Bălănescu & Ilinca, 2001).
In the next horizon, the texture becomes clay-clayey, volumetric weight increases quite a lot (1.57 g/cmc), and total porosity, as well as other attributes record low values, appearing an accentuated ram (penetration resistance reaches 63 kgf/mc).
Following the chemical properties, soil has a medium - hummus content in the arable layer (2.20%); it is poorly supplied with Nitrogen (0.104% N total), medium to good with Phosphorus (47 ppm P mobile) and good to very good supplied with Potassium (244.5 ppm K mobile), and the PH (in the water) is 7.6. The ground water depth is 8 up to 10 m.
The sunflower experiment was set up using the block method. Each experimental variant had a surface of 28 m^2: 2.8 x 10 m with a distance between rows of 0.7 cm over the course of three randomized repetitions.
The density of sunflower plants was 5.7 plants/m^2 (57.000 plants/ha).
For the sunflower crop, the hybrid who was used was Neoma hybrid, which is a simple hybrid, semi-early with an extraordinary and stable potential for production, resistant to the Express herbicide.
There were a number of 6 test variants used, as follows: V1 - $N_0P_0K_0$ (control); V2 - $N_{90}P_0K_0$; V3 - $N_{90}P_{60}K_0$; V4 - $N_{90}P_{60}K_{60}$; V5 - $N_{90}P_{60}K_{60}$ plus 1.5 liters/hectare of Fertigrain foliar fertilizer; V6 - $N_{120}P_{60}K_{60}$.
The works commenced on the fall of 2017. The first step in preparing the land was to make the ploughting at a depth ranging between 25 and 30 cm.
In the spring, the works continued with the soil disking in 2 passes, process executed using a heavy disc.

The final work in preparing the seedbed was performed using a combiner before planting the sunflower crop.
Fertilization was administered manually under the type of complexes before planting and the ammonium nitrate difference has been applied in vegetation.
Sowing took place on 23.04.2018. After that, Dual Gold 960 EC (active substance S-metolachlor 960 g/l) preemergent herbicide was used in a dose of 1.2 l/ha.
When the sunflower crop formed 4-6 leaves, the Select Super 120 EC (active substance Clethodim 120 g/l) herbicide was applied to annual and perennial monocotyledons weeds in a dose of 1.5 l/ha.
On the 8-10 leaves stage, a treatment made of Reveller fungicide (active substances Cyproconazole 80 g/l + Picoxystrobin 200 g/l) 0.6 l/ha and Decis Mega 50 EW insecticide (active substance Deltamethrin 50 g/l) 0.15 l/ha was applied.
Application of the foliar fertilizer occurred in 2 stages. The first application stage was conducted when the plants had formed 4-6 leaves, and the second stage before blooming period, in a dose of 1.5 l/ha.
Biometric determinations were performed on the sunflower plants, including the following: plant size and calatidium diameter.
The tests made on the sunflower seeds aimed at their quality, expressed by morphological indexes, such as: Thousand Kernel Weight TKW) and the hectolitre mass (HM).

RESULTS AND DISCUSSIONS

Climatic conditions registered in the 2017/2018 agricultural year by the weather station of Agricultural Research and Development Station Caracal were among the most favourable over the past 30 years.
The annual average of the temperature (Table 1) was between 0.8°C and 24.3°C registered in January and August, respectively.
Also, the multiannual average of temperatures ranged between 0.3°C and 23.9°C, resulting a difference of +1.1 ^{0}C between the annual and multiannual average.
While the monthly rainfall sum (Table 2) ranged between 6.8 and 147.8 mm, and the monthly average was between 26.3 and 69.7 mm, the difference between the rainfall sum and the monthly average was +301.9 mm.

Table 1. Temperatures registered at Agricultural Research and Development Station Caracal during the 2017/2018 agricultural year

Specifications	**Temperature °C**		
Month	**Monthly average**	**Multiannual media (last 30 years)**	**Deviation**
October	12.3	11.7	+ 0.6
November	6.5	5.4	+ 1.1
December	3.1	0.3	+ 2.8
January	0.8	1.3	+ 0.5
February	1.0	0.8	+ 0.2
March	3.8	6.0	- 2.2
April	16.3	12.0	+ 4.4
May	19.6	17.7	+ 1.9
June	22.1	21.6	+ 0.5
July	22.8	23.9	- 1.1
August	24.3	23.5	+ 0.8
September	19.6	18.1	+ 1.5
Total Average	**12.7**	**11.6**	**+ 1.1**

Table 2. Rainfall registered at Agricultural Research and Development Station Caracal during the 2017/2018 agricultural year

Specifications	**Rainfall (mm)**		
Month	**Monthly sum**	**Multiannual media (last 30 years)**	**Deviation**
October	144.6	46.0	+ 98.6
November	73.0	37.0	+ 36.0
December	63.8	39.1	+ 24.7
January	33.8	30.8	+ 3.0
February	56.4	26.3	+ 30.1
March	86.4	34.2	+ 52.2
April	10.6	47.8	- 37.2
May	55.6	58.6	- 3.0
June	134.2	69.7	+ 64.5
July	147.8	62.1	+ 85.7
August	30.6	46.6	- 16.0
September	6.8	43.5	- 36.7
Total Average	**843.6**	**541.7**	**+ 301.9**

The year 2018 turned out to be a climatically favourable year regarding the growth and development of the sunflower crop in the Oltenian plain.
Sunflower is a mesothermal plant, relatively demanding in heat which, to go through the

vegetation stages, needs at least 2,350°C (T > 0°C) or 1,600°C (T ≥ 5°C) (Bîlteanu et al., 1988).
During the research period, the sum of temperature degrees was 3,223.30°C, enough to meet the sunflowers biological demands.
The average of the air temperatures (Figure 1), between 16.3 and 22.1°C, recorded during the time span between emergence and inflorescence and those comprised between 22.8 and 24.3°C, registered since flowering until fruit-forming, were optimal temperatures to cover the vegetation period of the sunflower experiment.

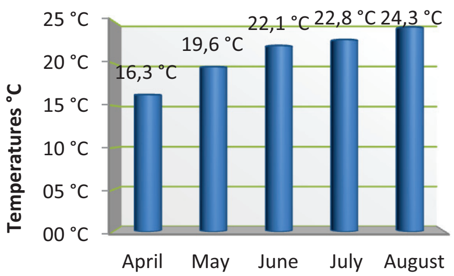

Figure 1. The average of temperatures during the vegetation period of sunflower

The humidity requirements for the sunflower are average, the rainfall estimated for the whole vegetation period being 400-500 mm (Bojan, 1986, quoted by Hera et al., 1989)
Rainfall average (Figure 2) was comprised between 10.6 and 55.6 mm between emergence and inflorescence emergence and 134.2 and 147.8, between flowering and fruit forming.

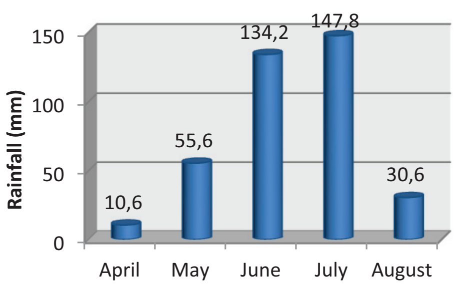

Figure 2. The average of rainfalls during the vegetation period of sunflower

The influence of fertilization on sunflower crop had a distinctly significant contribution regarding yield.

The yield differences (Tabel 3) between the variant $N_{90}P_{60}K_{60}$ plus Fertigrain foliar fertilizer and the variant $N_{90}P_{60}K_{60}$ is of 509 kg/ha, and compared to the control variant, it is of 1,801 kg/ha for the variant $N_{90}P_{60}K_{60}$ plus 1.5 liters/hectare of Fertigrain foliar fertilizer and 1,292 kg/ha for variant $N_{90}P_{60}K_{60}$.

Table 3. Yield differences between fertilization variants

A / B	V1	V2	V3	V4	V5	V6
	(A-B) kg/ha					
V1 - N0P0K0 (Control)	-	+ 706	+ 1110	+ 1292	+ 1801	+ 1410
V2 - N90P0K0		-	+ 404	+ 586	+ 1095	+ 704
V3 - N90P60K0			-	+ 182	+ 691	+ 300
V4 - N90P60K60				-	+ 509	+ 118
V5 - N90P60K60 plus foliar fertilizer					-	-391
V6 - N120P60K60						-

The best result was achieved by the variant $N_{90}P_{60}K_{60}$ plus foliar fertilizer, where the yield was 4,035 kg/ha, turning out to be the best fertilization variant, thanks to the foliar fertilizer used. Bellow this value was the variant $N_{90}P_{60}K_{60}$ with a production of 3,526 kg/ha (Table 4).
Even if at the variant 6 of fertilization, where $N_{120}P_{60}K_{60}$ was used, the yield obtained was 3,644 kg/ha, the yield difference of + 118 kg/ha, compared to variant $N_{90}P_{60}K_{60}$, is not significant, the higher amount of ammonium nitrate cannot be justified.
Very significant results (***) were also achieved at variant 3, where $N_{90}P_{60}K_0$ was applied. Therefore, the yield achieved was 3,344 kg/ha, exceeding the control variant with 1,110 kg/ha.
The distinctly significant difference (**), compared to the unfertilized control variant, was obtained at variant 2, where $N_{90}P_0K_0$ was applied. The registered yield was 2,940 kg/ha, recording a plus of 706 kg/ha.

Table 4. Influence of fertilization on the sunflower crop

Variant	Yield kg/ha	%	Differences	Signification
V1 - $N_0P_0K_0$ (control)	2,234	100	*Ct*	-
V2 - $N_{90}P_0K_0$	2,940	132	706	**
V3 - $N_{90}P_{60}K_0$	3,344	150	1,110	***
V4 - $N_{90}P_{60}K_{60}$	3,526	158	1,292	***
V5 - $N_{90}P_{60}K_{60}$ plus 1.5 l/ha of Fertigrain foliar fertilizer	4,035	181	1,801	***
V6 - $N_{120}P_{60}K_{60}$	3,644	163	1,410	***
	Kg/ha		%	
LSD 5%	273		12	
LSD 1%	413		19	
LSD 0,1%	664		30	

In terms of biometric determinations regarding plant size and head diameter (Table 5), which were carried out at flowering, they were different according to the fertilization degree.

The highest sizes were recorded at the following fertilization variants: $N_{90}P_{60}K_{60}$ plus foliar fertilizer variant, 184 cm, and $N_{120}P_{60}K_{60}$ variant, 176 cm.

Head diameter varied between 15.3 and 25.0 cm, the highest values was highlighted at variants $N_{90}P_{60}K_{60}$ plus foliar fertilizer, 25 cm, and $N_{120}P_{60}K_{60}$, 23.3 cm. The average achieved by variants was 20.2 cm, only 3 variants exceeding this result.

Table 5. Influence of fertilization on plant size and head diameter

Fertilization variant		Plant size (cm)	%	Head diameter (cm)	%
V1	$N_0P_0K_0$ (control)	151	100	15.3	100
V2	$N_{90}P_0K_0$	161	106	17.3	113
V3	$N_{90}P_{60}K_0$	168	111	19.3	126
V4	$N_{90}P_{60}K_{60}$	172	114	21.0	137
V5	$N_{90}P_{60}K_{60}$ + foliar fertilizer	184	121	25.0	163
V6	$N_{120}P_{60}K_{60}$	176	116	23.3	152
Average		**168.8**		**20.2**	

Determinations on the Thousand Kernel Weight (TKW) and the hectolitre mass (HM) turned out to be directly proportional to the fertilization degree.

Values regarding TKW were between 58.3 g at the unfertilized control variant and 64.3 g at the fertilization variant 5, where, besides $N_{90}P_{60}K_{60}$, foliar fertilizer was also applied.

The hectolitre mass (HM) registered values between 40.8 kg/hl at the control variant and 46.2 kg/hl at the fertilization variant $N_{90}P_{60}K_{60}$ plus 1.5 liters/ha of Fertigrain foliar fertilizer, (Table 6).

Table 6. Effect of fertilization on TKW and HM quality indicators

Fertilization variant		TKW (g)	HM (kg/hl)
V1	$N_0P_0K_0$ (control)	58.3	40.8
V2	$N_{90}P_0K_0$	60.9	41.5
V3	$N_{90}P_{60}K_0$	62.8	42.8
V4	$N_{90}P_{60}K_{60}$	63.2	43.5
V5	$N_{90}P_{60}K_{60}$ + foliar fertilizer	64.3	46.2
V6	$N_{120}P_{60}K_{60}$	63.4	44.5
Average		**62.1**	**43.2**

CONCLUSIONS

Sunflower is a plant who can adapt to various environmental conditions. In order to harness the biological potential of the plant, favourable ecological conditions need to exist, as well as a suitable agricultural practice.

Climatic conditions on the plain of Oltenia have proven to be some of the most favourable for the sunflower crop, both in terms of temperatures and precipitations, vegetation phases being strongly influenced by temperature.

The 2017/2018 agricultural year highlighted the results achieved behind fertilization by various fertilization doses.

Following the application of various fertilization doses to the sunflower crop, both very significant and distinctly significant results, were highlighted compared to the control variant.

The maximum level of production was recorded by variant $N_{90}P_{60}K_{60}$ plus foliar fertilizer, where a total of 4,035 kg/ha was obtained. The smallest production, compared to the unfertilized control variant, was obtained by the variant $N_{90}P_0K_0$ with 2,940 kg/ha, and the differences as against to the unfertilized control variant were between 706 and 1,801 kg/ha.

Also, the same variant $N_{90}P_{60}K_{60}$ plus foliar fertilizer registered the highest values in TKW and HM: 64.3 g and 46.2 kg/hl, respectively, compared to the unfertilized control variant.

It can be observed that the basic fertilization (NPK) in combination with the foliar fertilizer brings a significant production increase, detaching itself visibly from the other variants in terms of both production and seed quality.

ACKNOWLEDGEMENTS

The experiments in the field were carried out with the support of Agricultural Research and Development Station Caracal.

The foliar fertilizer was provided by Agri TecnoFertilizantes and the Neoma hybrid by Syngenta Company.

REFERENCES

Abubaker, B.A., Omer, A.A., Mohammad, A., Yu, S.E. (2014). Effect of different levels nitrogen and phosphorus fertilization on yield and chemical composition hybrid sunflower grown under irrigated condition. *Journal of Environmental and Agricultural Sciences, 1*(7), 01.

Ali, H., Ahmad, S., Muhammad, Y. (2004). Quantitative and qualitative traits of Sunflower (*Helianthus annuus* L.) as Influenced by Planting Dates and Nitrogen Application. *Int. J. Agri. Biol.*, *6*, 1–3.

Bălănescu, D., Ilinca, A. (2001). Agrochemical study of the soil in the research plot, Olt county. County Office for Agronomic and Pedological Studies, Scorniceşti.

Bîlteanu, Gh. (1988). *Mica enciclopedie agricolă*. Scientific and Encyclopedic Publishing House, Bucharest, 160–161.

Bîlteanu, Gh. (2003). *Phytotechnics*. Ceres Publishing House, Vol. 2, Bucharest, 13–61.

Bi, J., Liu, Z., Lin, Z., Alim, Md.A., Rehmani, M.I.A., Li, G., Wang, Q., Wang S., Ding. Y. (2013). Phosphorous accumulation in grains of japonica rice as affected by nitrogen fertilizer. *Plant Soil*, *369*, 231–240.

Ekwe, F. (2015). Effects of nitrogen and phosphorus on photosynthesis and growth of silver birch (*Betula pendula* Roth.) and sunflower (*Helianthus annuus* L.). MSc. Thesis, 45 hp, University of Gothenburg, Department of Biological and Environmental Sciences.

Elhassan, O.M.A., Elnaiem, A.A., Abbas, M.E., Mohamed, M.Y. (2006). Response of sunflower to different forms of fertilizer and nitrogen levels in the rain-fed area of the Blue Nile State. *Proceedings of the National Crop Husbandry Committee, Meeting, 41. ARC. Wad Medani. Sudan*, 26–34.

Harpole, W.S., Ngai, J.T., Cleland, E.E., Seabloom, E.W., Borer, E.T., Braken, M.E.S., Elser, J.J., Gruner, D.S., Helmut H., Shurin, J.B., Smith J.E. (2011). Nutrient co-limitation of primary producer communities. *Ecol Lett, 14*, 852–62.

Hera, Cr., Sin, Gh., Toncea, I. (1989). *Sunflower crop*. Ceres Publishing House, Bucharest.

Lauretti, D., Pieri, S., Vannozzi, G.P., Turi, M. Giovanardi, R. (2007). Nitrogen fertilization in wet and dry climate. *Helia, 30*(47), 135–140.

Lotfie, A.Y., Salah, B.M.A. (2013). Effect of seedbed preparation and nitrogen fertilizer on the performance of a sunflower (*Helianthus annuus* L.) hybrid under rain-fed conditions. *Res. J. Agril. Environ. Manage*., *2*, 27–32.

Malligawad, L.H., Parameshwarappa, K.G., Giriraj, K. (2004). Studies on the effect of ratios and level of NPK fertilizer nutrients of the productivity of hybrid sunflower under rainfed farming situations. *Proc. 16th Int. Sunflower Conference, Fargo, ND, USA, 1*, 377–386.

Mihalache, D., Sîrbu, C., Grigore, A., Cioroianu, Tr.M. (2014). Protein hydrolysates and amino-acids fertilizers–physicochemical characteristics. *Scientific Paper. Series A.*, *57*(2), 47.

Mihalache, D., Stanescu, A.M. (2017). Influence of some NPK fertilizers with protein hydrolysates on sunflower crop. *AgroLife Scientific Journal*, *6*(1), 159–165.

Moisii, I., Lazureanu, A., Sambotin, D., (2012). The impact of the environmental factors and of the fertilization`s level on production of oil and oil content on sunflower hybrids in the west side Romania. *Journal of Horticulture, Forestry and Biotechnology*, *16*(3), 120–123.

Panaitescu, L., Lungu, M., Niţă, S. (2010). Studies of sunflower crop in Constanta County. *Scientific Paper. Series A, Agronomy*, *LIII*. 385–391.

Weiss, E.A., (1983). *Oil seed crops*. Longman Inc. New York, U.S.A., 402–464.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EFFECT OF DIFFERENT NITROGEN DOSES IN DIFFERENT WINTER WHEAT PRODUCTION

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Abstract

One of the important ways to increase production of wheat is the application of fertilizers. Grain production at wheat is largely determined by the administration of nitrogen fertilizers, in close interdependence with the specific conditions of the year, the state of vegetation in winter entrance and its resumption in spring. Noteworthy is the fact that from the unfertilized agricultural fund N_0P_0 (mt) to the $N_{80}P_{70}$ was obtained an increase of 1628 kg/ha. This increase is reduced to 1144 kg/ha between $N_{80}P_{70}$ and $N_{120}P_{70}$ agricultural fund and 452 kg/ha between $N_{120}P_{70}$ and $N_{160}P_{70}$. With regard to the specific reaction at fertilizers application of different levels of fertility, Otilia variety had best capitalizes on the increasing nitrogen, achieving an increase (the average of the three agricultural funds) of 1068 kg/ha compared to Pitar control variety. The interaction between wheat varieties and fertilization doses revealed the Otilia variety and the $N_{160}P_{70}$ fertilization dose, with a yield of 8190 kg/ha. Concerning the behavior of varieties at increasing doses of chemical fertilizers, we find that the nine analyzed varieties obtain very significant production increases.

Key words*: winter wheat varieties, production, fertilization.*

INTRODUCTION

One of the world's largest grain crops is wheat, plant that provides the most nutritious food for humans and animals in the highest proportion.
Along with genetic factors, technological factors and above all, mineral fertilization is another way to increase and stabilize production of winter wheat production.
The results obtained worldwide shows that fertilization is one of the main factors in the growth of productions.
Productions obtained are linked and correlated with the quantities of fertilizers used. Without proper fertilization it is not possible to realize the productive potential of the new varieties created by geneticists and breeders (Hera Cr., 1984).
Wheat is known as a plant which reacts very well to the application of mineral and organic fertilizers, although the specific consumption of nutrients is relatively low: 2.3-3.3 kg N; 1.1-1.8 kg P_2O_5; 1.9-3.7 kg K_2O/l00 kg related berries + straw (Bîlteanu Gh., 1991).
Wheat is pretentious to fertilization due to particularities, namely: poorly developed root system, exploring a volume of soil on a low solubility depth and absorption of soil nutrients as well as the high need for fertilizers within a reduced interval (the flowering phase).
During the flowering wheat plants extract about 80% of the total amount of nitrogen and phosphorus as well as 85% of the total amount of potassium.
Of the technological factors, mineral fertilization is the main driving element of production and wheat quality management (Hera et al., 1971; Popescu et al., 1979).
The use of mineral fertilizers positively influences the quality indices and wheat production, by balancing the nutrients and in terms of optimal pedoclimatic conditions. Under normal water supply conditions, correct fertilization of wheat may increase protein content by 4-6% (Hera, 1979).
Quantities of mineral fertilizers to be used in the crop of winter wheat are imposed not only by climate and soil conditions, but also by the variety's ability to harness nutrients, his resistance to falling and diseases.
Production variations generated by the variation of climatic conditions can be reduced by practicing alternation of crops in the frame of the crop and a proper fertilization corresponding to the requirements of crops. (Sin Gh., Partal E., 2010)

The investigations carried out aimed to establish behavior of some winter wheat varieties created at NARDI Fundulea depending on some technological factors in Dobrogea pedoclimatic conditions.
Reduced production and poor harvest quality are often the result of poor quality technology but also insufficient fertilization or lack thereof failure to abstain and, last but not least, the use of inappropriate genotypes for the area where they are grown (Ceclan O.A. et al,.2015).

MATERIALS AND METHODS

Objectives of the researches carried out:
- checking the production potential of autumn wheat varieties lately created in Romania and choosing the most productive for Dobrogea;
-tracking the effect of different doses of mineral fertilizers on the productive potential of autumn wheat varieties.

The experience was placed on vermic chernozem from Agricultural Research and Development Station of Valu lui Traian, Constanta, according to the subdivided parcel method by two factors:

Factor A - Fertilization with 4 graduations:
- a_1 - N_0P_0;
- a_3 - $N_{120}P_{70}$
- a_2 - $N_{80}P_{70}$;
- a_4 - $N_{160}P_{70}$

Factor B - The winter wheat varieties:
•b_1 - Pitar; •b_2 - Litera; •b_3 - Voevod;
•b_4 - Pajura; •b_5 - Otilia; •b_6 - Miranda;
•b_7 – Izvor; • b_8 - Glosa; • b_9 - Boema

Production dates were calculated and interpreted according to the current statistical analysis of variance analysis.
The study of winter wheat varieties was done with the purpose to highlight the most appropriate in terms of production capacity, the constant of the productions from one year to the next, for their zoning in Dobrogea.
The pedoclimatic conditions in Dobrogea are relatively similar to those in the brown-red forest soil area of Fundulea, therefore the varieties tested have the origin of NARDI Fundulea.
A number of 9 winter wheat varieties have been tested, to determine the most suitable for expansion in Dobrogea agriculture. These winter wheat varieties tested are the result of the recombination of a very diverse germplasm.

RESULTS AND DISCUSSIONS

From recorded data at ARDS Valu lui Traian during 2012-2016 was observed that year 2012 was the driest, when was recorded a total rainfall of 394.9 mm with 43.7 mm below the annual average in 75 years.
The crop of winter wheat belongs to the category of those crop plants which maximizes the increasing levels of fertilization so that the allocation of 80 kg/ha active substance nitrogen together with 70 kg/ha active substance of P_2O_5 (Table 1) brings a very significant production increase, statistically insured, 1628 kg/ha (143%), the average of the nine wheat varieties tested.
Maintaining a constant phosphorus agricultural fund (70 kg/ha) in conjunction with the allocation of two more levels of nitrogen higher by 40 kg/ha a.s. N leads to substantial production increases.
The fact that at the last level of fertilization of 160 kg/ha a.s. N and 70 kg/ha a.s. P_2O_5 under non-irrigation conditions, production has no capping tendencies, leads to conclusion that the nine varieties of winter wheat could reap even higher levels of fertilization.
At the $N_{120}P_{70}$ fertilization dose, a very significant production increase is obtained of 2772 kg / ha (173%), statistically assured.
At the agricultural fund $N_{160}P_{70}$ is obtained the highest production increase of 3224 kg/ha (184%) very significant production increase.
Of note, is that from the unfertilized agricultural fund N_0P_0 (mt) to $N_{80}P_{70}$ there is an increase of 1628 kg/ha. It is reduced to 1144 kg/ha between $N_{80}P_{70}$ and $N_{120}P_{70}$ agricultural fund and at 452 kg/ha between $N_{120}P_{70}$ and $N_{160}P_{70}$ agricultural fund.
Concerning the specific application to fertilizer application of different levels of fertility (Table 2), we find that there are obvious genetic differences between the nine tested genotypes on the use of nitrogen and phosphorus fertilizers.

Table 1. Influence of different fertilization levels on average production of winter wheat grains (average of 9 varieties) at ARDS Valu lui Traian

No crt.	Agricultural fund Varieties	Grains yield (kg/ha)									Average yield / agrof. (kg/ha)	Difference		**Significance**
		Pitar	Litera	Voevod	Pajura	Otilia	Miranda	Izvor	Glosa	Boema		(kg/ha)	%	
1.	N_0P_0	3749	3686	3150	3623	4431	3896	3980	4106	3759	3822	mt	100	
2.	$N_{80}P_{70}$	5334	5187	4473	5103	6353	5534	5702	5912	5418	5450	1628	143	xxx
3.	$N_{120}P_{70}$	6489	6363	5397	6185	7560	6731	6878	7172	6542	6594	2772	173	xxx
4.	$N_{160}P_{70}$	6689	6899	5828	6699	8190	7193	7350	7581	6941	7046	3224	184	xxx

DL 5 % = 30 kg/ha
DL 1 % = 50 kg/ha
DL 0,1% = 70 kg/ha

Table 2. Production results obtained at nine varieties of winter wheat on four levels of fertilization, at ARDS Valu lui Traian

No crt.	Agricultural fund Variety	Yield kg/ha				Average yield (kg/ha)	Difference (kg/ha)	Significance
		N_0P_0	$N_{80}P_{70}$	$N_{120}P_{70}$	$N_{160}P_{70}$			
1	Pitar	3749	5334	6489	6689	5565	mt	
2	Litera	3686	5187	6363	6899	5534	-32	
3	Voevod	3150	4473	5397	5828	4712	-853	000
4	Pajura	3623	5103	6185	6699	5403	-163	00
5	Otilia	4431	6353	7560	8190	6634	1068	Xxx
6	Miranda	3896	5534	6731	7193	5839	273	Xxx
7	Izvor	3980	5702	6878	7350	5978	412	Xxx
8	Glosa	4106	5912	7172	7581	6193	628	Xxx
9	Boema	3759	5418	6542	6941	5665	100	

DL 5 % = 100 kg/ha
DL 1 % = 130 kg/ha
DL 0,1 % = 160 kg/ha

From this point of view, Otilia variety best capitalizes on increasing nitrogen doses, making a increase (average of the three agricultural fund) of 1068 kg/ha against the Pitar control variety, very significant production increase. Along with Otilia variety, three other varieties- Miranda, Izvor, and Glosa makes very significant increases for the probability of 0,1% (Glosa-628 kg/ha; Izvor-412 kg/ha; Miranda-273 kg/ha). Boema variety achieves an increase of 100 kg/ha, an increase that falls within the limits of experimental error. Smaller productions than the Pitar control variety are obtained from varieties: Litera, Pajura and Voevod. Litera variety is overtaken by 32 kg/ha of the Pitar control variety, differences that fall within the limits of experimental errors. Pajura variety is exceeded by 163 kg/ha of the Pitar variety, significant distinct production difference. Voevod variety, compared to the Pitar variety, produces less with 853 kg/ha, very significant production difference from a statistical point of view. This interaction is specific to each genotype. In the absence of chemical fertilizers against the Pitar variety, taken as control (Table 3) the highest yields are obtained by Otilia varieties with 4431 kg/ha and Glosa with 4106 kg/ha (682 kg/ha and 357 kg/ha of very significant yields). Izvor variety produces a yield of 3980 kg/ha, with an increase of 231 kg/ha (significant increase in yield).

Table 3. Behaviour of the nine varieties of winter wheat on different agricultural funds, at ARDS Valu lui Traian

Agricultural fund	**Varieties**	**Yield**		**Diff. by control**	**Significance**
		absolute (kg/ha)	**relative** (%)		
N_0P_0	Pitar	3749	100	mt	
	Litera	3686	98	-63	
	Voevod	3150	84	-599	000
	Pajura	3623	97	-126	
	Otilia	4431	118	682	***
	Miranda	3896	104	147	
	Izvor	3980	106	231	*
	Glosa	4106	110	357	***
	Boema	3759	100	10	
$N_{80}P_{70}$	Pitar	5334	100	mt	
	Litera	5187	97	-147	
	Voevod	4473	84	-861	000
	Pajura	5103	96	-231	0
	Otilia	6353	119	1019	***
	Miranda	5534	104	200	*
	Izvor	5702	107	368	***
	Glosa	5912	111	578	***
	Boema	5418	102	84	
$N_{120}P_{70}$	Pitar	6489	100	mt	
	Litera	6363	98	-126	
	Voevod	5397	83	-1092	000
	Pajura	6185	95	-304	00
	Otilia	7560	117	1071	***
	Miranda	6731	104	242	*
	Izvor	6878	106	389	***
	Glosa	7172	111	683	***
	Boema	6542	101	53	
$N_{160}P_{70}$	Pitar	6689	100	mt	
	Litera	6899	103	210	*
	Voevod	5828	87	-861	000
	Pajura	6699	100	10	
	Otilia	8190	122	1501	***
	Miranda	7193	108	504	***
	Izvor	7350	110	661	***
	Glosa	7581	113	892	***
	Boema	6941	104	252	*

DL 5% = 190 kg/ha
DL 1% = 250 kg/ha
DL 0,1% = 330 kg/ha

Larger yield than control Pitar and Boema varieties, with 3759 kg/ha and Miranda, with 3896 kg/ha, but yield increases fall within the limits of experimental errors. Pitar variety, performs higher yield to Litera variety (3686 kg/ha) and Pajura (3623 kg/ha), but yield differences do not exceed the probability of 5%. A single variety, Voevod, achieves a yield decrease of 599 kg/ha compared to the control variety Pitar, very significant difference. On the $N_{80}P_{70}$, agricultural fund we find that Otilia and Glosa varieties make very significant yied increases compared to the Pitar control variety. Izvor variety, achieves an increase that this time increases in significance, significant (N_0P_0) to very significant ($N_{80}P_{70}$). Along with these, there significant increase in the yield of the Miranda variety (200 kg/ha). Voevod variety is also outdated in this agricultural fund, Pitar variety, with a very significant yied difference, variety that exceeds Pajura variety also, with 231 kg/ha, significant yield gap. The other varieties - Litera and Boema - produces negative and positive yield differences, but not statistically assumed. Constantly maintaining the phosphorus dose to 70 kg/ha and increasing the nitrogen dose by 40 kg/ha, does not bring significant changes in hierarchy of reaction of varieties to reaction of control variety. Except for Pajura variety, which is exceeded by 304 kg/ha (significant production difference), the other varieties make similar statistical differences, with those on the $N_{80}P_{70}$ agricultural fund. Supplementing the nitrogen dose by another 40 kg/ha (N_{160}) and maintaining the phosphorus dose to 70 kg/ha, contributes to the slight modification of the varieties reaction hierarchy compared to the control variety.

At this level of fertilization, four varieties achieve very significant yield increases, compared to the Pitar variety, namely: Miranda, with an increase of 504 kg/ha; Izvor with an increase of 661 kg/ha; Glosa with an increase of 892 kg/ha and Otilia with an increase of 1430 kg/ha.

Table 4. Winter wheat varieties behaviour on different agricultural funds, at ARDS Valu lui Traian

No. Crt.	Variety	Agricultural fund	Yield (kg/ha)	Difference (kg/ha)	Significance
1	Pitar	N_0P_0	3749	mt	
		$N_{80}P_{70}$	5334	1585	xxx
		$N_{120}P_{70}$	6489	2740	xxx
		$N_{160}P_{70}$	6689	2940	xxx
2	Litera	N_0P_0	3686	mt	
		$N_{80}P_{70}$	5187	1501	xxx
		$N_{120}P_{70}$	6363	2677	xxx
		$N_{160}P_{70}$	6899	3213	xxx
3	Voevod	N_0P_0	3150	mt	
		$N_{80}P_{70}$	4473	1323	xxx
		$N_{120}P_{70}$	5397	2247	xxx
		$N_{160}P_{70}$	5828	2678	xxx
4	Pajura	N_0P_0	3623	mt	
		$N_{80}P_{70}$	5103	1480	xxx
		$N_{120}P_{70}$	6185	2562	xxx
		$N_{160}P_{70}$	6699	3076	xxx
5	Otilia	N_0P_0	4431	Mt	
		$N_{80}P_{70}$	6353	1922	xxx
		$N_{120}P_{70}$	7560	3129	xxx
		$N_{160}P_{70}$	8190	3759	xxx
6	Miranda	N_0P_0	3896	Mt	
		$N_{80}P_{70}$	5534	1638	xxx
		$N_{120}P_{70}$	6731	2835	xxx
		$N_{160}P_{70}$	7193	3297	xxx
7	Izvor	N_0P_0	3980	Mt	
		$N_{80}P_{70}$	5702	1722	xxx
		$N_{120}P_{70}$	6878	2898	xxx
		$N_{160}P_{70}$	7350	3370	xxx
8	Glosa	N_0P_0	4106	mt	
		$N_{80}P_{70}$	5912	1806	xxx
		$N_{120}P_{70}$	7172	3066	xxx
		$N_{160}P_{70}$	7581	3475	xxx
9	Boema	N_0P_0	3759	mt	
		$N_{80}P_{70}$	5418	1659	xxx
		$N_{120}P_{70}$	6542	2783	xxx
		$N_{160}P_{70}$	6941	3182	xxx

DL 5 % = 180 kg/ha
DL 1 % = 240 kg/ha
DL 0,1 % = 320 kg/ha

At this level of fertilization, Litera and Boema varieties obtain significant yield increases, Pajura variety exceeds the yield of Pitar variety, with an increase of 10 kg/ha, not statistically assured. Voevod variety, as well as the other levels of fertilization, is overtaken by the Pitar variety, with a very significant yield difference. As for behavior of varieties at increasing doses of chemical fertilizers (Table 4) have been found that the nine analyzed varieties obtain very significant yield increases. Thus, $N_{80}P_{70}$ agricultural fund performs against the N_0P_0 (control), the lowest yield of 1323 kg/ha to Voevod variety and the highest yied of 1922 kg/ha to Otilia variety. The same agricultural fund makes the following increases: 1480 kg/ha for Pajura variety; 1501 kg/ha for Litera variety; 1585 kg/ha to Pitar variety; 1659 kg/ha, Boema variety; 1722 kg/ha to Izvor variety and 1806 kg/ha to Glosa variety. At the next level of fertilization, $N_{120}P_{70}$, yield increases made against control N_0P_0 are between 2247 kg/ha to Voevod variety and 3129 kg/ha to Otilia variety. The growing order of contributions on this agricultural fund is: 2562 kg/ha for Pajura variety; 2677 kg/ha, for Litera variety; 2740 kg/ha, Pitar variety, 2783 kg/ha, Boema variety; 2835 kg/ha to Miranda variety and 3066 kg/ha to Glosa variety. At last level of fertilization $N_{160}P_{70}$, compared to the non-fertilized control variety there is a yield difference of 3076 kg/ha, to Pajura variety and 3759 kg/ha to Otilia variety. The following differences are observed for the other varieties analyzed: 2940 kg/ha to Pitar variety; 3076 kg/ha, for the Pajura variety; 3182 kg/ha to Boema variety; 3213 kg/ha, for Litera variety; 3297 kg/ha to Miranda variety; 3370 kg/ha for Izvor variety and 3475 kg/ha for Glosa variety.

CONCLUSIONS

Application of chemical fertilizers, especially those with nitrogen, has been shown to be the nutrient element which determines the largest yield increases.

Wheat efficiently harnesses chemical fertilizers, all tested varieties respond favourably to fertilization, through yield increases, thus there are differences between varieties related to the fertilization reaction.

Fertilizers with nitrogen must be accompanied by phosphorus and potassium, depending on the soil, the pre-emergence plant and the climatic conditions.

Phosphorus fertilizers contribute to increasing the effectiveness of applying nitrogen fertilizers.

The fractional application of nitrogen fertilizers ensures the supply of plants with this element throughout the vegetation period, giving the possibility of growing grain yields.

Growing in the same yield unit of several genetically diversified varieties, with different precocity periods and multiple varieties resistance, contributes to the continuous improvement of the yield potential, as well as to improving harvest stability.

REFERENCES

Bîlteanu, Gh. (1991). *Fitotehnie*. Editura Ceres, Bucureşti.

Ceclan, O.A., Racz, I., Kadar, R., Ceclan, L.A., Rusu, F. (2015). Influența nivelului de fertilizare asupra producției și a unor indici calitativi la un set de soiuri de grâu de toamnă. *Analele INCDA Fundulea, LXXXIII*. Electronic ISSN 2067-7758.

Hera, Cr. şi colab. (1971). Studiul interacţiunii ingrăşămintelor cu azot şi fosfor la graul cultivat în sud-estul ţării. Analele *ICCPT Fundulea, 37*(seria B).

Hera, Cr. şi colab. (1979). Influenţa unor factori tehnologici asupra conţinutului şi calităţii proteinei. *Analele ICCPT Fundulea, XLIV*, 115–128.

Hera, Cr., Eliade, Gh., Ghinea, I., Popescu, A. (1984). *Asigurarea azotului necesar culturilor agricole*. Editura Ceres, Bucureşti.

Popescu, S. şi colab. (1979). Influenţa factorilor tehnologici asupra conţinutului şi calităţii proteinei la grâu. *Analele ICCPT Fundulea, XLIV*, 1–20.

Sin, Gh., Partal, E. (2010). Influenţa rotaţiei şi a fertilizării asupra producţiilor de grâu şi porumb în contextul variaţiilor climatice. *Analele INCDA Fundulea, LXXVIII*(1). Electronic ISSN 2067-7758.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

MONITORING OF THE EVAPOTRANSPIRATION PROCESSES IN RIPARIAN GRASSLANDS

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Abstract

The paper presents the results obtained from monitoring the evapotranspiration processes in riparian grasslands located in Poeni village, Romania near Glavacioc River and the testing of portable instrumentation developed to monitor continuously the potential evapotranspiration (PET). The Penman-Monteith method was selected for the experimental study of PET groundwater dependent ecosystems in the INTER-ASPA project. The microclimate measurements for evapotranspiration assessments were performed between September 14 and 19, 2018. Following the preliminary mapping of the herbaceous vegetation, 12 species of grasses, 5 legumes and 23 species from other botanical families were identified. The PET varied from 0.71 to 1.07 (trees), 0.57 to 0.95 (shrubs), and 0.58 to 1.04 mm day^{-1} (grassland). The 7 days average of the geometrical mean of all vegetated surfaces was 0.86 mm day^{-1}, which can be considered a descriptive constant for the Poeni riparian ecosystem for the period of measurements. The PET rates varied significantly especially from wind speed, solar radiation and temperature fluctuations. It was found that grassland canopy had PET rates higher than the taller canopy of shrubs and very close to the tree canopy.

***Key words**: albedo, biometeorology, floristic composition, Penman-Monteith, Photosynthetically Active Radiation.*

INTRODUCTION

The hydrology of groundwater and streamflow clearly influences the dynamics of riparian and wetland ecosystems (Grimm et al., 1997) with direct effects on floristic composition. When modelling the flow in such vegetated systems, it is important to accurately quantify the seasonal riparian evapo-transpiration as a critical groundwater boundary condition (Goodrich et al., 2000). The way by which simulation of evapotranspiration (ET) is performed can modify calculated heads and consequently the system dynamics computations (Banta, 2000).

Riparian and wetland ecosystems usually host a substantial number of plant types and species. By using *plant functional groups* and their in-field identification, the complexity of individual species and phytosociological associations may be simplified into a relatively small number of general recurrent patterns (Maddock et al., 2012).

Species biological characteristics such as plant habitus and foliage architecture, and their ecophysiological response to the environmental conditions i.e., density, have key effects on the transpiration rates (Pearson & Ison, 1987). Meinzer et al. (1997) found that large woody plants detain various maximum rooting depths, hydraulic architecture and transpiration rates compared to smaller trees. Moreover, vegetated areas with high densities of herbaceous and woody plants were characterized by higher transpiration rates than scattered spatial arrangements of plant species (Monteith, 1981; Milică et al., 1982).

In Romania, riparian and wetland ecosystems often contain valuable grasslands of different typologies based on predominant grass species such as *Agrostis stolonifera*, *Alopecurus pratensis*, *Poa pratensis*, *Lolium perenne*, *Arrhenatherum elatius*, *Festuca pratensis* etc. depending on altitude. Among the woody species that forms the groves (riparian forests), *Alnus glutinosa* with *Alnus incana* (at higher altitudes), willows (*Salix fragilis* etc.), elms

(*Ulmus minor* etc.), poplars, and other species can be mentioned.

The riparian grasslands are spread over lowlands of riverside, terraces, river valleys and floodplains, but without water excess, on gley and alluvial soils, clay-loamy soils, weakly acidic or poorly alkaline. They are characterized by stable soil cover with vegetation (95-100%) and by the participation of the dominant species *Agrostis stolonifera* with 70%, which is a species with a high fodder value (Motcă et al., 1994). The herbaceous species forming the floristic composition of riparian grasslands generally have shallow root systems. In the mixed canopy of the riparian grasslands, grasses reach usually 40-50%, legumes 20-25%, rather high, and species from other botanical families approximately 25%.

Among all these species, there are plants with no forage value or harmful to the animal products: *Deschampsia caespitosa*, *Potentilla reptans*, *Rumex crispus*, *Mentha aquatica*, *Gratiola officinalis*, *Ranunculus repens*, *Stellaria graminea*, *Juncus effusus*, *Carex hirta* etc. The forage yield of these grasslands is 1.5-3.5 t ha^{-1} DM (7.5-17.5 t ha^{-1} green fodder) and the pastoral value is 1.75-2.75 (based on ground cover) or 35-55 (based on frequency), which falls within the category of intermediate quality grasslands (Motcă et al., 1994). *Agrostis stolonifera* grassland type supports an animal load of 0.7-1.0 LSU ha^{-1} (livestock unit), but because of the increased soil moisture, mowing is recommended. The following terms are related to the riparian evapotranspiration:

Evaporation and transpiration are important components of the water circuit in nature that is also important for groundwater dependent ecosystems (GDE). Evaporation is the loss of water in the form of vapours from a wide variety of surfaces, lakes, rivers, bare soil and vegetation (Dinca et al., 2017). Transpiration consists in the loss of water contained in plant tissues and the elimination of produced vapours in the surrounding atmosphere. Plants preponderantly lose water through stomata. Then, evaporation and transpiration occur simultaneously and it is not easy to distinguish between the two processes.

Potential evapotranspiration (PET) is the rate at which evapotranspiration can occur from a wide area, fully and evenly covered with vegetation in the growing stage that has unlimited access to soil water resources and without direct effects of the advection or heating processes (Dingman, 2014).

Evapotranspiration of the reference crop (ET_o) is the evapotranspiration amount specific to a cultivated species that is not limited in terms of water availability. FAO-56 adopts the specific characteristics of the reference crop with a certain height (0.12 m), surface resistance (70 s m^{-1}) and albedo (0.23). Then, ET_o is determined using the Penman-Monteith equation (McMahon et al., 2013).

Actual evapotranspiration (ET_a) is defined as the amount of water transferred as water vapour from the surface of an evaporation surface (surface water, bare soil, vegetation-covered area etc.). The process of evapotranspiration depends on meteorological, edaphic, and plant physiological factors.

In a heterogeneous approach, four groups of factors that influence the magnitude of the ET processes may be considered:

(i) *Meteorological parameters*: net radiation, air temperature, air relative humidity, and wind speed.

(ii) *Factors related to the phytosociological association or the cultivated species*: differences in plant resistance to transpiration or conductivity of the vegetal canopies, height of the canopy, canopy roughness, albedo, and characteristics of the root system.

(iii) *Environmental conditions*: ground cover, plant density and soil water content.

(iv) *Trophic and limiting factors*: macronutrients and pollutants.

ET can be measured directly in the field using instruments such as lysimeters, evaporimeters and eddy covariance systems for measuring the fluxes of energy over a variety of ecosystems (Burba & Anderson, 2005; Goss & Ehlers, 2009; Parisi et al., 2009).

These tools are expensive and difficult to maintain in order to provide point ET measurements, which sometimes are not valid to be applied for larger areas (being spatially limited).

For overcoming these problems, various models for ET assessment were developed depending on the number of variables used e.g., Thornthwaite, Blaney-Criddle, Penman, Penman-Monteith, FAO-56, Priestley-Taylor, FAO-24 Blaney-Criddle, Turc, Hargreaves-Samani, Morton etc. (Allen et al., 1998).

Models for ET estimation range from simple empirical equations to complex models based on radiative-advective balance using synthetic regional indices (e.g., land use/land cover, Leaf Area Index obtained from NDVI – Normalized Difference Vegetation Index, albedo, and FAPAR - Fraction of photosynthetically active radiation) obtained from remote sensing systems (McShane et al., 2017). Most of these models are applicable to the weather and vegetation conditions of a particular region, and the synthetic coefficients that apply to those locations limit their use in other regions (so, they may have a diminished degree of generalization). Then, such models are widely used to estimate ETs coefficients in various area of study followed by their calibration and validation (Goudriaan, 1977).

In a comparative study conducted in Romania using lysimeters, it was found that, according to the meteorological data available in the studied area, several methods i.e., Blaney-Criddle, Penman and Thornthwaite provide acceptable results for estimating ET_o. Blaney-Criddle provided 15-20% higher monthly ET_o values compared to the other two methods. The results obtained from Penman and Thornthwaite methods are almost equal (ET_o values supplied by Penman were slightly higher for the first months of the vegetation period, due to the quantification of spring wind characteristics). For Romania, Grumeza and Kleps (2005) recommended the Thornthwaite method because it does not involve the use of laborious calculations and is validated over time. If sufficient meteorological records are available, it is recommended to use the revised Penman method because it better accounts the conjugated effect of climatic factors.

Due to the availability of meteorological data at hydrographical basin scale and specific in-situ instrumentation to monitor the required parameters (inputs in the evapotranspiration algorithms), the Penman-Monteith and the Priestley-Taylor methods were selected for the experimental study of GDE evapotranspiration to be considered in the INTER-ASPA project (PN-III-P1-1.2-PCCDI-2017-0721), titled "*Tools for modelling processes at the interface between water, soil, plants and air in order to promote the sustainable management of groundwater dependent ecosystems and their integrating river basins*", and funded by the Romanian Government.

INTER-ASPA project aims to create an innovation ecosystem designed to support the mono-disciplinary, inter-disciplinary and trans-disciplinary development of research domain related to the interface processes between water, soil, plant and air on several strategic cycles. The project is developed between 2018 and 2020. In the framework of INTER-ASPA, one of the research objectives is the characterization of evapotranspiration processes at hydrographical basin scale for GDE including riparian ecosystems.

The paper presents the preliminary results obtained from monitoring the evapo-transpiration processes in riparian grasslands and the testing of portable instrumentation developed to monitor continuously the PET.

MATERIALS AND METHODS

As planned in the INTER-ASPA project, field measurements have been prepared to establish an adequate methodology for the determination of evapotranspiration in GDEs. The experimental site, considered relevant from the point of view of the project objectives, was chosen in the Glavacioc River basin (cadastral code: X-1.23.11.8) in the wetland area of Poeni village, Teleorman County, south of Romania.

The Glavacioc River is a tributary of the Câlniştea River having a length of 120 km, a sinuosity coefficient of 1.69 and an average altitude of 118 m. It has a basin of 682 km^2, oriented from northeast to southeast, more developed in the upper part. In the Poeni village administrative area, there are forests, agricultural lands and grasslands, but also oil extraction fields. Potential complex GDEs are located in the northwest and southeast of Poeni village.

Figure 1. The study area with riparian and wetland ecosystems located in Poeni, Teleorman County near the Glavacioc River (stereographic coordinates of central point N 322137.52 m; E 527447.87 m)

Figure 2. Instrumentation used for *in situ* monitoring of evapotranspiration at riparian grassland level in Poeni, Teleorman County (beam fraction sensor for PAR monitoring; digital anemometers; microclimate multiparameters; temperature sensors; soil moisture sensors)

Figure 1 presents the study area with riparian and wetland ecosystems located in Poeni, Teleorman County near the Glavacioc River (stereographic coordinates of central point N 322137.52 m; E 527447.87 m measured with a GPS receiver Garmin Oregon 650t).

The microclimate measurements for evapotranspiration assessments were performed between September 14 and 19, 2018 using a PAR monitoring system comprising a beam fraction sensor, 2 digital anemometers, 2 microclimate multi-parameters, air and soil temperature sensors, and soil moisture sensors (Figure 2).

PAR measurements were performed continuously between 11.00 a.m. and 1.00 p.m. at a sampling rate of 10 seconds.

The parameters related to the advective processes (temperature, relative air humidity and soil moisture) were measured discontinuously in 6 points (3 inside, and 3 outside the herbaceous canopy).

The distances between monitoring points were measured with a professional measuring wheel.

Albedo (the ratio between reflected and global radiation) was determined with a portable pyranometer for different vegetated surfaces (Figure 3).

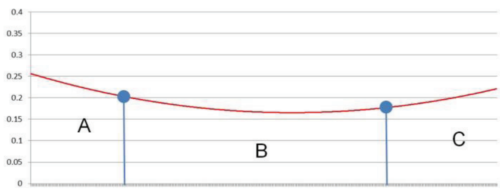

Figure 3. Albedo variations in correlation with the vegetation type (A-*grassland*; B-*trees*; C-*shrubs*)

In parallel with the microclimate measurements, a quick inventory of the floristic composition was performed to assess the plant species existing in the study area.
The data obtained from in-situ monitoring was used in the Penman-Monteith algorithm (Dingman, 2014) requiring the canopy height, the conductance properties of the leaf, the albedo, the LAI, the atmospheric pressure, the incident global radiation, the cloud cover, the air temperature, the relative humidity, the wind speed, and the soil moisture deficit.

$$PET = \frac{1}{\lambda}\frac{\Delta(R_n - g) + \rho_a c_a \,{(v'_a - v_a)}/{r_a}}{\Delta + \gamma(1 + {r_s}/{r_a})}$$

where: *PET* is Penman-Monteith's potential evapotranspiration; R_n is the daily net incident radiation on the vegetated surface; g is the soil heat flux; ρ_a is the average air density at constant pressure; c_a is the specific air heat; r_a is the aerodynamic resistance; r_s is the surface resistance; (v'_a - v_a) represents the pressure deficit of the air vapours.
PET was calculated for three types of vegetated surfaces existing in Poeni riparian ecosystems i.e., trees, grassland and shrubs during the monitoring period (September 14-19, 2018). Leaf conductance properties for riparian ecosystems were retrieved from literature. LAI was assessed using a scanning procedure and area meter recognition by software (Dunea & Moise, 2008). It is expected that in the 2nd phase of the INTER-ASPA project, a porometer for leaf conductance measurements and the Delta-T Devices Sunscan system for LAI will be used for more accurate *in situ* estimations. Table 1 presents the values of inputs used in this paper.

Table 1. Inputs used in the Penman-Monteith algorithm for the estimation of evapotranspiration in Poeni near Glavacioc River (September 14-19, 2018)

Variable	TREES	SHRUBS	GRASSLAND
Canopy Height (m)	8	1.2	0.45
Conductance factor	0.50	0.46	0.49
Maximum leaf conductance (mm/s)	2.30	2.16	2.20
Albedo	0.19	0.20	0.21
LAI (m^2 m^{-2})	3.3	2.9	2.6

RESULTS AND DISCUSSIONS

The heterogeneous canopies from wetlands and riparian ecosystems are characterized by various rates of solar energy conversion into dry matter, thus various biological efficiencies (light use efficiencies) and implicitly different rates of water use efficiency (McCree, 1973; Campbell, 1986; Sinoquet, 1993; Dunea et al., 2015). For this reason, the main objective of the field experiments planned within the INTER-ASPA project is to establish GDEs-specific evapotranspiration rates and the magnitude of the influence of macronutrients and pollutants on them. Also, the influence on eco-physiological indicators such as the efficiency of radiation use and the efficiency of water use will be also determined. The resulted coefficients will provide useful information for downscaling the information retrieved from the remote sensing systems such as MODIS, Sentinel 2, Landsat 8, and PROBA-V (Dunea et al., 2014).
Following the preliminary mapping of the herbaceous vegetation, 12 species of grasses, 5 legumes and 23 species from other botanical families were identified. In the riparian grassland, several types of heterogeneous canopies have been observed, which requires a unitary approach to characterize the eco-physiological processes influence by the evapotranspiration characteristics.
The perennial grasses accounted for 60-65% having a good ground cover (>80%) with the following species: *Agrostis stolonifera, Alopecurus pratensis, Poa pratensis, Lolium perenne, Agrostis canina, Festuca pratensis, Poa trivialis, Holcus lanatus, Agropyron repens, Poa palustris, Cynodon dactylon, Glyceria maxima. Agrostis stolonifera, Alopecurus pratensis, Poa pratensis, Lolium perenne, Arrhenatherum elatius, Festuca pratensis* etc. The dominant species was the creeping bentgrass *Agrostis stolonifera*, a sod-forming grass used on reclamation sites, lawns and for soil erosion control.
The identified legumes were *Trifolium repens, Trifolium pratense, Medicago lupulina, Galega officinalis, Vicia cracca* etc. having a low participation in the grassland canopy (10-15%).

Figure 4. Riparian grassland at Poeni, Teleorman County near Glavacioc River - *Agrostis stolonifera* type

Species from other botanical families (25-30%) were: *Typha angustifolia* (dominant species especially near river banks), *Taraxacum officinale, Plantago major, Cichorium inthybus, Symphytum officinale, Daucus carota, Leontodon autumnalis, Deschampsia caespitosa, Potentilla reptans, Rumex crispus, Mentha aquatica, Gratiola officinalis, Ranunculus repens, Stellaria graminea, Juncus effusus, Carex hirta, Rosa canina, Tragopogon pratensis, Prunella vulgaris, Glycyrrhiza echinata, Potentilla anserine, Arctium lappa,* and *Bidens tripartite*.
The most frequent woody species grouped in rare groves were: white poplar (*Populus alba*), willows (*Salix fragilis* etc.), elms (*Ulmus minor* etc.) and fruit-growing trees.
Consequently, we have assessed the evapotranspiration for three types of canopies (grassland; shrubs; trees) using the coefficients and values from Tables 1 and 2.
Table 3 shows the PET values provided by the Penman-Monteith algorithm for each day during September 14 and 19 (7 days).

Table 2. Daily averages of the meteorological inputs used in the Penman-Monteith algorithm for the estimation of evapotranspiration of riparian ecosystem in Poeni near Glavacioc River (September 14-19, 2018), altitude 117 m

Date	Pressure	Radiation	Cloud cover	Air Temp	Rel. Hum.	Wind speed
	kPa	MJ m^{-2}	-	°C	%	m s^{-1}
14.09	100.32	22	0.25	21.8	0.59	1.6
15.09	100.36	19	0.4	23.3	0.52	2.7
16.09	100.80	21	0.1	21.8	0.62	2.9
17.09	101.17	20	0.3	20.6	0.59	3.6
18.09	101.07	21	0.1	20.3	0.58	2.5
19.09	101.07	23	0	19.3	0.55	2.8
20.09	100.91	24	0	19.1	0.54	2.1

The PET varied from 0.71 to 1.07 (trees), 0.57 to 0.95 (shrubs) and 0.58 to 1.04 mm day^{-1} (grassland). A rate of 1 mm day^{-1} is equivalent with a loss of 10 m^3 ha^{-1} day^{-1}. The 7 days average of the geometrical mean of all vegetated surfaces was 0.86 mm day^{-1}, which can be considered a descriptive constant for the Poeni riparian ecosystem for the period of measurements. Interestingly, the PET varied significantly especially from wind speed, radiation and temperature fluctuations. Grassland canopy showed PET rates higher than the taller canopy of shrubs including *Typha* species, having the highest variance.

Table 3. Potential evapotranspiration computed with Penman-Monteith algorithm for various vegetated surfaces in Poeni near Glavacioc River

Date	PET trees	PET shrubs	PET grassland	*Geometrical mean*
14.09	0.968	0.866	0.945	***0.92***
15.09	0.707	0.569	0.58	***0.61***
16.09	0.964	0.827	0.882	***0.89***
17.09	0.889	0.719	0.734	***0.78***
18.09	0.883	0.839	0.891	***0.87***
19.09	1.031	0.884	0.943	***0.95***
20.09	1.069	0.952	1.039	***1.02***
Mean	***0.93***	***0.81***	***0.86***	***0.86***
CV%	*12.85*	*15.67*	*17.89*	*15.48*
Min.	*0.71*	*0.57*	*0.58*	*0.61*
Max.	*1.069*	*0.952*	*1.039*	*1.02*

Concomitantly with the PET measurements, PAR measurements were performed (Figure 5). The average of the PAR during 11.00 a.m. to

1.00 p.m. was 1343.5 μmols m^{-2} s^{-1} (CV = 14.5%), while the diffuse PAR was 250.2 μmols m^{-2} s^{-1} (CV = 6.2%). Regarding the albedo of riparian grassland, the overall average was 0.19, which is in line with other reported values for wetlands and riparian grasslands (Geiger, 1965; Baumgardner et al., 1985; Kotoda, 1986).

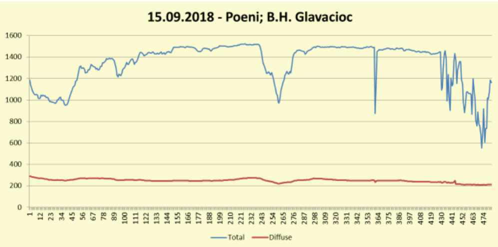

Figure 5. Example of PAR (μmols m^{-2} s^{-1}) measured from 11.00 a.m. to 1.00 p.m. in Poeni riparian grasslands

Following the monitoring campaign performed in Poeni riparian grasslands, it was concluded that continuous monitoring of PET would be more useful to understand the diurnal-nocturnal cycles of ET. To achieve this goal, the continuous monitoring has been started in the riparian grasslands of Ialomita River, Romania (Figure 6), by deploying a portable PET system based on adequate sensors interfaced on the Delta-T Devices GP2 data logger that uses the DeltaLINK 3.8 software, which is able to provide hourly calculations of PET based on Penman-Monteith algorithm(https://www.delta-t.co.uk/software/deltalink/).

The system has been tested to obtain hourly time series of PET that will be used to extract/validate useful key figures from remote sensing information.

CONCLUSIONS

The current study allowed the development of the methodological background to monitor evapotranspiration processes that occur in riparian grasslands. It is expected that the methodological approach will provide sustainable results also for cash crops monitoring for harvest index forecasting (Ion et al., 2015) and for characterization of the crop-weed interactions (Spitters & Aerts, 1983).

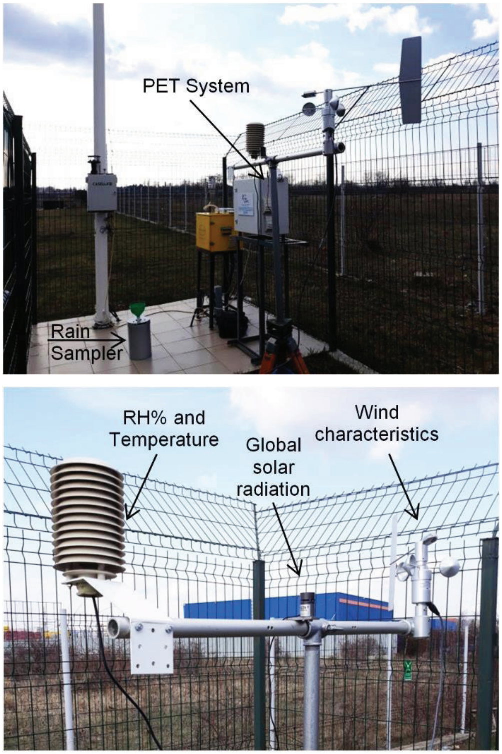

Figure 6. The PET continuous monitoring system deployed in the monitoring platform of the Department of Environmental Engineering - Valahia University of Targoviste, for assessing the canopy characteristics of riparian grasslands from Ialomita River, Romania

It was found that grassland canopy had PET rates higher than the taller canopy of shrubs and very close to the tree canopy. Since PET estimations are important for modelling hydrological processes and for groundwater evapotranspiration assessment, the presented methodology will be further refined in new monitoring campaigns in various riparian grasslands.

ACKNOWLEDGEMENTS

This research work was carried out with the support of INTER-ASPA_project "*Tools for modeling processes at the interface between water, soil, plants and air in order to promote the sustainable management of groundwater dependent ecosystems and their integrating river basins*", PN-III-P1-1.2-PCCDI-2017-0721, funded by the Romanian Government, http://inter-aspa.ro/index_en.html

REFERENCES

Allen, R.G., Pereira, L.S., Raes, D., Smith, M. (1998). Crop evapotranspiration - Guidelines for computing crop water requirements, FAO Irrigation and Drainage, pp. 56. Food and Agriculture Organization of the United Nations.

Banta, E.R. (2000). MODFLOW-2000, the U.S. Geological Survey modular ground-water model-Documentation of packages for simulating evapotranspiration with a segmented function (ETS1) and drains with return flow (DRT1): U.S. Geological Survey Open-File Report 00-466.

Baumgardner, M.F., Sylva, L.F., Biehl, L.L., Stoner, E.R. (1985). Reflectance Properties of Soils. *Adv. Agron., 38*, 1–44.

Burba, G., Anderson, D. (2005). A Brief Practical Guide to Eddy Covariance Flux Measurements: Principles and Workflow Examples for Scientific and Industrial Applications, LI-COR Biosciences, Retrieved March 10, 2019, from https://www.licor.com/env/pdf/eddy_covariance/Brief_Intro_Eddy_Covariance.pdf)

Campbell, G.S. (1986). Extinction coefficients for radiation in plant canopies using an ellipsoidal inclination angle distribution. *Agric. For. Meteor., 36*, 317–321.

Dinca, N., Dunea, D., Casadei, S., Petrescu, N., Barbu, S. (2017). An assessment of the water use efficiency in alfalfa canopy under the climate regime of Targoviste Piedmont Plain. *Scientific Papers-Series A, Agronomy, 60*, 235–240.

Dingman, S.L. (2014). *Physical Hydrology*. Waveland Press, Inc.; 3rd edition.

Dunea, D., Moise, V. (2008). Artificial neural networks as support for leaf area modelling in crop canopies, New Aspects of computers. *Proceedings of the 12th WSEAS International Conference on COMPUTERS*, 440–446.

Dunea, D., Neagu Frasin, L.B., Dinca, N. (2015). Ecophysiological responses of white clover-hybrid ryegrass mixture to foliar fertilisation. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, *43*(1), 173–178.

Dunea, D., Dinca, N., Iordache, S. (2014). Spatio-temporal characterization of natural grasslands from Bucegi Natural Park using remote sensing resources. *Scientific Papers. Series A. Agronomy, 57*, 180–185.

Geiger, R. (1965). *The Climate near the Ground.* Harvard Univ. Press: Cambridge, MA.

Goodrich, D.C., Scott, R., Qi, J., Goff, B., Unkrich, C.L., et al. (2000). Seasonal estimates of riparian evapotranspiration using remote and *in situ* measurements. *Agricultural and Forest Meteorology*, *105*, 281–309.

Goss, M.J., Ehlers, W. (2009). The role of lysimeters in the development of our understanding of soil water and nutrient dynamics in ecosystems. *Soil Use and Management*, *25*, 213–223.

Goudriaan, J. (1977). Crop Micrometeorology: A Simulation Study. Centre for Agricultural Publication Documentation, Wageningen, Netherlands.

Grimm, N.B., Chacon, A., Dahm, C.N., Hostetler, S.W., Lind, O.T., Starkweather, P.L., Wurtsbaugh, W.W. (1997). Sensitivity of aquatic ecosystems to climatic and anthropogenic changes: the Basin and Range, American Southwest and Mexico. *Hydrological Processes, 11*, 1023–1041.

Grumeza, N., Kleps, C. (2005). *Irrigation Arrangements in Romania*. Bucureşti, RO: Ed. Ceres,

Ion, V., Dicu, G., Dumbravă, M., Temocico, G., Alecu, I.N., Băşa, A.G., State, D. (2015). Harvest index at maize in different growing conditions. *Romanian Biotechnological Letters*, *20*(6), 10951.

Kotoda, K., (1986). Estimation of River Basin Evapotranspiration. *Environmental Research Center Papers, University of Tsukuba, 8*.

Maddock, T., Baird, K.J., Hanson, R.T., Schmid, W., Ajami, H. (2012). RIP-ET: A riparian evapotranspiration package for MODFLOW-2005: U.S. Geological Survey Techniques and Methods 6-A39, 76 p.

McCree, K.J. (1973). The measurement of photosynthetically active radiation*. Solar Energy*, *15*, 83–87.

Meinzer, F.C., Andrade, J.L., Goldstein, G., Holbrook, N.M., Cavelier, J., Jackson, P. (1997). Control of transpiration from the upper canopy of a tropical forest: the role of stomatal layer and hydraulic architecture components. *Plant, Cell and Environment*, *20*, 1242–1252.

McMahon, T.A., Peel, M.C., Lowe, L., Srikanthan, R., McVicar, T.R. (2013). Estimating actual, potential, reference crop and pan evaporation using standard meteorological data: a pragmatic synthesis. *Hydrol. Earth Syst. Sci.*, *17*, 1331–1363.

McShane, R.R., Driscoll, K.P., Sando, R. (2017). A review of surface energy balance models for estimating actual evapotranspiration with remote sensing at high spatiotemporal resolution over large extents: U.S. Geological Survey Scientific Investigations Report 2017–5087, 19 p., https://doi.org/10.3133/sir20175087.

Milică, C. et al. (1982). *Fiziologie vegetală*. Bucureşti, RO: Editura Didactică şi Pedagogică.

Monteith, J.L. (1981). *Physiological Processes Limiting Plant. Productivity*. London, UK: Johnson Editure.

Motcă, Gh., Oancea, I., Geamănu, L.I. (1994). *Pajiştile României, tipologie şi tehnologie*. Bucureşti, RO: Editura Tehnică Agricolă,

Parisi, S., Mariani, L., Cola, G., and Maggiore, T. (2009). Mini-Lysimeters Evapotranspiration Measurements on Suburban Environment. *Italian Journal of Agrometeorology*, *13-16*(3).

Pearson, C.J., Ison, R.L. (1987). *Agronomy of Grassland Systems*. Cambridge: Cambridge University Press.

Sinoquet, H. (1993). Modelling radiative transfer in heterogeneous canopies and intercropping systems. In: Varlet-Grancher C., Bonhomme R., Sinoquet H. (eds.) *Crop structure and light microclimate*. Paris, FR: INRA Editions, 229–252.

Spitters, C.J.T., Aerts, R. (1983). Simulation of competition for light and water in crop-weed associations. *Aspects Appl. Biol*., *4*, 467–483.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

GRAIN YIELD OF MAIZE HYBRIDS FROM DIFFERENT MATURITY GROUPS INFLUENCED BY NITROGEN AND PHOSPHORUS FERTILISATION

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Abstract

Understanding the relationship between yield and macronutrients fertilization is essential in maize cultivation technology, especially in non-irrigated conditions that require the rational use of water and fertilizers to ensure efficient crops. From this point of view, the purpose of the research was to determine the influence of mineral fertilization with nitrogen (N) and phosphorus (P) on the yield of different maturity groups maize hybrids and the identification of those hybrids with the best adaptation to the specific soil and climatic conditions of the area (Sarichioi, Tulcea County, Dobrogea). The research involved sowing eight maize hybrids from different maturity groups: early hybrids (DKC 3623 - FAO 260 and P8567 - FAO 275), mid-early hybrids (DKC 4590 - FAO 350 and P9903 - FAO 360), mid-late hybrids (DKC 4717 - FAO 410, P0023 - FAO 400 and P0412 - FAO 480) and late hybrids (DKC 5632 - FAO 510) under different mineral fertilization conditions with doses of nitrogen (N) and phosphorus (P) (N_0P_0; $N_{60}P_0$; $N_{60}P_{60}$; $N_{120}P_{60}$). Nitrogen fertilization led to a yield increase with values between 7.50 kg/kg N a.s. (for the hybrid P99038) and 10.75 kg/kg N a.s. (at P99038 hybrid), and application of phosphorus resulted in yield increases from 4.17 kg/kg P a.s. (P8567 and P0412) to 5.67 kg/kg P a.s. (DKC 4590). The eight hybrids responded to fertilization with nitrogen and phosphorus, their yield ranging from 6.46 t/ha for the hybrid DKC 3623 hybrid unfertilized to 9.09 t/ha to for the hybrid P0412 fertilized $N_{120}P_{60}$. On average, mid-late hybrids (FAO 400-480) recorded the highest yields with values between 7.35 t/ha and 8.77 t/ha.

Key words: *Zea may hibrids, FAO maturity groups, grain yield, fertilization, Dobrogea area.*

INTRODUCTION

Fertilization is essential for the corn crop technology, directly related to its yield and other productivity elements.
Nitrogen (N) plays a fundamental role in the nutrition of corn plants (Sangoi L., 2001; Miao Y. et al., 2006; Al-Naggar A.M.M., 2015; Gidea M. et al., 2015; Prisecaru G. et al., 2017), in their vegetative development and in yield implicitly. Following water, nitrogen is the second most important factor that influences corn yield (Ran H. et al., 2017). Phosphorus (P) is of particular importance in root formation and increase in drought resistance, photosynthesis and in the transport and activation of enzymes, while the lack of phosphorus can directly influence yield (Pettigrew W.T., 2008). Numerous researches demonstrated the direct proportional relationship between fertilization with these macronutrients and corn yield. Nitrogen fertilization produces the highest yield increases of 2 to 6 t/ha depending on soil fertility (Berca M. & Buzatu C.S., 2011).
Long-term research on a clay-sandy soil in NV China has shown that in the absence of fertilization grain yield has diminished at the end of the experimentation period, representing 28.2% of the yield obtained in the first year of research, but the yield obtained in response to fertilization with nitrogen and phosphorus have improved over time (Yang S. et al., 2004).
In the field conditions of southern Romania (INCDA Fundulea, on a chernozem soil type), to ensure a yield of more than 9 t/ha, the required amount of nitrogen was between 140 and 166 kg N/ha and phosphorus must be provided at doses of 41-73 kg P/ha (Habdan V.G., 2009). In irrigated conditions, the average yield was between 12.1 and 26.8 kg grains/kg N, and for non-irrigated crop yield increases were from 10.8 to 24.6 kg grains/kg N (Habdan V.G., 2009).

Research in Kansas, United States, demonstrated a maximization of corn yield under the application of a 185 kg N/ha (Gehl R.J. et al., 2005).
Research carried out in eastern Romania (SCDA Secuieni, on a cambic chernozem) shows that with nitrogen fertilization, for every kg of applied fertilizer the average grain yield increase was between 12.21 and 19.54 kg/kg N, while the increases brought by phosphorus fertilization were between 6.42-8.83 kg/kg P (Lupu C., 2012). Lazin V.L. (2014) obtains, within the research carried out in Călăraşi County, southern Romania yields ranging from 600 to 1400 kg/ha due to nitrogen and phosphorus fertilization.
Phosphorus fertilization using doses of 60-80 kg P/ha during a 5 years research in China highlighted the assurance of a yield between 5-6 t/ha, the efficiency of phosphorus capitalization being 29%. (Xu T. et al., 2008).
Although the positive influence of fertilization on corn yield was evidenced by extensive research, this is equally influenced by environmental factors (soil type, rainfall, soil water reserve) as well as other technological factors (crop rotation, soil tillage systems, integrated pest management technologies (Marin D.I et al., 2011) generating differentiated results, which impose optimal, locally adapted fertilization strategies.
Use of fertilizers without regard to soil and climatic conditions specific to the growing area, and the use hybrids inappropriate to these conditions are the main yield limits of the corn crop (Kogbe J.O.S., Adediran J.A., 2003, Tahir M. et al., 2008). The objective of the research was to determine the influence of mineral fertilization with N and P on the yield of corn hybrids from different maturity groups and to identify those hybrids with the best adaptation to the climatic and pedologic conditions in Dobrogea (Sarichioi, Tulcea).

MATERIALS AND METHODS

The field research was located in Sarichioi, Tulcea County, within the SC Transmeteorit SRL farm. The soil specific to this area is part of the Chernozems class.
During the growing season of corn crop, the climatic data presented in Figure 1 highlight temperature values higher than the multiannual values.
Thus, we can observe that in both agricultural years there was an increase of the average temperature in May-September compared to the multiannual value specific for the area, with differences of 1.3°C in 2016 and 1.5°C in 2017.

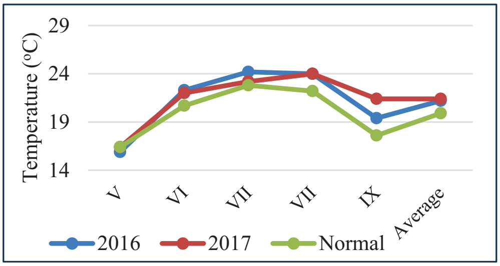

Figure 1. Temperature (°C) during maize vegetative development, Sarichioi, Tulcea County

In 2016, during the corn crop vegetative period, rainfalls of 164.9 mm were recorded, with a negative difference of 10.8 mm from the multiannual average (Figure 2). The highest rainfall values were recorded in May and June, with differences from the multiannual value of 39.3 mm and 8.3 mm, respectively. In 2017, the amount of rainfalls between May and September was 47.8 mm higher than the multi-year value (175.7 mm), with positive differences in June (38.5 mm) and July (59.8 mm).

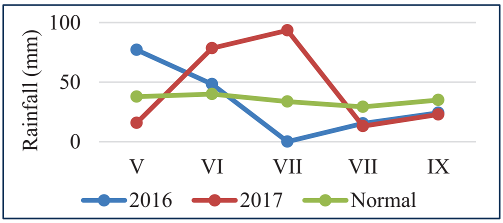

Figure 2. Rainfall (mm) during maize vegetative development, Sarichioi, Tulcea County

To achieve the research objective, a bifactorial 4 x 8 type field experience was established. The field was organized by the split plot method, with 4 replications. Within the research, the following factors were tested:
Factor A, representing the fertilization level with the following graduations:
$a_1 = N_0P_0$
$a_2 = N_{60}P_0$
$a_3 = N_{60}P_{60}$
$a_4 = N_{120}P_{60}$

Factor B, different FAO groups hybrids, with the following graduations:
b_1 = DKC 3623 (FAO 260) - early hybrid;
b_2 = P8567 (FAO 275) - early hybrid;
b_3 = DKC 4590 (FAO 350) - mid-early hybrid;
b_4 = P9903 (FAO 360) - mid-early hybrid;
b_5 = DKC 4717 (FAO 410) - mid-late hybrid;
b_6 = P0023 (FAO 400) - mid-late hybrid;
b_7 = P0412 (FAO 480) - mid-late hybrid;
b_8 = DKC 5632 (FAO 510) - late hybrid.

Corn was sown after winter wheat; soil tillage was done by plowing in autumn at a depth of 25 cm and seedbed preparation was carried out in spring at a depth of 5-6 cm.
Sowing was carried out on 13.04.2016 and 25.04.2017 at a density of 60,000 germinal seeds/ha. The crop was harvested in the second decade of September.
Fertilization was performed at the preparation of the seedbed and during vegetative development.
Weed control was carried out with Adengo 465 SC, at a dose of 0.35 l/ha applied pre-emergence and with Equip g/l at a dose of 1.7 l/ha applied post-emergence. Control of *Tanymecus dilaticollis* pests was performed using Calypso 480 SC at a dose of 90 ml/ha.

RESULTS AND DISCUSSIONS

Fertilization influence on maize yield, Sarichioi, Tulcea

Mineral fertilization with nitrogen and phosphorus resulted in yield increases statistically assured compared to the unfertilized control variant. $N_{60}P_0$ fertilization produced a minimal yield increase compared to control for all researched hybrids (Table 1), the statistically assured yield growths ranging from 0.66 t/ha for DKC 4590 (FAO 350) to 1.09 t/ha for P0412 (FAO 480). The highest yield increases, very significant in statistical terms, were generated by complex fertilization $N_{120}P_{60}$ (Table 1). Yield increases determined by 120 kg N/ha and 60 kg P/ha active substance fertilization had values between 1.19 t/ha for the hybrid P9903 (FAO 360) and 1.54 t/ha for the hybrid P0412 (FAO 480).
Due to $N_{60}P_{60}$ complex mineral fertilization, yield increased significantly, with values between 0.98 t/ha for P9903 (FAO 360) hybrid and 1.34 t/ha for P0412 hybrid (FAO 480).

Table 1. Fertilization influence on grain yield (GY, t/ha), Sarichioi, Tulcea County

Maize hybrid	NP level	Grain yield			
		GY t/ha	Diff. %	Diff. t/ha	Signf.
DKC 3623 (FAO 260)	**N_0P_0**	**6.46**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.20	111.55	0.75	**
	$N_{60}P_{60}$	7.46	115.57	1.01	***
	$N_{120}P_{60}$	7.71	107.05	1.25	***
P8567 (FAO 275)	**N_0P_0**	**6.80**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.61	111.92	0.81	**
	$N_{60}P_{60}$	7.86	115.60	1.06	***
	$N_{120}P_{60}$	8.09	106.38	1.30	***
DKC 4590 (FAO 350)	**N_0P_0**	**6.90**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.57	109.63	0.66	**
	$N_{60}P_{60}$	7.91	114.58	1.01	***
	$N_{120}P_{60}$	8.29	109.53	1.39	***
P9903 (FAO 360)	**N_0P_0**	**7.26**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.95	109.46	0.69	**
	$N_{60}P_{60}$	8.25	113.56	0.98	***
	$N_{120}P_{60}$	8.46	106.37	1.19	***
DKC 4717 (FAO 410)	**N_0P_0**	**7.05**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.89	111.88	0.84	**
	$N_{60}P_{60}$	8.17	115.77	1.11	***
	$N_{120}P_{60}$	8.35	105.83	1.30	***
P0023 (FAO 400)	N_0P_0	**7.45**	**100.00**	**Ct**	-
	$N_{60}P_0$	8.24	110.61	0.79	**
	$N_{60}P_{60}$	8.55	114.84	1.11	***
	$N_{120}P_{60}$	8.87	107.71	1.43	***
P0412 (FAO 480)	N_0P_0	**7.55**	**100.00**	**Ct**	-
	$N_{60}P_0$	8.64	114.44	1.09	**
	$N_{60}P_{60}$	8.89	117.75	1.34	***
	$N_{120}P_{60}$	9.09	105.21	1.54	***
DKC 5632 (FAO 510)	N_0P_0	**7.20**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.96	110.68	0.77	**
	$N_{60}P_{60}$	8.26	114.82	1.07	***
	$N_{120}P_{60}$	8.47	106.38	1.28	***
Hybrids average	N_0P_0	**7.08**	**100.00**	**Ct**	-
	$N_{60}P_0$	7.88	111.28	0.80	**
	$N_{60}P_{60}$	8.17	115.32	1.09	***
	$N_{120}P_{60}$	8.42	118.83	1.33	***

LSD 5% = 0.44 t/ha; LSD 1% = 0.61 t/ha; LSD 0.1% = 0.84 t/ha

*ns - not significant; * significant; ** distinctly significant; *** very significant*

Analyzing the influence of mineral fertilization on the average yield of the eight hybrids, increases recorded compared to unfertilized control were between 11.28% ($N_{60}P_0$) and 11.83% ($N_{120}P_{60}$), statistically assured for all fertilization levels (Table 1).
Yield increase achieved by applying one kg of nitrogen (N) active substance was on average 8.75 kg, ranging between 7.50 kg for P9903 and 10.75 kg for P0412 (Figure 3).

The average grain yield growth (Figure 3) per kg of phosphorus (P) active substance was 4.83 kg, the highest capitalization of a kg of phosphorus active substance was recorded by DKC 4590 (5.67 kg) hybrid, and the lowest by the hybrids P8567 and P0412 (4.17 kg).

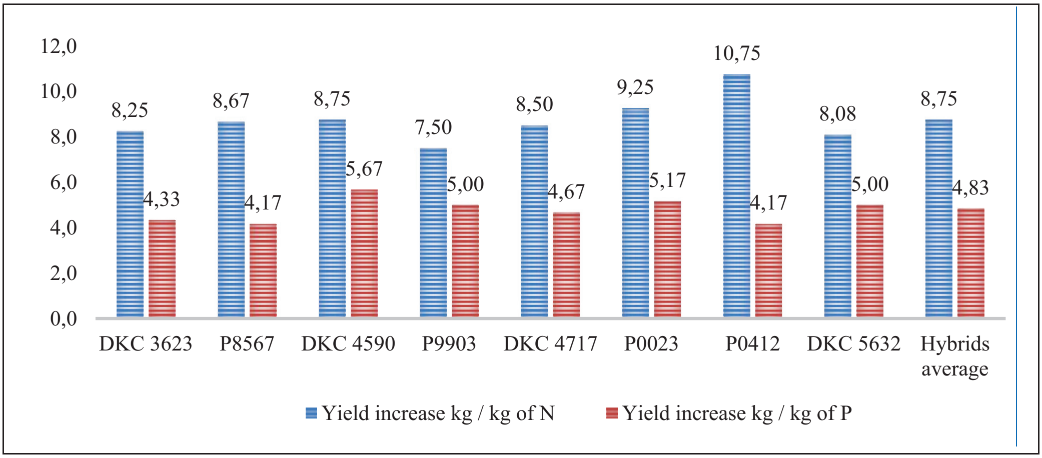

Figure 3. Yield increase kg grains per kg of mineral fertilizer active substance: nitrogen and phosphorus

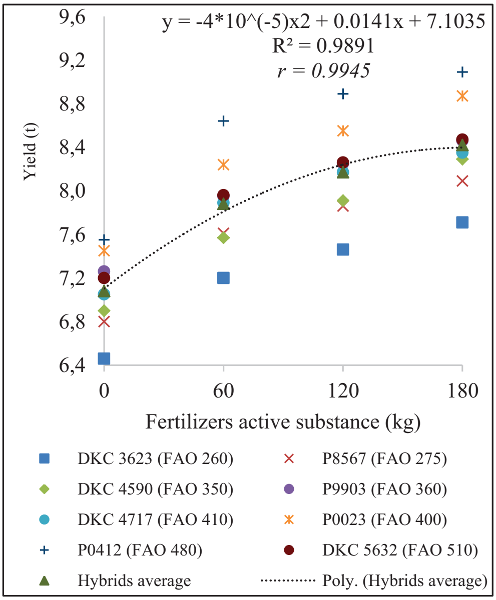

Figure 4. Graphical distribution of grain yields as influenced by doses of active substance fertilizers

For all eight hybrids data presented in Figure 4 highlights a relationship of interdependence between the grain yield and the amount of fertilizer active substance applied.

On average, under non-irrigated conditions, the correlation coefficient of 0.9945 reveals a strong dependence between yield and the amount of fertilizer the active substance applied. The value of the regression coefficient indicates that about 98.91% of the corn crop yield is provided by the applied fertilizer dose of the active substance (Figure 4).

Hybrid influence on maize yield, Sarichioi, Tulcea

The eight corn hybrids recorded an average yield of 7.08 t/ha for the non-fertilized variant (N_0P_0) and 8.42 t/ha when mineral fertilization $N_{120}P_{60}$ was (Table 2).

The lowest yield values were obtained by the hybrid DKC 3623 (between 6.46 t/ha in the unfertilized variant and 7.71 t/ha for the fertilization level $N_{120}P_{60}$) and the P0412 hybrid recorded the highest yields (between 7.55 t/ha, unfertilized and 9.09 t/ha, fertilized $N_{120}P_{60}$).

Under the influence of the hybrid, compared to the average yield the eight hybrids grain yield recorded differences were between -0.71 t/ha and 0.76 t/ha. The hybrids DKC 3623, P8567 and DKC 4590 obtained for all fertilizer levels researched lower yield values, with differences between -0.71 t/ha (at DKC 3623 fertilized with $N_{60}P_{60}$ and $N_{120}P_{60}$) and -0.13 t/ha (at hybrid DKC 4590 fertilized with $N_{120}P_{60}$).

For DKC 3623 hybrid, yield difference compared to control (average yield of the hybrids) was significantly negative, while for the other hybrids the differences recorded were not statistically assured.

Table 2. Hybrid influence on grain yield (GY, t/ha), Sarichioi, Tulcea County

NP fertilization level	Maize hybrid	Grain yield			
		GY (t/ha)	Diff. %	Diff. t/ha	Signif.
N_0P_0	DKC 3623 (FAO 260)	6.46	91.16	-0.63	oo
	P8567 (FAO 275)	6.80	95.94	-0.29	ns
	DKC 4590 (FAO 350)	6.90	97.47	-0.18	ns
	P9903 (FAO 360)	7.26	102.54	0.18	ns
	DKC 4717 (FAO 410)	7.05	99.57	-0.03	ns
	P0023 (FAO 400)	7.45	105.12	0.36	ns
	DKC 5632 (FAO 510)	7.20	101.60	0.11	ns
	P0412 (FAO 480)	7.55	106.60	0.47	*
	Average hybrids	**7.08**	**100.00**	**Ct**	**-**
$N_{60}P_0$	DKC 3623 (FAO 260)	7.20	91.38	-0.68	oo
	P8567 (FAO 275)	7.61	96.49	-0.28	ns
	DKC 4590 (FAO 350)	7.57	96.02	-0.31	ns
	P9903 (FAO 360)	7.95	100.86	0.07	ns
	DKC 4717 (FAO 410)	7.89	100.11	0.01	ns
	P0023 (FAO 400)	8.24	104.48	0.35	ns
	DKC 5632 (FAO 510)	7.96	101.05	0.08	ns
	P0412 (FAO 480)	8.64	109.62	0.76	**
	Average hybrids	**7.88**	**100.00**	**Ct**	**-**
$N_{60}P_{60}$	DKC 3623 (FAO 260)	7.46	91.35	-0.71	oo
	P8567 (FAO 275)	7.86	96.17	-0.31	ns
	DKC 4590 (FAO 350)	7.91	96.85	-0.26	ns
	P9903 (FAO 360)	8.25	100.98	0.08	ns
	DKC 4717 (FAO 410)	8.17	99.97	0.00	ns
	P0023 (FAO 400)	8.55	104.68	0.38	ns
	DKC 5632 (FAO 510)	8.26	101.16	0.09	ns
	P0412 (FAO 480)	8.89	108.84	0.72	**
	Average hybrids	**8.17**	**100.00**	**Ct**	**-**
$N_{120}P_{60}$	DKC 3623 (FAO 260)	7.71	91.61	-0.71	oo
	P8567 (FAO 275)	8.09	96.13	-0.33	ns
	DKC 4590 (FAO 350)	8.29	98.50	-0.13	ns
	P9903 (FAO 360)	8.46	100.48	0.04	ns
	DKC 4717 (FAO 410)	8.35	99.21	-0.07	ns
	P0023 (FAO 400)	8.87	105.39	0.45	*
	DKC 5632 (FAO 510)	8.47	100.67	0.06	ns
	P0412 (FAO 480)	9.09	108.01	0.67	**
	Average hybrids	**8.42**	**103.04**	**Ct**	**-**

LSD 5% = 0.46 t/ha; LSD 1% = 0.60 t/ha; LSD 0.1% = 0.78 t/ha

*ns – not significant, * significant, ** distinctly significant, *** very significant*

On the opposite side hybrids P9903, P0023, DKC 5632 and P0412 (Table 2) recorded higher yields for all fertilization levels compared to the average hybrids yield, and differences were positives with values between 0.04 t/ha (P9903 hybrid fertilized with $N_{120}P_{60}$) and 0.76 t/ha (in P0412 hybrid fertilized with $N_{60}P_0$), being significantly positive for hybrid P0412, unfertilized (0.47 t/ha) and for the hybrid fertilized P0023 $N_{120}P_{60}$ (0.45 t/ha) distinctly positive for P0412 fertilized with $N_{60}P_0$, $N_{60}P_{60}$ and $N_{120}P_{60}$ and not statistically assured for the other hybrids.

The hybrid DKC 4717 recorded yield differences between -0.07 t/ha ($N_{120}P_{60}$) and 0.18 t/ha (N_0P_0) compared to the control (Table 2), but not assured statistically.

Analyzing the influence of the maturity group on hybrids yield, the data presented in Table 3 shows that the semi-late hybrids recorded the highest yield values for all fertilization levels ranging between 7.35 t/ha for the unfertilized variant and 8.77 t/ha for $N_{120}P_{60}$ fertilization. The lowest yield values were obtained by early hybrids, with values between 6.63 t/ha (unfertilized) and 7.90 t/ha ($N_{120}P_{60}$).

Table 3. Grain yield (t/ha) differences among maturity groups, Sarichioi, Tulcea County

NP fertilization level	Grain yield (t/ha)				Differences among maturity groups (t/ha)****					
	Early hybrids (b_{1-2})	*Mid-early hybrids (b_{3-4})*	*Mid-late hybrids (b_{5-7})*	*Late hybrids (b_8)*	b_{3-4} - b_{1-2}	b_{5-7}-b_{1-2}	b_8-b_{1-2}	b_{5-7}-b_{3-4}	b_8-b_{3-4}	b_8-b_{5-7}
N_0P_0 (a_1)	6.63	7.08	7.35	7.20	0.46^{*}	0.72^{**}	0.57^{**}	0.27^{ns}	0.11^{ns}	-0.15^{ns}
$N_{60}P_0$ (a_2)	7.40	7.76	8.26	7.96	0.36^{ns}	0.85^{***}	0.56^{**}	0.50^{*}	0.21^{ns}	-0.29^{ns}
$N_{60}P_{60}$ (a_3)	7.66	8.08	8.54	8.26	0.68^{**}	1.13^{***}	0.86^{***}	0.46^{*}	0.18^{ns}	-0.27^{ns}
$N_{120}P_{60}$ (a_4)	7.90	8.37	8.77	8.47	0.97^{***}	1.37^{***}	1.07^{***}	0.40^{*}	0.10^{ns}	-0.30^{ns}
LSD 5% = 0.39 t/ha; LSD 1% = 0.54 t/ha; LSD 0.1% = 0.74 t/ha										

*ns - not significant, * significant, ** distinctly significant, *** very significant; ****differences were calculated among the average yield of the hybrids belonging to a FAO maturity group*

Compared to early hybrids (Table 3), mid-early hybrids recorded yield increases of 0.36 t/ha (fertilization level $N_{60}P_0$) and 0.97 t/ha (fertilization level $N_{120}P_{60}$), statistically assured for the unfertilized variant and for the use of $N_{60}P_{60}$ and $N_{120}P_{60}$.

Compared to early hybrids both late and mid-late hybrids recorded higher yields with distinctly significant differences of 0.72 t/ha and 0.57 t/ha respectively for the unfertilized variant and very significant differences between 0.85 t/ha and 1.37 t/ha for the variants where mineral fertilization with nitrogen and phosphorus was applied (Table 3).

Compared with mid-early hybrids, semi-late hybrids recorded higher yields with statistically significant differences between 0.40 t/ha and 0.50 t/ha obtained where mineral fertilization was applied (Table 3).

The yield of late hybrids was higher than that recorded by mid-early hybrids, and increased with values between 0.10 t/ha and 0.21 t/ha were not statistically assured. Compared to mid-late hybrids, the late ones recorded lower yields, with statistically significant differences, ranging from 0.15 t/ha to 0.30 t/ha (Table 3).

CONCLUSIONS

The results obtained during the period 2016-2017 under the conditions of Dobrogea area (Sarichioi locality, Tulcea County) reveal the positive influence of mineral fertilization with nitrogen and phosphorus on the grain yield of the eight corn hybrids.

Under the influence of fertilization, the yield of the eight corn hybrids recorded statistical assured growths compared to the unfertilized control variant, ranging from 0.66 t/ha for DKC 4590 fertilized with $N_{60}P_0$ to 1.43 t/ha for P0023 fertilized with $N_{120}P_{60}$.

Nitrogen fertilization resulted in an average yield increase of 8.75 kg grains per kg N, and phosphorus fertilization provided an average increase of 4.83 kg grains per kg of P.

The highest yields were recorded by the mid-. late hybrid P0412, with values between 7.55 t/ha (unfertilized) and 9.09 t/ha ($N_{120}P_{60}$).

Under the pedologic and climatic conditions of Sarichioi, Tulcea county, the highest yields are recorded by the mid-late hybrids, with values between 7.35 t/ha (unfertilized) and 8.7 t/ha ($N_{120}P_{60}$).

REFERENCES

Al-Naggar, A.M.M., Shabana, R.A., Atta, M.M., Al-Khalil, T.H. (2015). Maize response to elevated plant density combined with lowered N-fertilizer rate is genotype-dependent. *The Crop Journal, 3*(2), 96–109.

Berca, M., Buzatu, C.S. (2011). Maize answer modelling at nutrition with nitrogen and phosphorous within the Romanian space, *Scientific Papers. Series A. Agronomy*, 54.

Gehl, R.J., Schmidt, P.P., Maddux, L.D., Gordon, W.B. (2005). Corn Yield Response to Nitrogen Rate and Timing in Sandy Irrigated Soils. *Agron. J.*, *97*, 1230–1238.

Gidea, M., Ciontu, C., Sandoiu, D.I., Penescu, A., Schiopu, T., Nichita, M. (2015). The Role of Rotation and Nitrogen Fertilization Level upon the Economic Indicators at Wheat and Corn Crops in Condition of a Long-Term Experience. *Agriculture and Agricultural Science Procedia*, *6*, 24–29.

Hăbădan (Zaharia) V.G. (2009). Contribuții la optimizare nutriției cu azot la cultura porumbului Teză de doctorat, București.

Kogbe, J.O.S., Adediran, J.A. (2003). Influence of nitrogen, phosphorus and potassium application on the yield of maize in the savanna zone of Nigeria. *African Journal of Biotechnology, 2*(10), 345–349.

Lazin, V.L. (2014). Research on the behavior of some corn hybrids under the influence of technological components in the context of sustainable agriculture. *Lucrări Ştiinţifice, Seria Agronomie, 57(1).*

Lupu, C., Naie, M., Pomohaci, T. (2012). Efectul fertilizării de lungă durată cu fosfor şi azot asupra producţiei de boabe la porumb în condiţiile S.C.D.A. Secuieni. *I.N.C.D.A. Fundulea, LXXX.*

Marin, D.I., Mihalache, M., Ciontu, C., Bolohan, C, Ilie, L. (2011). Influence of soil tillage of pea, wheat and maize crop in the Moara Domneasca-Ilfov area. *5th International Symposium-Soil Minimum Tillage System*, 111–118.

Miao, Y., Mulla, D.J., Robert, P.C., Hernandez, J.A. (2006). Within-field variation in corn yield and grain quality responses to nitrogen fertilization and hybrid selection. *Agronomy Journal*, *98*(1), 129–140.

Pettigrew, W.T. (2008). Potassium influences on yield and quality production for maize, wheat, soybean and cotton. *Physiologia Plantarum*, *133*(4), 670–681.

Prisecaru, G., Rujescu, C., Sala, F. (2017). The influence of nitrogen fertilization on the productivity elements on corn cobs. *Journal of Horticulture, Forestry and Biotechnology, 21*(3), 44–51.

Ran, H., Kang, S., Li, F., Du, T., Ding, R., Li, S., Tong, L. (2017). Responses of water productivity to irrigation and N supply for hybrid maize seed production in an arid region of Northwest China. *Journal of Arid Land*, *9*(4), 504–514.

Sangoi, L., Ender, M., Guidolin, A.F., Almeida, M.L.D., Konflanz, V.A. (2001). Nitrogen fertilization impact on agronomic traits of maize hybrids released at different decades. *Pesquisa Agropecuária Brasileira*, *36*(5), 757–764.

Tahir, M., Tanveer, A., Ali, A., Abbas, M., Wasaya, A. (2008). Comparative yield performance of different maize (*Zea mays* L.) hybrids under local conditions of Faisalabad-Pakistan. *Pakistan Journal of Life and Social Sciences*, *6*, 118–120.

Xu, Tang, Jumei, Li, Yibing, Ma, Xiying Hao, Xiuying, Li. (2008). Phosphorus efficiency in long-term (15 years) wheat–maize cropping systems with various soil and climate conditions.

Yang, S., Li, F., Wang, P, Malhi, S.S., Liu, S. (2004). Long-Term Fertilization Effects on Crop Yield and Nitrate-N Accumulation in Soil in Northwestern China.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

OPPORTUNITIES FOR CULTIVATION OF MEDICAL CANNABIS (*Cannabis sativa* L.) IN GREECE

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Abstract

The cultivation of Cannabis sativa L. for medical uses was legalized in Greece in March of 2018, ending the 90 years ban. The cannabis regulations in Greece attempt to protect the public health and safety by controlling for quality in a reactive manner. The necessary knowledge and know-how for this new crop were acquired by the Laboratory of Agronomy of the Agricultural University of Athens. In this review, the Greek legislation and licensing procedure are briefly presented. The essential relevant steps needed for making a production line are two. The installation permit for the production unit is the first step and then on the investment services sought to establish the conditions under which authorized investment. In the case of greenhouse facility, considering the crop requirements (temperature and photoperiod), the cultivation is suggested for Southern and Central Greece regions. The production process flow is endless during the year, and the duration of the biological cycle is 13-14 weeks. Finally, the required mechanical equipment, human resources, and final products for 1-hectare production with all the necessary conditions to cover GMPs are described.

Key words: *cannabis, Greek legislation, location criteria, medical use, production line.*

INTRODUCTION

1. Historical review

The origin of word cannabis is from Greek kannabis (κάνναβης or kánnavis). Herodotus, the Greek Historian, in 5th century B.C.E., described extensively about the Scythians and their ways of using cannabis (Grimmer, 2013). Later, circa 40-90 C.E., the Greek physician, pharmacologist and botanist, Pedanius Dioscorides wrote about the medical qualities of cannabis and mentioned about treating pains of the ear. During the declines of the Greek Empire, the knowledge pass to European and Byzantines through Rome and Dioscorides texts. In 1855, in a Bavarian text about pharmacology, cannabis was mentioned as a plant which was indigenous in the East and Persia and was cultivated in Europe and Greece. The big gab from ancient time was closed between 1870 and 1880, while the Piraeus port was developed and constituted the major trade depot in the Aegean region (Abel, 1980). During this period, immigrants from Egypt, Cyprus, and countries from the Middle East lived moved to the port because of work, and they brought their recreational habit, which was hashish (Stefanis et al., 1975).

In 1906, the Greek government introduced customs restrictions and tax on cannabis cultivation, accepting British pressures, extinguishing the Greek grapes market.

In 1919, the Treaty of Versailles banned the cultivation and marketing of hashish due to addiction and harmful effects on health. Greece has enacted the hash trade ban for ten years until 1st January 1936 (Papadopoulos, 1974).

2. Chemotype of cannabis

Cannabis components are many, while from the Cannabinoids, the well-known for their healing properties are cannabidiol (CBD) and tetrahydrocannabinol (Δ^9-THC).

Some of the cannabinoids come from raw cannabis, and others are produced after heating. They can be found more in the bud of blooming plant or before blooming. There is also the possibility to appear only after drying. However, in cannabis, there are also terpenoids that give a characteristic fragrance to the plant.

There are three cannabis chemotypes: the drug type, the fibre type and the intermediate type, which are easily distinguished by their Δ^9-THC content that represents the psychoactive constituent of the cannabis plant. The drug type is characterized by a high Δ^9-THC content (>2%), high in resin and is mainly found in good climatic conditions areas and sufficient sunshine. The fibre type has a low Δ^9-THC content (<0.5%) as opposed to high CBD content (>0.5%) and is cultivated for paper, cloth and litter production. The Δ^9-THC content in the fibre type doesn't exceed 0.03%. Intermediate type characterized by the high content of Δ^9-THC and CBD (>0.5%) (Kinghorn et al., 2017).

3. Legislation in Greece

In Greece, the medical cannabis cultivation is governed by the law 4523/2018, which was drawn up in 2018. For the granting of approval for installation and authorization, the applicant must submit documents for all stages (production, excise, transport, storage, supply of raw materials and substances of cannabis varieties of *Cannabis sativa* L. with a tetrahydrocannabinol (THC) content greater than 0.2%, the production, import and marketing of propagating material, processing unit, processing and production of finished medicinal products of cannabis). The final product will be purchased either in the state monopoly or exported. The supporting documentation for the installation approval and operating license is valid for 5 years. Any amendments require new supporting documents. On the basis of legislation, the cultivation will take place under enclosed conditions (greenhouse or indoor), the production area must be fenced off. In addition, there will be private security guards. The quantity of propagation material introduced is limited up to 30 kg of seed per hectare. Any infringements and incomplete data due to refusal or concealment are subjected to sanctions. These sanctions have correction deadline to withdrawal of approval (Hellenic Republic, 2018a, b).

The starting material for the manufacture of a herbal medicinal product can be a medicinal plant, a herbal substance or a herbal preparation.

Herbal medicinal product should be provided with the herbal substance data. For the purpose of ensuring the consistency of the finished product, particular attention should be paid in the agricultural production, including the selection of seeds, cultivation, and harvesting conditions. In order to succeed it, instructions are given in the Herbal Medicinal Product Committee (HMPC) guidance document entitled "Guideline on Good Agricultural and Collection Practice for starting materials of herbal origin".

There are five activities that receive the application of Good Practice. The guidance document of Good Agricultural and Collection Practice (GACP) provides the appropriate instructions for cultivation, collection, and harvesting of plants, algae, fungi, lichens, and collection of exudates. In addition, it includes the activity of cutting, drying of plants, algae, fungi, lichens, and exudates. The guidance document of Good Manufacturing Practice (GMP) is followed by 4 activities. Part II of the GMP guide is responsible for the proper operation of cutting, drying of plants, algae, fungi, lichens and exudates, expression from plants and distillation and comminution, processing of exudates, extraction from plants, fractionation, purification, concentration or fermentation of herbal substances. Part I of the GMP guide provides all the instructions that should be followed by the above-mentioned activities, including the further processing into a dosage form and packaging as a medicinal product.

Depending on the herbal material (active substance, intermediate or a finished product), the GMP classification is applied differently. Manufacturer assumes the proper choice of GMP classification for the medicinal product, additionally is responsible that the activities follow the marketing authorization-registration. GACP is applicable for the initial steps in the field, while GMP for further steps.

In that case, the quality of the product within the approved specifications should be ensured that it is acceptable that the activity of expression from plants and distillation will take place in the field and will be an integral part of harvesting, given that cultivation is in accordance with GACP. These situations should be regarded as exceptional and justified in the relevant marketing authorization - registration documentation. GMP principles undertake the proper operation of the activities that carried out in the field. Furthermore, GMP inspections occur during the activities by the regulatory authorities in order to evaluate compliance (European Commission, 2008).

PRODUCTION PROCESS/LINE

1. Selection of the region, location of the greenhouse cannabis production

Cannabis cultivation in a greenhouse is based on control of the environment in such a way as to provide the conditions that are most favorable for maximum yield. The location of the greenhouse and its orientation are factors that greatly influence the production potential. Solar radiation during winter, temperature, wind, and soil are needed to evaluate the suitability of a region (Mavrogiannopoulos, 2017). Solar radiation is indispensable for plant growth as well as for heat. Sunlight offers greenhouse plants optimal full-spectrum light, yet the day length is not enough. A region with high solar radiation can significantly reduce the use of technical light and therefore the cost. With regard to temperature, there are many areas in Greece where there are relatively high minimum temperatures in the winter and low maximum in the summer. Moreover, there is the option of areas, where the snow accumulation could be avoided because it is dangerous for construction. A prerequisite parameter is the water availability and the heavy metal contamination in water. Soil quality is not a significant parameter for medical cannabis cultivation as the plants are grown in pots. Wind direction and speed in a region could cause significant energy loss in heated greenhouses and can sometimes be a dangerous factor for the construction. For the reasons mentioned above, greenhouse orientation must be carefully considered during the installation. Also, the greenhouse location should be 1000 meters away from schools (Hellenic Republic, 2018b).

Greece is characterized by a predominant Mediterranean climate, with mean annual temperatures ranging from 12 to 19°C. Although, there is a remarkable range of micro-climates and local variations, most of the regions of Greece meet the demands of cannabis cultivation. Considering the crop requirements (temperature, photoperiod or light duration) in a case of greenhouse facility, the suggested areas include the whole region of Central, West, and South mainland of Greece. More particularly, regions of Thessaly, Central Greece, and Peloponnese could be preferred.

2. Production process

The production process is based on clone plants and until now the pharmaceutical final products *Cannabis sativa* L. will be (a) Dried inflorescence of *Cannabis sativa* L., and (b) *Cannabis sativa* L. (THC Oil) pharmaceutical oil which are pesticide free. For each stage, *Cannabis sativa* L. plants will be coded for safety and reference to yield (Mediavilla et al., 1998).

2.1. Growth and maintaining of mother plants

The cultivation begins with growing or introduction of the mother plants which are: i) feminized cannabis seeds, in order to be developed the first mother plants or, ii) already developed mother plants which can be imported. Mother plants should be healthy and have plenty of stems so that if one is removed, it will not harm the plant. In the first instance, seeds are imported and planted in biodegradable jiffy pots. Temperature, moisture air and light condition must be correct for seeds to germinate (Hall et al., 2014). Seed germination, depending on cannabis strain as the conditions in chamber room are standard. Seedlings should be exposed to cool fluorescent light (18 h photoperiod) for the initial vegetative growth (Clarke & Watson, 2002). When the plants have 2-6 leaves, they are transplanted. The growth of mother plants will last 8-9 weeks and will only take place once, at the beginning of production. The growth of plants from feminized seeds will be done in a chamber at 20 hours of light/day, 50-70% RH and 24-26°C. Great care must be taken to prevent mother plants from flowering because the clones use stored carbohydrates to grow a new root system (Kinghorn et al., 2017). In the second case, the already developed plants are kept in the controlled chambers as mentioned above. The mother plants must be stored in chambers with the appropriate equipment in order to maintain the desired conditions. In order to ensure that a mother plant provides vigorous cuttings, the fertilization schedule must be modified to suit long living of them, and the mother plant's age must be noticed.

2.2. Growth of clone plants

Since the mother plants will be at the ideal stage, then the second stage of the production process, propagation, is followed. The plant clones are transported and stored in a suitable place to form a rhizome and a completely new plant. There are two methods for propagation, vegetative propagation or tissue culture. With both methods, the genetically identical version of the donor plant is achieved. By using this method, the crop genetic uniformity will be developed, provided that the propagating conditions are controlled (Clarke & Watson, 2002). Tissue culture propagation constitutes a common way in plant science research, and it is successfully applied in the field of cannabis cultivation. It is a complicated method than vegetative propagation; however, the potential value of this method is appreciated by producers due to high volume management of numerous varieties and strains. An apical branch is about 6-10 cm containing at least two branches, as at least one node must be covered by soil. This branch is cut at a 45° angle straight below the node and dipped in distilled water immediately. Then, the first 2 cm of the cut is dipped into a root hormone and is planted in pots (5 x 5 cm). Plants are regularly irrigated and kept under controlled environmental conditions. In particular, the growth of clone plants requires more than 18 hours of light/day, 80-90% RH, and 24-26°C. The growth of clone plants lasts for up to 4 weeks. The rooting begins in two to three weeks, and this is followed by transplantation to larger pots (e.g. 30 x 30 cm) after six weeks.

2.3. Vegetative growth of clone plants

When plants acquire the appropriate root system, they are transplanted into 20-liter pots and transported to the greenhouse where they remain for 4-5 weeks at steady conditions photoperiod 18-24 hours of light/day, 50-70% RH and 24-29°C.

2.4. Flowering plant growth

At the flowering stage, the plants are transported to a greenhouse with 12 hours of light/day, 50% RH and 20-25°C for 5 to 6 weeks. The plant clones remain there until harvesting. By cutting the inflorescences, the plant crop residues are destroyed (burning or composting) and the soil from the pots is sunburned in a constructed area outside the greenhouse.

2.5. Harvesting

After the flowering stage has been completed, the plants are cut at the base of the overhang and placed in large basins to be transported to the site where the central leaf-foliar and inflorescence separation will take place. When 75% of hair- like structures turn brown, the flowers are ready to harvest (UNODC, 2009). Pots with soil and root system are transported out of the greenhouse, into the sun-cleansing area. In the area of solar decontamination, the contents of the pots are dispersed, the roots are transported to the composting site, and the soil remains there.

2.6. Central leaf-foliar and inflorescence separation

With the stem removal machine, the inflorescences and leaves are also gently removed from the stem. Once the plant has been cleaned down, the branches are separated from the stem, and then the branches are passed through the holes.

2.7. *Trimming*

Trimming constitutes one of the most important aspects of cannabis cultivation and production. The breech leaf is removed from inflorescence by special machines. The trimming machine can save a significant amount of money compared to paying a crew of hand trimmers, especially for large operations. Trimmer Machine will be used for trimming, as it offers the ability to adapt to more inflorescence conditions (i.e., strains, conditions, moisture levels, flower density etc.). A wet trimmer has the advantage to keep the moisture of flower, and thus the context of flower does not be destroyed. In case of the dry trimmer, flowers are dried up to 8-9% humidity.

2.8. Dehydration of inflorescence

The inflorescences after trimming are deposited for 12 days in a drying chamber at 40-45°C. Generally, it is recommended that the drying temperature maintained between 40-45°C because many terpenoids (molecules that are partly responsible for the psychoactive effects but also largely responsive to the odor of the plant) evaporate at temperatures above 45°C. Unplanned drying moisture levels should generally be between 45-55%. Higher moisture levels are associated with mold appearance. Lower moisture levels tend to dry the products very quickly. If the plant material dries very quickly, chlorophyll will fail to convert, which will lead to suboptimal the average organoleptic characteristics (Tang et al., 2015; Kinghorn et al., 2017).

2.9.A.1. Packaging of dried inflorescence of Cannabis sativa L.
Dry inflorescences are packed in special 500 g or 1000 g vacuum packs or FDA-approved polyethylene bags placed in sealable fiber drums (short term: 18-20°C; long term: 10°C) (Kinghorn et al., 2017).

2.9.A.2. Storage of dried inflorescence of Cannabis sativa L.
The inflorescences are stored in a dark warehouse, 30% H and 22°C.

2.9.B.1. Extraction of pharmaceutical oil of Cannabis sativa L. *(THC Oil)*
Liquid cannabis is a concentrated liquid extract of either herbal cannabis material or cannabis resin. The reason that liquid cannabis is produced is to concentrate the psychoactive ingredient THC. Immediately after drying, they are placed in the extraction machine. The extraction process is done with CO_2. This process is also referred to as Extract Liquid and is today a popular technology for fast and non-transmissible extraction in the food industries and pharmaceutical industries. It is important that in the extraction machine are also placed the waste generated during the trimming process and thus reduces the production losses.

2.9.B.2. Packaging of pharmaceutical oil of Cannabis sativa L. *(THC Oil)*
The oil is packed in 10, 50, 150, 250 and 500 g dark glass oil packs.

2.9.B.3. Storage of pharmaceutical oil of Cannabis sativa L. *(THC Oil)*
The packaged *Cannabis sativa* L. pharmaceutical oil (THC Oil) is stored in a dark warehouse, 30% RH and 22°C. A major concern when storing *C. sativa* is the stability of many of the cannabinoids, e.g. the optimum Δ^9-THC content. Degradation of Δ^9-THC is negligible during processing, especially when the material is well-dried and sealed; however, it is still extremely sensitive to oxygen and UV light, and slow degradation occurs during room temperature storage through oxidation to cannabidiol (CBD). Also, Δ^9-THC readily converts to (-)-Δ^8-*trans*-tetrahydrocannabinol (Δ^8-THC) under thermodynamic control. Therefore, the preferred conditions for long-term storage are low temperature and absence of light (Kinghorn et al., 2017).

THE ENVIRONMENT OF THE PLANT AND THE MECHANICAL EQUIPMENT

1. The above-ground part and its environment
In addition to the proper construction and location of the greenhouse, which they are significant for a successful production, it is crucial to have the equipment, which allows the precise regulation of environmental factors that influence the growth of plants, but primarily they provide GMPs. Additionally, the variation of Δ-9-tetrahydrocannbinol in cannabis products is high, and be attributed in climatic and growing conditions (Kinghorn et al., 2017). The equipment of a greenhouse for medical cannabis consists of devices for artificial lighting, shading, sun shading curtains, heating, CO_2 enrichment, irrigation, nutrient solution disinfection, and automation. The choice between equipment with similar capabilities and distinctive features should be based on the duration of its use, operating costs and best economic result.

1.1. Duration of light
An important factor for continuous production is light. For medical cannabis cultivation, natural light is not enough, and it often requires artificial light to supplement illumination up to 20 h per day at a density of 120 μmol m^{-2} s^{-1} and above. Flowering is affected by the night length, and often it is necessary to the night length falls below their critical photoperiod, when the plant is during this particular stage. For shading, blackout curtains are used. The growth of plants is affected by:

a. Radiation spectrum (wavelength), for photosynthesis, photoreduction and photomorphogenesis
b. Intensity, for photosynthesis, photomorphogenesis and phototropism, and
c. Duration, for photosynthesis

The wavelength of 400-700 nm (Visible light) is important for cannabis and its photosynthesis. Artificial lighting is used in different ways. In the mother room, there is a complete replacement of the lighting while in the vegetative and flowering stage the light is complementary, in periods with low daylight. While the total hours required for each stage of development are stable, the quantity and range of artificial light required is determined by various factors such as daytime, sunshine, latitude and weather, and, finally, greenhouse permeability light (Albright et al., 2000).

It has been demonstrated that the photosynthetic ability of plants in light correlates better with the number of photons (mol m^{-2} s^{-1}) than with power units (Wm^{-2}) (McCree, 1972). Thus, artificial light will be substituted by High Pressure Sodium lamps (HPS), which are efficient lamps in the 400-700 nm (100 lm W^{-1}) range. Light intensity was set to ~450 µmol m^{-2} s^{-1} measured from the canopy top (de Wit, 1965). The emission spectrum is very concentrated in the yellow-orange-red range (500-650 nm) but is quite low in the blue range (450-500 nm). High Pressure Sodium lamps are widespread in greenhouse cultivations and are used during the flowering cycle of crops such as medical cannabis cultivation (Critten, 1993). The cost of artificial light is quite high, and it costs both in the initial installation and the duration of electricity consumed.

1.2. Ventilation

In order to ensure uniform fresh air in greenhouse, a ventilation system is preferred to keep stable conditions. Hot air is extracted, and appropriate filters introduced fresh air from the outside, and, as a result, the temperature in the greenhouse is not increased. In Greek conditions, the needs for ventilation are high from spring to late autumn (Carpenter & Willis, 1959).

Ventilation must be dynamic (or mechanical), i.e., the interior and exterior pressure differences greenhouse will be created by mechanical means to ensure stable conditions and no problems with GMP's. Ventilation is a critical control point because it depends on the reduction of the high temperature of the greenhouse due to the high intensity of the solar radiation. In a greenhouse of medical cannabis, it is more important as there is artificial light. Ventilation also exhausts vapors due to the transpiration of plants and offers full exploitation of CO_2 (Ganguly & Ghosh, 2011).

For the purpose of creating the homogenous conditions and keeping a controlled environment during cold/humid hours, the ventilation may not be sufficient to a lower temperature inside the greenhouse. On the other hand, high air velocities in the greenhouse result in excessively high transpiration with negative effects on cannabis clones growth.

1.3. Heating

Heat and consequently temperature have an effect on the growth and production of cannabis clone plants. Temperature affects photosynthesis, respiration etc. (Walker et al., 1982). It is also a factor that has its effect on the production cost in the greenhouse after artificial light. A greenhouse for medical cannabis belongs to fully heated greenhouses that offer the optimum temperature.

During the day, solar radiation is the main source of energy for heating (Kampkes et al., 2000). On a cloudless day or early in the morning or on a day with low outside temperatures, sunlight is not enough, and the room temperature is below the desired levels.

Cannabis production must not be affected by temperature drops because at the harvest stage the inflorescences will be at a different stage, so they cannot be harvested at the same time. For heating in a cannabis greenhouse, central heating with hot water or air heaters will be used to complete the air distribution to achieve a uniform distribution of air (Nederhoff, 2006).

Temperature regulation enables production planning, quantity increase and improved product quality throughout the year, and the production of pharmaceutical cannabis is important as the production flow is continuous. Moreover, stationary humidity reduces plant diseases and improve buds quality (Nederhoff, 2006).

1.4. Humidity control

The humidity is one of the determinants of the greenhouse environment. It usually tends to be high due to the evaporation of the crop. The management of humidity belongs to environmental control strategies (Labidi et al., 2017). However, humidity encourages diseases and other problems in a growing environment, while the use of pesticides is not allowed in medical cannabis cultivation.

1.5. Enrichment with carbon dioxide

The rate of photosynthesis affected not only by the availability of light and carbon dioxide (CO_2) but also by the ability of the photosynthetic mechanism that engages the CO_2. Enriching the greenhouse with CO_2 pointed to enhance the production of the crop through the enrichment of the greenhouse atmosphere (Akilli et al., 2000).

The enrichment of the greenhouse with CO_2 gas should be done during the day, when, the photosynthesis is done, the windows are closed, and the ventilators are off (Chalabi & Zhou, 1997). In particular, ventilation should begin one hour after dawn, and continue until sunset. It is a necessary element for producing inflorescences throughout the year (Heij & van Uffelen, 1984). The enrichment method commonly used in Greece is the liquid CO_2 evaporation, which is in a high-pressure tank and distributed in the greenhouse via a series of pressure regulating valves (Mavrogiannopoulos, 2017).

2. The underground part and its environment

2.1. Irrigation, water and oxygen

The water along with oxygen are the most important factors in the root environment. The irrigation system is fully automated with a drip method which offers the best control over the amount of water. It is the most widespread watering system in closed-type crops. Black plastic tubes of 12-20 mm in diameter are used on which drippers are installed or incorporated. The tubes are usually placed on the ground surface, one for each plant line or one for two plant lines. In the second case, from the center tubes start very thin tubes of 1-2 mm diameter ending in each pot. The end of the tube is secured to a specific pile close to the plant (Kim et al., 2015).

2.2. Fertilization - pH

The fertilization of the medical cannabis crop will be done along with irrigation. The optimum substrate pH for the plant is from 6.5 to 7.2 (Papastylianou et al., 2017). In hydroponic growth, the nutrient solution is best at 5.2 to 5.8, making cannabis well suited to hydroponics, and thus indoor production, because this pH range is hostile to most bacteria and fungi. (UNODC, 2009).

CONCLUSIONS

A few years ago, any discussion of medical cannabis was kept under wraps. Today, the medical cannabis cultivation is a great opportunity. The cannabis industry has many strengths such as human resources needs. For one-hectare productive unit, around 50 scientific or technical staff is needed. Additionally, new medicines will be produced to manage epilepsy, depression or chronic pain. This new industry for Greece encourages new businesses for product design. The main lack of the Greek market is the familiarization of people with this kind of medical products. Until now, for the approval granting for construction, 32 application forms have been submitted. In the next months, more application forms are expected as the consumers believe in the health benefit of cannabis as a medicine. Greece has great prospects for growth and build of a medical cannabis valley.

REFERENCES

Abel, E.L. (1980). *Marihuana: The First Twelve Thousand Years*. USA: Springer.

Akilli, M., Osmerzi, A., Ercan, N. (2000). Effect of CO_2 enrichment on yield of some vegetables grown in greenhouse. *Acta Horticulturae*, *534*, 231–234.

Albright, L.D., Both, A.J., Chiu, A.J. (2000). Controlling greenhouse light to a consistent daily integral. *Transactions of the ASAE*, *43*(2), 421–431.

Carpenter, W.J., Willis, W.S. (1959). Comparisons of evaporative fan-and-pad and high-pressure mist systems for greenhouse cooling. *Proceedings of the American Society for Horticultural Science*, *74*, 711–718.

Chalabi, Z.S., Zhou, W. (1997). Optimization of CO_2 enrichment in greenhouse constrained by limited CO_2 supply and CO_2 dynamics. *IFAC Proceedings*, *30*(26), 109–112.

Clarke, R.C., Watson, D.P. (2002). Botany of Natural *Cannabis* Medicines. In: F. Grotenhermen, and E. Russo. *Cannabis and Cannabinoids: Pharmacology,* *toxicology, and therapeutic potential* (pp. 3–14). New York, USA: Haworth Integrative Healing Press.

Critten, D.L. (1993). A review of light transmission into greenhouse crops. *Acta Horticulturae*, *328*, 9–32.

de Wit, C.T. (1965). Photosynthesis of leaf canopies. Agricultural Research Report No. 663. PUDOC, Wageningen, The Netherlands.

European Commission (2008). Enterprise and Industry Directorate. Manufacture of Sterile Medicinal Products. In: EudraLex. The Rules Governing Products in the European Union. Volume 4. EU Guidelines to Good Manufacturing Practice. Medicinal Products for Human and Veterinary Use. European Commission, Brussels, Belgium, 2008. Annex 1.

Ganguly, A., Ghosh, S. (2011). A review of ventilation and cooling technologies in agricultural greenhouse application. *Iranica Journal of Energy and Environment*, *2*(1), 32–46.

Grimmer, B. (2013). 10,000 Years: An etymologically guided history of Cannabis. 2013 PAT (Phi Alpha Theta) Northwest Conference. USA: Oregon.

Hall, J., Bhattarai, S.P., Midmore, D.J. (2014). Effect of industrial hemp (*Cannabis sativa* L.) planting density on weed suppression, crop growth, physiological responses, and fibre yield in the subtropics. *Renewable Bioresources*, *2*(1). doi:10.7243/2052-6237-2-1.

Heij, G., van Uffelen, J.A.M. (1984). Effect of CO_2 concentration on growth and production of glasshouse vegetable crops. *Acta Horticulturae*, *148*, 591–595.

Hellenic Republic (2018a). Provisions for the production of final products of medical cannabis and other provisions. Law No. 4523. Government Gazette, Series I, No. 41, 7-3-2018.

Hellenic Republic (2018b). Terms and conditions for cultivating and processing pharmaceutical cannabis. Decision No. οικ. 51483/700/Φ.15 Government Gazette, Series II, No. 1692, 15-5-2018.

Kampkes, F.L.K., Van de Braak, N.J., Bakker, J.C. (2000). Effect of heating system position on vertical distribution of crop temperature and transpiration in greenhouse tomatoes. *Journal of Agricultural Engineering Research*, *75*(1), 57–64.

Kim, H.J., Lee, D.H., Ahn, S.W., Kim, W.K., Hur, S.O., Chol, J.Y., Chung, S.O. (2015). Design and testing of an autonomous irrigation controller for precision water management of greenhouse crops. *Engineering in Agriculture, Environment and Food*, *8*(4), 228–234.

Kinghorn, A.D., Falk, H., Gibbons, S., Kobayashi (2017). *Phytocannabinoids: Unraveling the Complex Chemistry and Pharmacology of Cannabis sativa*. Progress in the Chemistry of Organic Natural Products, 103. Switzerland: Springer.

Labidi, A., Chouchaine, A., Mami, A.K. (2017). Control of relative humidity inside an agricultural greenhouse. 18th International Conference on Sciences and Techniques of Automatic Control and Computer Engineering (STA), Monastir, 109–114. doi: 10.1109/STA.2017.8314955.

Mavrogiannopoulos, G.N. (2017). *Greenhouses*, 4th editions. Athens, Greece: Stamoulis Publications (in Greek).

McCree, K.J. (1972). The action spectrum, absorptance and quantum yield of photosynthesis in crop plants. *Agricultural Meteorology*, *9*, 191–216.

Mediavilla, V., Jonquera, M., Schmid Slembrouck, I., Soldati, A. (1998). Decimal code for growth stages of hemp (*Cannabis sativa* L.). *Journal of the International Hemp Association*, *5*(2), 68–72.

Nederhoff, E. (2006). Closed greenhouse and heat producing greenhouse. *Commercial Grower*, *10*, 67–69.

Papadopoulos, D.N. (1974). *The Cannabis*, 2nd edition. Athens, Greece: The Ministry of Rural Development and Food (in Greek).

Papastylianou, P., Kakabouki, I., Travlos, I. (2017). Effect of Nitrogen Fertilization on Growth and Yield of Industrial Hemp (*Cannabis sativa* L.). *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, *46*(1), 197–201.

Stefanis, C., Ballas, C., Medianou, D. (1975). Sociocultural and epidemiological aspects of hashish use in Greece. In: V. Rubin, *Cannabis and Culture*. The Hague: Mouton.

United Nations Office on Drugs and Crime (UNODC) (2009). Recommended methods for the identification and analysis of cannabis and cannabis products. Manual for use by National Drug Analysis Laboratories, 12–15.

Tang, Y., Bao, Q., Tian, G., Fu, K., Cheng, H. (2015). Heavy metal cadmium tolerance on the growth characteristics of industrial hemp (*Cannabis sativa* L.) in China. International Conference on Advances in Energy, Environment and Chemical Engineering (AEECE-2015). doi: 10.2991/aeece-15.2015.58

Walker, P., Scarborough, J., Rand, H. (1982). An experimental surface-heated greenhouse. *Transactions of the ASAE*, *25*(4), 1022–1025.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESEARCHES CONCERNING EUROPEAN CORN BORER (*Ostrinia nubilalis* Hbn.) CONTROL, IN SOUTH-EAST OF THE ROMANIA

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Abstract

European corn borer (Ostrinia nubilalis Hbn) is one of the most important maize pests in Romania, especially in the central and western region of the country, in hilly areas and along the main rivers, in the Danube flooded plains. However, in last years, ECB produce high damages at maize crops in south and south-east of the Romania, too. In this paper there were presented results of a study concerning both chemical and biological control of this pest at maize crop, between 2016 and 2018, in the climatic conditions from south-east of the Romania. The experiences were carried out at NARDI Fundulea, Calarasi County. Five days before foliar spraying, maize plants were artificial infested witch ECB egg batches produced in laboratory conditions, from insects reared consecutive generations, using same artificial diet. Climatic conditions from first 15 days of July, when it has made artificial infestation of the maize plants were favourable for ECB attack in all studied years. As result, the attack of this pest at maize untreated plants was high. Foliar application of both, chemical and biological insecticides, provide effective protection of the maize plants, against ECB larva attack, in first week after the treatments. However, in the climatic conditions of the years 2016 and 2017, at treated variants, it has observed highest amount of ECB larva/plant, at assessment made in autumn, comparative with first assessment made at 5 days after the treatments.

Key words: *maize, borer, control, artificial, infestation.*

INTRODUCTION

Maize is one of the most important crops in Romania (Soare & Dobre, 2016; Tudor et al., 2017; Popescu A., 2018). According Eurostat reports, in 2018, our country occupied first place in EU, both for maize area and yield. However maize production per hectare, realized in Romania is lower, comparative with countries from Western Europe (Eurostat data, 2015; 2016; 2017). The main reasons for maize yield losses in Romania are because of drought (Stan & Naiescu, 1997; Sabau et al., 2002; Bonea & Urechean, 2011; Panaitescu et al., 2012; Ion et al., 2013; Cociu & Cizmas, 2015; Vizitiu et al., 2016), low temperatures during emergence period (Elena et al., 2011; Has et al., 2012; Rusu & Moraru, 2015; Balas-Baconschi et al., 2019), high temperatures during flowering period (Cuculeanu et al., 1999; Mateescu & Alexandru, 2010; Cociu et al., 2012: Pravalie et al., 2017), weeds (Bogdan et al., 2007; Borza et al., 2009; Stanila et al., 2013; Rusu et al., 2015), diseases and pests attack (Muresan & Mustea, 1995; Popov, 2002; Meiselle et al., 2010; Antonie et al., 2012; Ivas & Muresan, 2013; Georgescu et al., 2016; Trotus et al., 2018). According Trotus et al. (2011) only because of the pest attack, maize yield losses can arrive at 23%. In the climatic conditions from Romania, the main pests of the maize crop are wire worms (*Agriotes* spp.), maize leaf weevil (*Tanymecus dilaticollis*), european corn borer (*Ostrinia nubilalis*), western corn rootworm (*Diabrotica virgifera virgifera*), corn earworm (*Helicoverpa armigera* sin. *Chloridea obsolete*) and other species from *Noctuidae* family (Popov et al., 2001; Popov & Barbulescu, 2007; Rosca & Istrate, 2009; Trotus et al., 2013; Manole et al., 2017). European corn borer (*Ostrinia nubilalis*) it is one of the main pests of the maize crops from Romania (Paulian et al., 1976; Barbulescu et al., 1988; Mustea, 1990; Rosca & Barbulescu, 1997; Popov et al., 2005; Trotus et al., 2017). According Cristea et al. (2004), favorable areas from this pest are located in western and central part of the country (Transylvania) in hilly areas and along the main rivers or in the Danube flooded plains. Same author mentioned that in south and south-east, ECB is second pest like economic importance, after maize leaf weevil (*Tanymecus dilaticollis*). According data from the literature, in Romania, maize yield losses, because of ECB attack, can arrive at 60% (Paulian et al., 1976). In the middle of the years '70, Sapunaru and Hartman (1975) reported maize yield losses from 1400 to 2360 kg/ha, in different farms from east of the country. In Transylvania, at the beginning of the years '80, Mustea (1981) reported maize yield loses from 5.4 to 9.8%. More recent data, suggest that average maize yield losses in Romania, because of ECB attack were 7.5% (Popov & Rosca, 2007). After 2000, it hasn't reported important maize yield loses as result of ECB attack. However, after 2010, in journals for farmers it has reported higher attack of this pest at maize crops in western part of the country (Alexandri A., 2011; Plants Health, 2012; Farm, 2013). In last years, it has registered high attack of ECB in south-east of the Romania, too (Georgescu et al., 2016). Possible explication for this situation is because of the climatic changes that can favor insect in first stages of development (Olesen et al., 2011). Same author mentioned that climates changes from the Central and South-East Europe countries can have negative impact on local agriculture and, in same time, can favor pests attack. Researches for control of ECB attack in Romania it was made at NARDI Fundulea (for south-east of the country) and ARDS Turda (for central part of the country). ECB larva was reared on laboratory conditions, using same artificial diet for obtain egg-batches used for artificial infestation of the maize plants (Barbulescu, 1977; 1978; 1979; 1980). The purpose of these researches was to evaluate reaction of the maize inbreed lines and hybrids at ECB larva attack (Barbulescu et al., 1985; 1999; 2001; Barbulescu & Cosmin, 1987; 1997; Mustea, 1990), effectiveness of the foliar treatments in controlling of this pest (Barbulescu, 1989; Muresan & Mustea, 1995; Rosca & Barbulescu, 1997; Georgescu et al., 2016) and biological control effectiveness against this pest (Rosca & Barbulescu, 1986). Now days, only at NARDI Fundulea continue the activities concerning rearing of ECB larva, in laboratory conditions, consecutive generations using same artificial diet. As result of increasing of the area cultivated with maize, and higher attack of ECB larva, new researches are necessary, concerning control of this pest. In this paper, author collective present results concerning effectiveness of both, chemical and biological control of the ECB larva, at maize crops, in conditions of artificial infestation of the maize plants with ECB egg batches, in south-east of the Romania.

MATERIALS AND METHODS

The experiment was carried out at the experimental field of the Plants and Environment Collective, from National Agricultural Research Development Institute (NARDI) Fundulea, Calarasi County, Romania (44° 30′ N, 24° 1′ E). Experimental plots were arranged according randomized blocks design, each variant has four replications. Maize plants were sowed in plots, 10 m length and 4.2 m width (six rows) that correspond on an area of 42 m. For this experiment it has used Olt maize hybrid (FAO 450). In 2016, maize was sowing on 19 April, in 2017 maize was sowing on 18 May while in 2018 maize was sowing on 25 April. Because of high amount of rains occurred in April, 2017, it has registered a delay of sowing maize, approximate 4 weeks, comparative with normal period. Also, as result of drought registered in April, and first 10 days of May in 2018, most of the maize plants emerged after 20 May.

Table 1. Active ingredients used in research concerning effectiveness of ECB larva control, at NARDI Fundulea (2016-2018)

Variant no.	**Active ingredient** (concentration)	**Rate** (ml, g c. p./ha)
1	control (untreated)	-
2	deltamethrin (100 g/l)	75
3	lambda-cihalothrin (50 g/l)	150
4	indoxacarb (150 g/l)	250
5	chlorantraniliprole (20 %)	250
6	*B. thuringiensis* (54 %)	500
7	*B. thuringiensis* (54 %)	750
8	*B. thuringiensis* (54 %)	1000

Active ingredients used in this study were listed on Table 1. From pyrethroid class it has tested deltamethrin and lambda-cihalothrin active ingredients, from oxidazyne class it has tested indoxacarb active ingredient while from ryanoid class it has tested chlorantraniliprole active ingredient. Also it has tested a formulation on a base of *B. thuringiensis* subsp. *kurstaki* (ABTS-351 strain), used for biological control of the pests (Roh et al., 2007). In this study it has tested three doses of this formulation (500, 750 and 1000 g commercial product/ha).

The experiment was carried out in conditions of artificial infestation. Five days before foliar spraying, maize plants were infested with 10 egg batches/plant. Egg-batches used for artificial infestation are in "black-head" stage, when larva head become visible. From each experimental plot it has infested 40 plants. Plants were infested with ECB egg batches produced in the laboratory conditions, on continuous flux, after a technology described by Barbulescu (1980). Egg batches were placed with a tweezer at maize leaf base, from upper part of the plants (Figure 1). Attack level of the ECB larva at maize plants were analyzed both, in summer, at five days after foliar spraying (July) and in autumn (September), before harvesting (BBCH 99).

Figure 1. Artificial infestation of the maize plants with ECB egg batches, produced in laboratory conditions, at NARDI Fundulea

For these assessments, the stalk of the maize plants, that were artificial infested, before foliar spraying, was cooped in twice. During summer assessments it has determined the number of larva/plant and the number of holes/plant. During autumn assessments it has determined the number of alive ECB larva/plant and cavities length (cm) per plant. Also, at summer assessment, it has determined ECB attack incidence (%).

Meteorological data were collected from automatic meteo stations (iMethos), placed in the experimental field. It has registered average air temperature (°C) and rainfalls amount. This data was recorded every hour.

The data were **statistical analyzed** using Student-Newman-Keuls (SNK) test for multiple comparisons used to identify sample means that are significantly different from each other (Student, 1927; Neuman, 1939; Keuls, 1952).

RESULTS AND DISCUSSIONS

Climatic conditions from period that were made artificial infestation of the maize plants with ECB egg batches are very important. High temperatures and drought has result of higher mortality percent of ECB first instars larva and low percentage of larva eclosion (Paulian et al., 1976; Barbulescu et al., 2001). Same author mentioned that heavy storms registered in this period can have negative effect on population dynamic, of this pest. However, if air temperature is moderate and air humidity is high can result an increasing of larva eclosion percentage and lover mortality percent of 1st instar larva (Rosca & Rada, 2009).

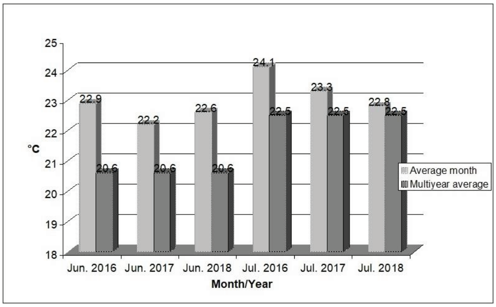

Figure 2. Average air temperature registered in June and July, period 2016-2018, at NARDI Fundulea

During this experiment, at NARDI Fundulea, in 2016, artificial infestation of the maize plants were made on 5 July and foliar treatments on 10 July, in 2017, artificial infestation were made on 8 July and foliar treatments on 13 July while in 2018, artificial infestation were made on 1 July and foliar treatments on 6 July.

Data recorded at meteorological stations from experimental field of NARDI Fundulea, reveal that, in summer period, between 2016 and 2018, average air temperature registered in June and July was higher then multiyear average (Figure 2). Higher deviation from the multiyear average was recorded, both in June and July, 2016 (+2.3 and +1.6°C). Also, in June, 2018, it has recorded high deviation of the average temperature recorded in this month, comparative with multiyear average (+2.0°C).

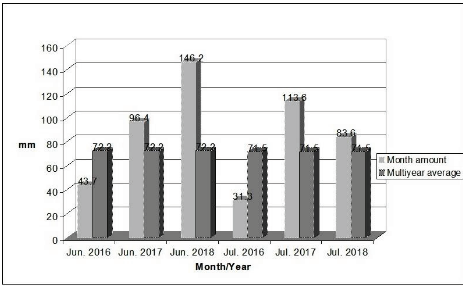

Figure 3. Rainfalls amount registered in June and July, period 2016-2018, at NARDI Fundulea

However, deviation of the average monthly temperature comparative with multiyear average was higher in June comparative with July, in all studied years.

Regard as rainfall amount, registered at automatic meteo station, from experimental field of NARDI Fundulea, in June and July, between 2016 and 2018, it has ascertained higher variability of this parameter (Figure 3). In 2016, rainfalls amount registered in June and July was below multiyear average while in 2017 and 2018 was higher than averages. Higher deviation from the average was recorded in June, 2018 (+74.0 mm).

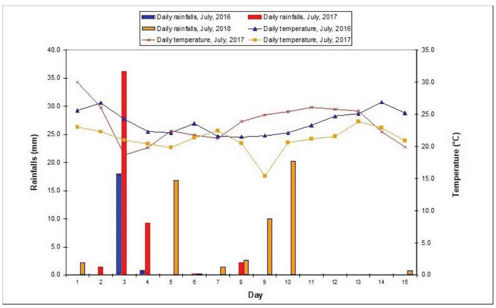

Figure 4. Daily temperatures and rainfalls recorded in first 15 days of July, between 2016 and 2018, at NARDI Fundulea

Daily temperatures and rainfalls amount are very important for ECB population dynamic. The most sensitive stage of this pest to weather conditions is during egg laying stage-larva eclosion-larva 1st instars. In 2016, daily rainfalls distribution and temperatures evolution, in first 15 days of July, was favorable for ECB larva (Figure 4). High rain amount has occurred two days before artificial infestations, as result the air humidity were favorable for larva eclosion. Similar situation was recorded in first 15 days of July, 2017. Between 3 and 4 July, it as registered 45.4 mm of rain, as result air humidity at 8 July, when it has made artificial infestation of maize plants with ECB egg batches were favorable for larva eclosion. Rainfalls amount registered in first 15 days of July, 2018 was high. After artificial infestation, on 1 July it has recorded 2.2 mm of rain. Between 5 and 10 July, 2018, it has recorded 51.2 mm of rains and moderate temperatures. However, temperatures recorded in first 48 hours after artificial infestation, were high and air humidity was low, that represent less favorable conditions for larva eclosion.

Table 2. Effectiveness of some active ingredients used in controlling of the *O. nubilalis* larva attack, at NARDI Fundulea, summer assessments (July)

Active ingredient	Rate (ml, g c. p./ha)	Attack incidence (%)			Number of larva/plant		
		2016	2017	2018	2016	2017	2018
control (untreated)	-	100 A	100 a	62.50 a	14.75 a	13.80 A	2.05 S
deltamethrin	75	86.25 Ab	86.25 b	3.75 b	7.95 b	5.20 B	0.08 B
lambda-cihalothrin	150	92.50 Ab	68.75 c	21.25 b	7.79 b	4.80 Bc	0.23 B
indoxacarb	250	97.50 A	85.00 b	13.75 b	6.08 b	3.10 C	0.15 B
chlorantraniliprole	250	67.50 C	67.50 c	21.25 b	7.75 b	3.45 Bc	0.45 B
B. thuringiensis	500	78.75 Bc	82.50 bc	13.75 b	7.55 b	4.33 Bc	0.35 B
B. thuringiensis	750	77.50 Bc	78.75 bc	11.25 b	5.95 b	4.05 Bc	0.28 B
B. thuringiensis	1000	75.00 Bc	76.25 bc	10.00 b	5.93 b	3.85 Bc	0.15 B

*Means followed by same letter or symbol do not significantly differ (P = 05, Student-Newman-Keuls)

In Table 2 there were presented results of assessments made in summer, at 5 days from foliar spray and 10 days from artificial infestation of maize plants with ECB egg batches. Regard as attack incidence of *O. nubilalis*, at five days after foliar spraying, it can have ascertained that, in climatic conditions of the years 2016 and 2017, at untreated variant, value of this parameter was maximum (100%). In 2016, at treated variants, attack incidence of the ECB larva at maize plants, ranged from 67.50 to 97.50% while in 2017, attack incidence of this pest, at treated variant, ranged from 67.50 to 86.25%. Lower values of the attack incidence it has registered in 2018, at all experimental variants. Regard as number of larva/plant, at five days after foliar spraying, data from Table 2 reveal that, at untreated variant, in 2016 it has registered 14.75 larva/plant while, one year later, at same variant, it has registered 13.80 larva/plant. However, in 2018, at control (untreated) variant it has registered only 2.05 larva/plant. In the climatic conditions of the years 2016 and 2017, in conditions of a heavy infestation pressure, and favorable climatic conditions for pest development, foliar spraying of the maize plants with, both chemical and biological insecticides, provide effective protection against this pest, in first days after treatments. Same results were observed in conditions of the year 2018, when it has registered low pest pressure, especially because unfavorable climatic conditions, after artificial infestation of the maize plants with ECB egg batches, produced in laboratory conditions.

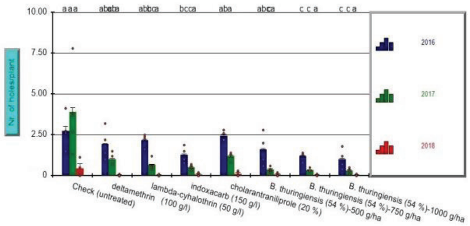

Figure 5. Number of holes/plant, at five days after foliar spraying (NARDI Fundulea, 2016-2018)

In all three years of this experiment, at five days after foliar spraying it has ascertained that there were significant statistic differences between biological products on a base of *B. thuringiensis* subsp. *kurstaki* (ABTS-351 strain) and untreated variant, concerning both, attack incidence and number of ECB larva/plant. However, there weren't statistical differences between the three doses of this biological product (p<.05). Concerning of ECB attack incidence, at five days from foliar treatment, according Student-Newman-Keuls (SNK) test, highest statistical differences comparative with untreated variant it has registered in case of chlorantraniliprole active ingredient, in 2016 and 2017. In the climatic conditions of 2018, there weren't registered statistical differences between treated variants, both with chemical and biological insecticides. Using SNK test, it has ascertained that, concerning number of larva/plant, in conditions of the high pest pressure from 2016 and low pest pressure in 2018, there weren't statistical differences between treated variants and control (untreated) variant (p<.05), while in 2017, highest statistical differences, comparative with untreated variant it has registered in case of indoxacarb active ingredient.

Table 3. Effectiveness of some active ingredients used in controlling of the *O. nubilalis* larva attack, at NARDI Fundulea, autumn assessments (September)

Active ingredient	Rate (ml, g c. p./ha)	Cavities length/plant (%)			Number of larva/plant		
		2016	2017	2018	2016	2017	2018
control (untreated)	—	20.25 A	36.18 a	24.33 a	12.63 a	12.32 a	4.45 a
deltamethrin	75	8.33 b	22.35 a	18.68 ab	5.50 b	9.63 abc	2.93 b
lambda-cihalothrin	150	8.31 b	19.50 a	14.43 b	5.70 b	8.18 abc	2.65 b
indoxacarb	250	8.22 b	18.94 A	19.03 ab	4.50 b	7.58 bc	2.75 b
chlorantraniliprole	250	5.30 c	12.11 C	4.88 c	4.01 b	6.51 c	1.03 c
B. thuringiensis	500	8.88 b	25.08 B	18.90 ab	5.93 b	12.29 a	3.08 b
B. thuringiensis	750	8.44 b	24.68 B	16.98 ab	5.60 b	11.75 ab	3.10 b
B. thuringiensis	1000	7.81 b	23.53 B	16.70 ab	5.40 b	10.72 abc	2.96 b

*Means followed by same letter or symbol do not significantly differ (P=.05, Student-Newman-Keuls)

Similar situation it has registered in case of holes number/plant, made by ECB larva, in climatic conditions of the years 2017 and 2018 (Figure 5). However, in 2016, highest statistical differences comparative with control variant, it has registered in case of last two doses of biological insecticide, on base of *B. thuringiensis* (750 and 1000 g c. p./ha).

In Table 3 there were presented results of assessments made in autumn, before harvest. In the climatic conditions of the year 2016, it has ascertained that cavities length/plant, at untreated variant was 20.25 cm. At treated variants this parameter ranged between 5.30 and 8.88 cm. According SNK test, highest statistical difference comparative with untreated variant it has registered in case of variant treated with chlorantraniliprole (p<.05). In conditions of the year 2017, at all experimental variants it has registered high values of the cavities length/plant. At maize plants from control (untraded) variant, cavities length/plant was higher than 36 cm. Higher statistical differences comparative with untreated variant it was registered in case at chlorantraniliprole variant. Similar situation it has recorded in conditions of the year 2018.

Concerning number of the larva/plant, in autumn, when maize plants are in BBCH 99 stage, assessments results reveal that, in 2017 and 2018, at all treated variants, number of larva/plant was higher comparative with summer period (at five days from foliar spray). A possible reason for this is because of attack of natural population of ECB or appearance of a new generation of this pest, in the climatic conditions from south-east of the Romania. According SNK test, it has ascertained that, there weren't statistical differences between all treated variants, concerning number of larva/plant. In same time the differences between treated variants and control (untreated) variant were significant ($p<.05$). In conditions of 2017 and 2018, highest statistical differences comparative with untreated variants, it has registered in case of variant treated with chlorantraniliprole active ingredient ($<.05$).

In conditions of high pest pressure as result of artificial infestation, at NARDI Fundulea, foliar treatment with, both chemical and biological insecticides provide effective protection of the maize plants, against this pest, first five days after treatments. However single foliar treatment didn't provide effective protection of the maize plants, until the end of the vegetation period, in autumn.

CONCLUSIONS

Rearing of the European corn borer (*Ostrinia nubilalis* Hbn.), consecutive generations, in laboratory conditions, using same artificial diet is a good method to evaluate effectiveness of the insecticides applied like foliar spraying for control this pest.

Climatic conditions from summer period, registered at NARDI Fundulea were favorable for ECB larva development in 2016 and 2017 and less favorable in summer of the 2018.

In the conditions of artificial infestation of the maize plants, with ECB egg batches, foliar spraying assign effective protection of the maize plants against pest attack only in summer period.

In this experience, between 2016 and 2018, in the climatic conditions from south-east of the Romania, highest effectiveness in control of this pest was registered in case of chlorantraniliprole active ingredient.

Between 2016 and 2018, in the climatic conditions from south-east of the Romania, biological insecticide on a base of *B. thuringiensis* subsp. *kurstaki* (ABTS-351 strain) can provide effective protection for maize plants against ECB larva.

Further studies are necessary to evaluate impact of the climatic changes on ECB populations dynamic in Romania.

ACKNOWLEDGEMENTS

This research work was carried out with the support of National Agricultural Research and Development Institute, Fundulea, Plants and Environment Collective.

REFERENCES

Alexandri, A. (2011). *Ostrinia nubilalis-Diabrotica virgifera virgifera. Plants Health*, *159*, 12.

Antonie, I., Stanciu, M., Sand, C., Blaj, R. (2012). The researches regarding the biodiversity of the entomologic of the corn cultures in the Sibiu County. *Scientific Papers, Series Management, Economy. Engineering. in Agriculture and Rural Development*, *12*(1), 5–10.

Barbulescu, A. (1977). Rearing of the European corn borer (*Ostrinia nubilalis* Hbn.) on different artificial diets. *Problems of Plant Protection*, *5*(2), 141–150.

Barbulescu, A. (1978). Insects rearing on artificial diets. *Problems of Plant Protection*, *6*(4), 375–400.

Barbulescu, A. (1979). The influence of temperature on egg incubation in the European corn borer *(Ostrinia nubilalis* Hb.). *Problems of Plant Protection*, *7*(4), 377–384.

Barbulescu, A. (1980). Mass rearing of the European corn borer (*Ostrinia nubilalis* Hbn.) on artificial diet. *Problems of Plant Protection*, *8*(1), 1–12.

Barbulescu, A., Cosmin, O., Sarca, T., Restea, T. (1985). Testing some maize lines for resistance to *Ostrinia nubilalis* Hb, during 1982-1984. *Problems of Plant Protection*, *13*(3), 267–272.

Barbulescu, A., Cosmin, O. (1987). Maize inbred lines with some degree of resistance *to Ostrinia nubilalis* Hb. *Problems of Plant Protection*, *15*(4), 301–306.

Barbulescu, A., Bigiu, L., Bratu, R., Ciurdarescu, G., Craiciu, M., Gheorghe, M., Mateias, M.C., Pelmus, A., Pelmus, V., Popov, C., Rugina, M., Tusa, C., Voinescu, I., Vonica, I. (1988). Evolution of some diseases and pests of grains, industrial and forage crops in our country during 1987. *Problems of Plant Protection*, *16*(1), 57–74.

Barbulescu, A., Cosmin, O. (1997). Maize inbreed lines obtained at Fundulea, characterized by a certain

resistance degree to *Ostrinia nubilalis* Hb. *Problems of Plant Protection*, *25*(1), 1–8.
Barbulescu, A., Cosmin, O., Sabau, I. (1999). Data obtained at Fundulea, concerning maize resistance at European corn borer *Ostrinia nubilalis* attack. *Problems of Plant Protection*, *27*(2), 173–180.
Barbulescu, A., Popov, C., Sabau, I. (2001). The behaviour of a Monsanto maize hybrid-Dekalb 512 bt to the attack by the European corn borer in Romania. *Romanian Agricultural Research*, *15*, 65–73.
Balas-Baconschi, C.G., Pamfil, D., Savati, M., Tinca, E., Călugăr, R., Haș, V. (2019). The effect of simulating hail and late spring frost on certain parental forms of registered maize hybrids in the north west of Transylvania. *Romanian Agricultural Research*, *36*, 3–12.
Bogdan, I., Gus, P., Rusu, T., Pop, A., Vaju, A., Morar, P. (2007). Research concerning weed control in maize crop. *Agronomical Researches in Moldavia*, *1*(129), 16–21.
Bonea, D., Urechean, V. (2011). The evaluation of water stress in maize (*Zea mays* L.) using selection indices. *Romanian Agricultural Research*, *28*, 79–86.
Borza, I.M., Domuta, C., Sandor, M., Ciobanu, C., Samuel, A., Brejea, R. (2009). Researches concerning the weeds influence on water use efficiency in maize from North-Western Romania. *Annals of Oradea University, Fascicule: Environment Protection*, *14*, 47–54.
Cociu, A.I. (2012). Air temperature and precipitation influence on maize grain yield within different annual and perennial crop rotations. *Romanian Agricultural Research*, *29*, 149–154.
Cristea, M., Cabulea, I., Sarca, T. (2004) *Maize. Monographic study, Volume 1*, Romanian Academy Publishing house, Cap. 14, 589–626.
Cuculeanu, V., Marica, A., Simota, C. (1999). Climate change impact on agricultural crops and adaptation options in Romania. *Climate Research*, *12*(2-3), 153–160.
Elena, D.A., Constantin, D.D., Flavia, B. (2009). Agricultural Insurance in Romania. *Annals of Faculty of Economics, University of Oradea, Faculty of Economics*, *3*(1), 169–175.
Georgescu, E., Toader, M., Balaban, N., Rîsnoveanu, L., Cana, L. (2016). Testing of the new active ingredients for controlling of the *Ostrinia nubilalis* Hbn. at maize crop, in conditions of artificial infestation, at NARDI Fundulea, *Annals of Craiova University, series Agriculture – Montanology – Cadastre*, *XLVI*(2), 121–126.
Grozea, I., Badea, A.M., Prunar, F., Adam, F. (2006). Diversity of maize pests and their damaging effect in an agroecosystem from the western side of the country. *Agronomical Researches in Moldavia 3*(1), 15–22.
Has, I., Has, V., Copandean, A., Schiop, T., Rotar, C. (2012). Evaluation of Maize" Turda" Germplasm as Sources of Earliness in Breeding Programs. *Bulletin of the University of Agricultural Sciences & Veterinary Medicine Cluj-Napoca, Agriculture*, *69*(1).
Ion, V., Dicu, G., Basa, A.G. (2013). Yield components at some hybrids of sunflower (*Helianthus annuus* L.) under drought conditions from South Romania. *AgroLife Scientific Journal*, *2*(2), 9–14.
Keuls, M. (1952). The use of the "studentized range" in connection with an analysis of variance. *Euphytica*, *1*, 112–122.
Manole, T., Chireceanu, C., Teodoru, A. (2017). Current Status of *Diabrotica virgifera virgifera* LeConte, 1868 (Coleoptera: Chrysomelidae) in Romania. *Acta Zoologica Bulgarica*, *Supplement, 9*, 143–148.
Meissle, M., Mouron, P., Musa, T., Bigler, F., Pons, X., Vasileiadis, V.P., Otto, S., Antichi, D., Kiss, J., Pálinkás, Z., Dorner, Z. (2010). Pests, pesticide use and alternative options in European maize production: current status and future prospects. *Journal of Applied Entomology*, *134*(5), 357–375.
Mateescu, E., Alexandru D. (2010). Management recommendations and options to improve crop systems and yields on south-east Romania in the context of regional climate change scenarios over. *Scientific Papers, Series A. Agronomy, 53*. 328–334.
Muresan, F., Mustea, D. (1995). Results obtained in European corn borer control *Ostrinia nubilalis* Hbn. at the agricultural research station Turda. *Problems of Plant Protection*, *23*(1), 23–34.
Mustea, D., (1981). Elements for the estimation of the losses caused by *Ostrinia nubilalis* Hbn. in maize crops. *Problems of Plant Protection*, *9*(4), 349–357.
Mustea, D., (1990). Studies of some aspects of maize resistance at *Ostrinia nubilalis* Hbn. in recurrent selection process. *Problems of Applied and Theoretical Genetics*, *22*(2), 61–71.
Newman, D. (1939). The distribution of range in samples from a normal population, expressed in terms of an independent estimate of standard deviation. *Biometrika*, *31*(1), 20–30.
Olesen, J.E., Tmka, M., Kersebaumc, K.C., Skjelvågd, A. O., Seguine, B., Peltonen-Sainiof, P., Rossig, F., Kozyrah, J., Micalei, F. (2011) Impacts and adaptation of European crop production systems to climate change. *European Journal of Agronomy*, *34*(2), 96–112.
Panaitescu, L., Lungu, M., Nita, S. (2012). The influence of the semi-dry climate of Dobrudja on the maize production. *Present Environment and Sustainable Development*, *6*(2), 381–385.
Paulian, F., Mustea, D., Burdea, V., Baniţă, E., Enica, D., Peteanu, S., Petcu, L., Sapunaru, T., Sandru, I. (1976). Evolution of European corn borer-*Ostrinia nubilalis* Hb and damaging potential registered in 1971-1975 in R.S. Romania. *Problems of Plant Protection*, *4*(1), 23–51.
Pravalie, R., Bandoc, G., Patriche, C., Tomescu, M. (2017). Spatio-temporal trends of mean air temperature during 1961–2009 and impacts on crop (maize) yields in the most important agricultural region of Romania. *Stochastic Environmental Research and Risk Assessment*, *31*(8).
Popescu, A. (2018). Maize and wheat-top agricultural products produced, exported and imported by Romania. *Scientific Papers. Series" Management,*

Economic Engineering in Agriculture and Rural Development, *18*(3), 339–352.
Popov, C. (2002). Researches concerning the protection of the cereals, leguminosae for grains, technical and forage plants against pathogen agents and pests, year 2001. *Problems of Plant Protection*, *30*(2), 109–190.
Popov, C., Barbulescu, A., Trotus, E., Vasilescu, S., Bucurean, E. (2001). Control of wireworms by seed treatment in Romania. *Romanian Agricultural Research*, *15*, 69–75.
Popov, C., Guran, M., Raranciuc, S., Rotarescu, M., Spiridon, C., Vasilescu, S., Gogu, F. (2005). Phytosanitary state of the cereals, leguminous for grains, forage and technical crops from Romania, year 2004. *Problems of Plant Protection*, *33*(1-2), 7–30.
Popov, C., Barbulescu, A. (2007). 50 years of scientific activity in field crop protection area, against pests and diseases. *Annals of NARDI Fundulea*, *75*, 371–404.
Popov, C., Rosca, I. (2007) Technology of European corn borer (*Ostrinia nubilalis* Hbn.) mass rearing, in continuous system and successive generations. *Entomological Research*, *37*(1), 126.
Roh, J.Y., Choi, J.Y., Li, M.S., Jin, B R., Je, Y.H. (2007). *Bacillus thuringiensis* as a specific, safe, and effective tool for insect pest control. *Journal of Microbiology and Biotechnology*, *17(*4), 547.
Rosca, I,, Barbulescu, A. (1986). Attempts of biological testing of *Bacilus thuringiensis*, products with *Ostrinia nubilalis* Hb. as a test species. *Problems of Plant Protection*, *14*(2), 133–140.
Rosca, I., Barbulescu, A. (1997). Researches on labelling with „Calco Red Dye" of the European corn borer (*Ostrinia nubilalis* Hb.). *Problems of Plant Protection*, *25*(2), 199–206.
Rosca, I., Rada I. (2009). *Entomology (Agriculture, Horticulture, Forest)*. Bucharest, Romania: Alpha MDN Publishing house, 699 pp. (Cap. II: 119–141).
Rusu, T., Moraru, P.I. (2015). Impact of climate change on crop land and technological recommendations for the main crops in Transylvanian Plain, Romania. *Romanian Agricultural Research*, *32*, 103–111.
Rusu, T., Moraru, P.I., Pop, A.I., Salagean, T., Duda, B. M. (2015). Influence of tillage system and weed control methods on the weeding and soil weed seed bank. *Conference Proceedings SGEM*, *2*. 191–197).
Trotus, E., Buburuz, A.A., Zaharia, P. (2011). Researches on the protection of maize crops against soil pests. *Agronomical Researches in Moldavia*, *4*, 45–51.
Trotus, E., Zahari,a P., Nae, M., Buburuz, A.A., Pochiscanu, S. F. (2013). Research on the evolution of *Agrotis segetum* Den. et Schiff. species under conditions of central Moldavia. *Annals of NARDI Fundulea*, *81*, 141–153.
Trotus, E., Buburuz, A.A., Ursache, P.L. (2017). *New data regarding the corn crops protection against specific pests*. *ARDS Secuieni, Neamt (1962-2017)*, 55 years of Research - Development, Anniversary volume, Iasi, Romania: Ion Ionescu de la Brad Publishing house, 115–130.
Trotus, E., Buburuz, A.A., Ursache, P.L. (2018). New data regarding the appearance, evolution and the attack produced by *Ostrinia nubilalis* Hbn. species, at maize crop, under the center of Moldavia conditions. *Romanian Agricultural Research*, *35*, 229–236.
Tudor, V., Popa, D., Gimbasanu, G.F. (2017). The analysis of the cultivated areas, the production and the selling price for maize crops during the pre-and post-accession periods of Romania to the European Union and trends of evolution of these indicators, 2017. *Scientific Papers: Management, Economic Engineering in Agriculture & Rural Development*, *17*(2), 387–394.
Sabau, N.C., Domuta, C., Man, T.E., Sandor, M., Brejea, R. (2002). Drought analysis by the climate indexes in link with the yield of the main crops from the Crişurilor Plain, Romania. *International Conference on Drought Mitigation and Prevention of Land Desertification*, 21–25.
Sapunaru, T., Hatman M. (1975). The particularities of the phytosanitary state from area served by agricultural research station Podu-Iloaiei. *Problems of Plant Protection*, *3*(4), 331–343.
Soare, E., Dobre, I. (2016) Changes and trends of corn production in Romania. *Scientific Papers: Management, Economic Engineering in Agriculture & Rural Development*, *16*(1), 511–516.
Stan, I., Naescu, V. (1997). Maize response to water deficit. *Romanian Agriculture Research*, 7–8.
Stanila, S., Gheres, M., Duda, M., Mogosan, C. (2013). Influence of minimum tillage systems on the control of *Convolvulus arvensis* L. on wheat, maize and soybean. *Journal of Food, Agriculture & Environment*, *11*(2), 563–566.
Student. (1927). Errors of Routine Analysis. *Biometrika*, *19*(1/2), 151–164.
Vizitiu, O.P., Calciu, I.C., Simota, C.C. (2016). Drought intensity on arable land in Romania–processes and tendencies, *Geographic seminar papers "Dimitrie Cantemir"*, *42*(1), 67–80.
***EUROSTAT (2015). http://ec.europa.eu/eurostat/data/database
***EUROSTAT (2016). http://ec.europa.eu/eurostat/data/database
***EUROSTAT (2017).
http://ec.europa.eu/eurostat/data/database
***Farm (2013). *Problems with Ostrinia*. 125, 62.
***Plants Health (2012). *Ostrinia attack at maize in many agricultural areas from Romania*. 170, 38.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE EFFECTS OF MICRO-GRANULATED FERTILIZATION ON POLLINATION AND YIELD OF HYBRID SEED CORN

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Abstract

This study is carried on to identify the effects of micro-granulated fertilization on pollination and yield of hybrid seed corn. On the trial that set up as random parcels, the inbreds of a Monsanto simple corn hybrid are used. We have control parcels without micro-granulated fertilizer and test parcels with micro-granulated fertilizer applied. The results of the study help us draw valuable conclusions regarding the efficiency of the application of micro-granulated fertilizer at planting in seed corn and its effect on pollination nicking: the time from planting to emergence was shorter on average by 2 days which helped fast development during early vegetation stages; in 4 leaf stage the root of the corn with micro-granulated fertiliser applied at planting was 20% more developed than the root of the control; at 8 leaf stage we can observe the 20% difference in root development; the plants had a faster development which helped reach flowering time 4-5 days before the control; the earlier start of flowering compared to the control resulted in a better protection from draught limiting its protandric effects thus generating a higher percentage of pollinated kernels; plant height and vigour were obviously increased compared to the control, showing better resistance to disease and flattening; we observed a superior ear development from a size standpoint and a higher Thousand Kernel Weight. Most importantly following the study we can observe that the parcel fertilized with micro-granulated fertilizer had a higher yield by 1104 kg per hectare.

Key words: *micro-granulated fertilizer, pollination, nicking, yield.*

INTRODUCTION

Attaining the full potential of hybrid corn and its productive qualities is strongly tied with the quality of the hybrid seed obtained after the multiplication process.

The process of breeding and multiplication of seeds is a long term process which need huge efforts and vast budgets for research and development.

The seed production system must be organized so it is able to allow the propagation into production in the shortest time of new valuable varieties and hybrids created in the country or registered in the EU and at the same the periodical renewal of the seeds of the varieties and hybrids already cultivated in the country.

The problem of fertilization in seed corn fields is becoming increasingly important as seed companies (Monsanto, Pioneer etc.) are facing a continuous increase in demand of high quality seed with increasingly varied traits and increased yield capability. Fertilization used for seed corn fields must take into account the newest ideas in the fields of agro-chemistry and plant growing, the grower having the duty of maintaining hybrid characteristics and technically not to influence the synchronisation at pollination of the two parental inbred lines. Any different assimilation of fertilizer might affect the synchronisation thus, increasing or decreasing the quantity of viable pollen at the time of fecundation.

For a long time, seed growers have used classic fertilization schemes which included complex fertilizer as 16:16:16, 20:20:0 or simple fertilizer as Ammonium Nitrate and even Urea despite its high volatilisation rate during summer. Nitrogen fertilisation is an expensive but necessary input in any agricultural system. Nitrogen fertilisation furthermore enables farmers to achieve high yields that drive modern agriculture (Brady & Weil, 2008).

Time of N application studies have been reported, extensively in the literature. The general conclusion among researches has been that N should be applied nearest to the time it is needed by the crop, i.e., dressed several weeks after corn emergence (Aldrich, 1984; Fox et al., 1986; Olson & Kurtz, 1982; Russelle et al., 1981; Stanley & Rhoades, 1977; Welch et al., 1971). The context of higher importance of fertilizers costs leading farmers to "just enough" habits with more sophisticated products as well as an increased awareness of precision farming methods and micro-fertilizers agronomic benefits (Laurent Lemarchand et al., 2016).

Taking into account the increase in required yield of hybrid seed and taking into account the low vigour of inbreds, during the last few years seed growers have turned their attention to fertilisation in general, but more so on starter and precision fertilisation to ensure the inbreds have a good development even from the earliest vegetative stages. Micro-granulated fertilizers influence the vegetative development of plants, encouraging the formation of taller, vigorous plants, with large dark green leaves (Crista F et al., 2004).

Micro-granulated fertilizers are target distributed near the seed ensuring the necessary nutritional start, especially phosphorus which has a key role in the development of the root system immediately after the water absorption by the seed and at germination, respectively at the development of the radicle. One of the unknowns that occurs when using micro-granulated fertilizer is its effect on pollination timing taking into account the fact that most hybrid seed corn is planted split in time, because of different maturities of the parental inbreds and also to ensure that most pollen is available from the male inbred at the time of the silking of female inbreds.

It is obvious that any delay determined by using this type of fertilizer, for example the early or late opening of male anthers affects the final percentage of pollinated ovules and thus the final yield. Therefore a study of the effects of micro-granulated fertilization on polination and yield in seed corn is important and valuable for seed corn production technology.

The aim of this study was to assess the behaviour of the inbred lines in terms of pollination and seed yield, mainly of a Monsanto maize hybrid, under different fertilization conditions, including fertilization at sowing with micro-granulated fertilizer.

MATERIALS AND METHODS

The field experiments were carried out during the 2017 growing season in the soil and climate conditions of the company Integrasem SRL from Bivolari Commune, Iasi County, Romania. Several variants of fertilization of parental forms with classical and micro-grained fertilizers have been tested in the field experience and the comparison of the results obtained with the control represented by the fertilization scheme established and agreed with the agronomist of Monsanto.

To start the experience, the inbred lines of a semi-early hybrid, group FAO 360, a hybrid best suited to zoning due to climate change, were chosen. This is a hybrid with good tolerance to drought, with an early flowering that protects it from the summer heat, with a deep and vigorous root system and with an excellent start into vegetation. The sowing scheme dictated the time difference between the sowing of the two parental forms to ensure the flowering coincidence of the female and male inbreds and a longer period of viable pollen to have the fullest pollination.

The classic fertilizers used in this experiment are complex fertilizers with nitrogen, phosphorus and potash of the type 16:16:16; 20:20:0 and fertilizers based on ammonium nitrate ammonium. The micro-grain fertilizer used is one with a specific formulation to ensure an optimal dose of nutrients with N, and especially with P, Ca, S, Zn, at germination and plant growth.

Physiostart is a starter product designed specifically to meet plant requirements in the first phase of vegetation. It is administered to spring crops at a dose of 20-30 kg/ha. Physiostart contains nitrogen in ammoniacal form, preferred by young plants, and does not affect germination of seeds. Phosphorus is available immediately because it has a high solubility and its effect is seen on the development of the root system. The product also contains calcium, sulfur and zinc microelements. It is a formula created specifically to ensure a good start for crops. Being a micro-granulated product, we need to know that it cannot cover all the nutrient requirements of agricultural crops and that is why basic fertilization with chemical fertilizers should be done.

Physiostart is ideal for spring crops as it provides a strong root development, thus increasing plant resistance to drought, ensuring a faster start of culture, avoiding critical phases during strong heat, and helping to continually increase of the plant of culture.

The experience was performed on a surface of 5.47 ha organized as random blocks with 4 variants and 3 replications, each variant having 0.5 ha size. The sowing parity was "narrow rows" 6 female rows to 2 male rows with the distance of 60 cm betweeen female parental rows, 60 cm between the female and male 1, and 45 cm between the two rows of Male 1 and Male 2, with a distance of 17 cm between seeds in a row, resulting in high density crops.

Analyzes and determinations made in the field and laboratory were:

- ✓ Plant height - in the 4-leaf stage;
- ✓ Plant height - in the 8-leaf stage;
- ✓ The number of leaves per plant;
- ✓ Evolution of the root system up to the phase of 4 leaves and 8 leaves;
- ✓ Leaf -7 length;
- ✓ Duration from sowing to emergence;
- ✓ Duration from emergence to flowering;
- ✓ The flowering gap between Male inbred and the Female inbred;
- ✓ The average number of kernels on the ear;
- ✓ Grain Yield/ha.

Statistical analysis

The statistical analysis of the data was performed using ANOVA Analysis. Relationship between different traits was determined with the Linear Regression Procedure.

RESULTS AND DISCUSSIONS

The results revealed a significant correlation between all the analyzed characters and the yield per hectare. For each comparison the fertilized version of the micro-grain fertilizer, Physiostart, has outperformed the control variant. Thus, there was a production increase of 1015 kg compared to the control variant (Table 1).

A first observation is the much faster emergence of the plants at the micro-granulated applied version at sowing, i.e. about 6 days after sowing compared to the control variant at which emergence occurred 9 days after sowing.

In Figure 1 is shown the effect in the linear regression between Sowing-Emergence time and Yield. Significant phosphorus intake near the first seed root accelerated the emergence of the version fertilized with micro-granulated fertilizer, the difference being visible also in later stages of development.

Table 1. The mean values of morpho-physiological and agronomic characters recorded in micro-granulated fertilization variants and maize control variants

NS	ND S-E	NR - 4 L	H-4 L	NR - 8 L	H - 8 L	LL 7	ND E-F	FG T1-M2	NK/E	PR	V
1	**8**	**7**	**23**	**10**	**48**	**30**	**70**	**3**	**417**	**4720**	**MG**
2	**7**	**8**	**25**	**11**	**51**	**32**	**67**	**2**	**427**	**4840**	
3	**7**	**8**	**24**	**11**	**50**	**31**	**68**	**2**	**420**	**4770**	
4	**6**	**9**	**26**	**12**	**52**	**33**	**66**	**2**	**431**	**4870**	
5	**7**	**8**	**24**	**11**	**49**	**30**	**70**	**3**	**419**	**4760**	
Max	**8**	**9**	**26**	**12**	**52**	**33**	**70**	**3**	**431**	**4870**	
Min	**6**	**7**	**23**	**10**	**48**	**30**	**66**	**2**	**417**	**4720**	
Average	**7**	**8**	**24.4**	**11**	**50**	**31.2**	**68.2**	**2.4**	**422.8**	**4783**	
1	9	4	15	7	33	16	74	4	406	3730	**M**
2	10	3	13	6	31	14	75	5	395	3760	
3	9	4	14	7	32	15	74	5	398	3710	
4	9	4	16	7	34	16	73	4	410	3780	
5	8	5	17	8	35	17	72	4	412	3860	
Min	8	3	13	6	31	14	72	4	395	3710	
Max	10	5	17	8	35	17	75	5	412	3860	
Average	**9**	**4**	**15**	**7**	**33**	**15.6**	**73.6**	**4.4**	**404.2**	**3768**	

NS - sample number; ND S-E - number of days from sowing to emergenece; NR-4 L - roots number - 4 leaves; H-4L - height - 4 leaves; NR-8 L - roots number - 8 leaves; H-8L - height - 8 leaves; LL-7 - leaf length 7 (cm); ND E-F - number of days emergence-flowering; FG T1-M - Flowering gap between male inbred T1 and female inbred M (days); NK/E - number of kernels on ear; PR - seed production/ha (kg); V - variant; MG - micro-granulated fertilizer; M - control

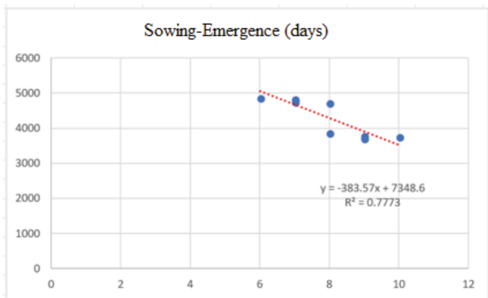

Figure 1. Linear regression between Sowing-Emergence Time and Grain Yield (kg/ha)

Significant phosphorus intake near the first seed root accelerated the emergence of the version fertilized with micro-granulated fertilizer, the difference being visible also in later stages of development. In the four leaf stage of the two variants, the number of roots and the height of the plants were evaluated (Figure 2). The variant tested showed a higher root development of an average of 9 roots developed versus 6 in the control variant.

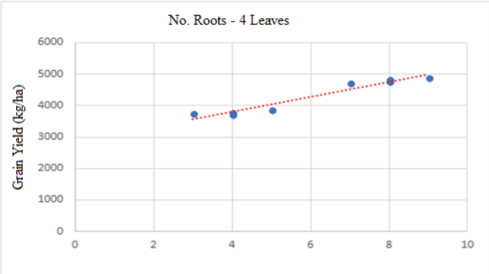

Figure 2. Linear regression between No. Roots - 4 leaves and Grain Yield (kg/ha)

Regression Statistics	
Multiple R	0.971462885
R Square	0.943740137
Adjusted R Square	0.936707654
Standard Error	136.5140269
Observations	10

ANOVA

	df	SS	MS	F	Significance F
Regression	1	2500911.364	2500911.364	134.1972896	2.80329E-06
Residual	8	149088.6364	18636.07955		
Total	9	2650000			

	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95.0%	Upper 95.0%
Intercept	2849.545455	130.8102037	21.78381635	2.07972E-08	2547.896584	3151.194325	2547.896584	3151.194325
Numar Radacini 4 Frunze	238.4090909	20.5802639	11.58435538	2.80329E-06	190.9509172	285.8672646	190.9509172	285.8672646

Figure 3. Regression Analysis of No. Roots - 4 leaves and Grain Yield (kg/ha)

The independent variable (x-root number) was tested with the dependent variable (y-grain yield/hectare), the yield obtained extrapolated per hectare to assess the existence of a correlation relationship between the two. The statistical result $P<0.05$ denied the statistical independence hypothesis between the two factors and demonstrated a significant positive correlation between the number of roots developed in the 4 leaf stage and the yield obtained. The high R2 value shows that our linear regression very well explains the variability and relationship of the variables (Figure 3).
The plants height of 4-leaves is higher in the micro-granulate fertilized variant, with a 9 cm on average larger height, the increase from the fertilizer ensures a harmonious development above the ground, stimulating the growth of the foliar mass (Figure 4).
The statistical test shows a positive correlation between this variable and the yield obtained, and thus the plants with a larger size at this stage of development have a higher production.

In the 8-leaves stage, before hoeing and before the explosive maize growing period following assimilation of the extra nitrogen applied at hoeing, was determined the plant size, the number of roots, the length of the 7th most developed leaf at that moment.

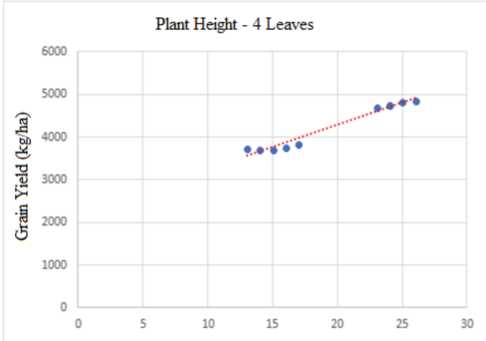

Figure 4. Linear regression between Plant Height-4 leaves and Grain Yield (kg/ha)

The radicular system of the test variant proves a much higher vitality with a higher number of roots, 11 compared to 7 in the control variant, with a longer root length and much more root hairs (Figure 5). This superior development of the radicular system correlates positively with seed production following statistical tests proving a direct proportional undeniable relationship between the number of roots and the final production.

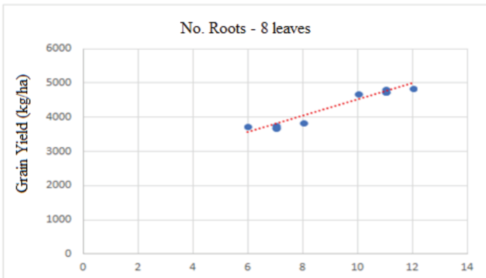

Figure 5. Linear regression between No. Roots - 8 leaves and Grain Yield (kg / ha)

The difference in the height of the plants in the 8-leaves stage further confirms the positive relationship between superior foliage development and higher yield. This superior development provides increased photosynthetic activity, resulting in an increase in the development gap between the plants included in the two experimental variants (Figure 6).

The size of the most developed leaf in this leaf-7th phase is a good example to illustrate the massive difference from the foliar canopy development, so for the fertilized micro-granulate variant we observe a leaf length of 7 inches of 31 cm on average compared to 16 cm in the case of the control variant (Figure 7). This superior development illustrates the advantage of early emergence due to microganized fertilization.

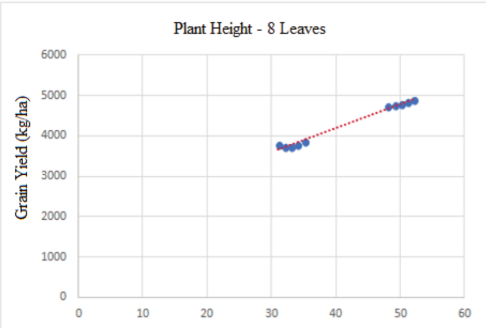

Figure 6. Linear regression between Plant height-8 leaves and Grain Yield (kg/ha)

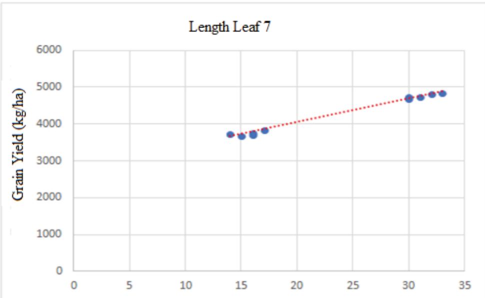

Figure 7. Linear regression between the length of leaf 7 and Grain Yield (kg/ha)

Two determinations were carried out to check the duration from emergence to blooming and the gap between the flowering of Male Inbred 1 and Female Inbred 2. The corn flowering studies show that the anthers of the masculine part, the tassel, open 5-7 days before the silks of the female, but the maximum amount of pollen is released after 2-3 days of flowering. This characteristic of corn makes it very important in the technology of producing maize to obtain a best nicking in the flowering of the two inflorescences so that the quantity of pollen is maximized (Figure 8).

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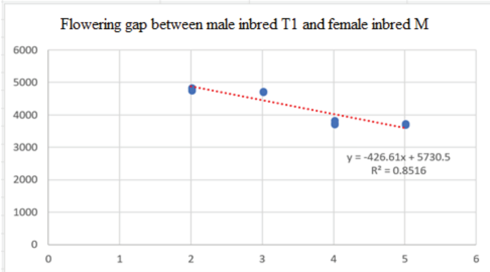

Figure 8. Linear regression between T1-M Flowering gap and Grain Yield (kg/ha)

Field determinations showed that for the tested variant the flowering gap between Male and Female was almost ideal at 2-3 days, providing a better supply of pollen and implicitly a higher percentage of pollination. In the case of the control variant, the gap between the two was 4-5 days due to longer exposure to drought conditions, which accentuated the protandric character of the masculine inflorescence.

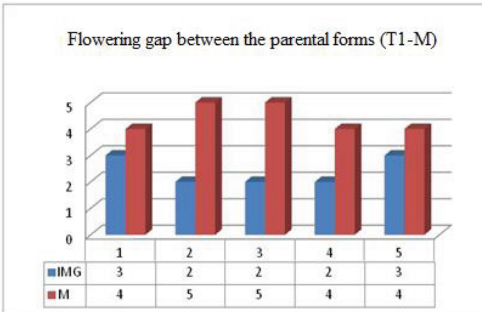

Figure 9. The graphical representation of the size of the gap in time of flowering between the parental forms

The statistical test shows a negative correlation between the delay at flowering of the two inbreds and production, the smaller delay between the two parent forms providing a better supply of pollen and a larger number of kernels on the ears.

Very important is the time from emergence to flowering, a shorter period being desirable to avoid the intense drought since mid-July and avoiding the increase of protandria. The positive effect of this variable being statistically proven, the correlation being inversely proportional to the lower the number of days between emergence and flowering, resulting in a higher yield (Figures 10, 11).

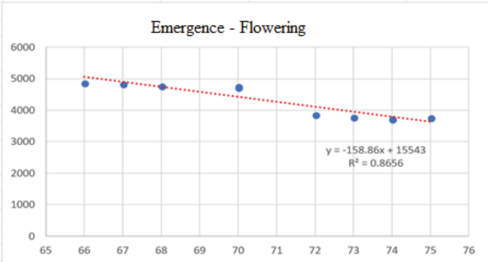

Figure 10. Linear regression between Emergence-Flowering time and Grain Yiled (kg/ha)

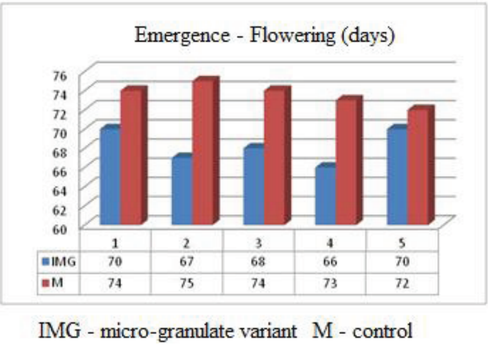

Figure 11. Graphical representation of the duration from emergence to flowering

An important determination was that of the number of kernels on the ears, which is a critical production item that plays a key role in determining production.

As a result of the analysis of the field samples, there is a higher number of grains on the ear in the case of the ears obtained in the experimental variant than in the control variant (Figure 12).

This is largely determined by the cumulative effect of faster growth of the fertilized plant with micro-granulated fertilizers and of ensuring optimal flowering nicking between parental forms.

The statistical analysis only confirms the strong correlation between the number of kernel on the ears and the final production, which is a corn production item.

In the experience we were able to highlight and demonstrate scientifically a number of quantitative and qualitative elements (number of leaves, waist, duration from emergence to flowering, the gap between parental forms, number of kernels on ear) that directly and significantly influence the production seed harvested within a hybridization parcel.

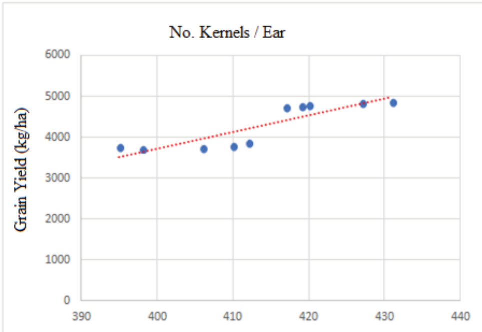

Figure 12. Linear regression between No. Kernels on ear and Grain Yield (kg/ha)

For each element, the plants fertilized with micro-granulated fertilizer at sowing proved to be clearly superior to those of the control variant, this being confirmed by the difference in production between the two variants of 1014 kg/ha in favor of the test variant.

CONCLUSIONS

The use of micro-granulated fertilizer in seed corn production brings an improvement in terms of the quality and quantity of the yield, thus giving seed growers a real help in the context of increases in required yields.

Early start of vegetation and the shortening of the planting-emergence period even with 1-2 days means a lot at flowering time and fecundation helping avoid effects of protandry and ensuring pollen viability.

The obtained results suggest that micro-granulated fertilizer applied at planting has a benefic effect on the plant with a quicker emergence, a better developed root system, shortening the time from emergence to flowering and the delay at flowering between the 2 inbred parentals and not the least superior yield.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.
We would like to thank Mr. Liviu Zbant, PhD. Agronomist engineer and Mr. Liviu Nicorici, Agronomist engineer for all their support in the field experience.

REFERENCES

Aldrich, S., (1984). Nitrogen management to minimize adverse effects on the environment. In R.D. Hauck (ed.) Nitrogen in crop production. ASA, CSSA, and SSSA, Madison, *WI*. 663–673.

Brady, N.C., Weil, R.R., (2008). Nature and Properties of Soils. Prentice-Hall, Inc., Upper Saddle River, New Jersey.

Crista, F., Boldea, M., Radulov, I, Crista, L., Lato, A., Dragomir, C., Berbecea, A., Okros, A., Lato, I., (2014). Changing the Quality of Maize Grain after Applying Micro-Granular Fertilizers. *Research Journal of Agricultural Science*, *46*(1), 166‒171.

Fox, R.H., J.M. Kern, and W.P. Piekielek, (1986). Nitrogen fertilizer source, and method and time of application effects on no-till corn yields and nitrogen uptake. *Agron. J.*, *78*, 741–746.

Lemarchand L., Revellat J., Benedetti, A. (2016). CVA Fertilizer Practice - Micro-Granular Fertilizers Market Study and Route-to-Market (www.corporate-value.com).

Jeffrey A.V. and Gyles W.R., (2003). Corn Production as Affected by Nitrogen Application Timming and Tillage. *Agronomy Journal. Published by American Society of Agronomy*, 502‒509.

Olson, R.A., and Kurtz, L.T. (1982). Crop nitrogen requirements, utili zation and fertilization. p. 567–604. In F. J. Stevenson (ed.) Nitrogen in agricultural soils. Agron. Monogr. 22. ASA, CSSA, and SSSA, Madison, WI.

Russelle, M.P., Deibert, E.J., Hauck, R.D., Stevanovic, M., Olson, R.A. (1981). Effects of water and nitrogen management on yield and 15N-depleted fertilizer use efficiency of irrigated corn. *Soil Sci Soc. Am. J.*, *45*, 553–558.

Stanley, R.L., Jr., and Rhoads, F.M. (1977). Effect of time, rate, and increment of applied fertilizer on nutrient uptake and yield of corn (*Zea mays* L.) *Proc. Soil Sci. Soc. Fla.*, *36*, 181–184.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

COMPARATIVE STUDY CONCERNING THE VARIABILITY OF FEW QUANTITATIVE CHARACTERS OF SOME NEW WHEAT GERMPLASM

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Abstract

An assortment of ten amphidiploids and twenty-five mutant/recombinant new genotypes plus their parental forms of winter wheat were studied for generating information on genetic variability of some quantitative characteristics. Field experiments were conducted at Caracal Agricultural Research and Development Station during 2015-2018. The data wer recorded after harvest and refers to plants height, number of fertile tillers, spike length, number of spikelets/spike, number of grains/spike, grains weight/spike, thousand grains weight and yield.
Analysis of variance revealed significant differences among the category of genotypes for all the characters due mostly to the influence of the climatic conditions of the region. The correlations established between the traits indicate same type of relationships, not very different depending on the method of obtaining of the genotypes and these could help researchers in the process of selection of genotypes with desired traits. For this experiment, it can conclude that some of the experimented material can be used in further breeding programmes. In the category of amphidiploid genotypes higher amplitude of variation was identified especially for yield while for the mutant/recombinant genotypes, characters variability was smaller and correlations were stronger. Also, for this category, values registered for the analised traits were closer to parental forms.

***Key words**: variability, correlations, amphidiploid wheat, mutant/recombinant germplasm.*

INTRODUCTION

Wheat is considered to be the most cultivated agricultural crop in the world. In our country, it is cultivated on approx. 25% of the arable land area and 40% of the area sown with cereals. Oltenia is one of the warmest regions of Romania, but where cereal crops still occupy vast areas. The Agricultural Research and Development Station Caracal is operating in a vast fertile area (the soil is clay-lily chernozem), but where climatic conditions are often restrictive (a strong aridization process has been installed in the last 20 to 30 years).
In the context of the expected climate change, the production of future cereals requires genotypes with good adaptability and various forms of resistance. NARDI Fundulea deals with the diversification of the genetic basis of wheat germplasm by using foreign introgreses or the use of appropriate modern techniques. Thus, a series of amphidiploids (introgression lines derived from wheat-derived hybrids x related species) and mutant/recombinant lines (obtained using a specific mutagenic protocol of two wheat genotypes, two irradiation cycles application, hybridization and DH technology using *Zea* system) (Giura A., 2013) and distributed within a research project (ADER 116) for cultivation under various conditions. Crops improved through biotechnology are producing higher yields and of quality (Iancu et al., 2018).
Induced mutations are now being widely used for developing improved crop varieties and for the discovery of genes controlling important traits and understanding the functions and mechanisms of actions of these genes (Singh et al., 2014). According to the federal government of Mutant Variety Database, thousands of new varieties have been created through mutation breeding, either by radiation like gamma rays, thermal neutrons, X-rays or by exposure to certain chemicals.
Consistent efforts are made to improve and/or sustain wheat production and the development of new varieties is based now on the advanced genetics. Amphidiploids are plants created by interspecific or intergeneric hybridization incoporating diploid chromosome complexes of two parental components. Also, the application of mutation techniques has generated a vast amount of genetic variability and is playing a significant role in plant breeding. Mustăţea and

Săulescu (2011) remarked that the grain yield increase in recently created winter wheat varieties was entirely due to an increase in the number of kernels per unit area, while the grain weight showed a negative, but relatively smaller trend.
This paper presents the way of their behavior for several elements of productivity and the types of correlations between them, under ecological conditions specific to the southern part of Romania in order to increase the genetic variability.

MATHERIALS AND METHODS

The biological material was obtained at the Laboratory of Genetics of NARDI Fundulea and was represented by 10 amphidiploid lines and 25 mutant/recombinant lines, as well as their parental forms, Izvor variety and the improved line F00628-34. Amphidiploid lines come from crosses between *T. durum* genotype and *Ae. squarrossa* biotipes (http://www.incda-fundulea.ro/cercet/ader/ader116.pdf).
Field trials were sowed every year in the autumn using randomised blocks method in three replicates at ARDS Caracal (44"7' N latitude 24"21' E longitude, 98 m altitude) on a chernozem soil (pH 7.5-7.7, humus 3.18%) with rape, sun-flower and maize as preceding crop. Standard crop technolgy was applied. At harvest, the analized characters were made for 50 plants and yield was determined by weithering the hole quantity of grains resulted from the plots. Plant height (cm) was measured at maturity stage from ground level up to the tip of spike of main tiller excluding awns. For TKW (g), 1000 seeds were counted randomly by seed counter and weighted by electric balance. The values presented in the tables represent the average of the three years calculated using analysis of variance method and correlation index.
The climate is temperate-continental with mediteranean influences, characterized through alternate frost-defrost winters and 2-6 drought month with maximum precipitation in June and minimum in August-September. The annual average temperature is about 11°C, of January -3...-20° and July from 20 to above 23°C. Rainfall amount during the winter wheat vegetation period (October to July) totalled 477.6 mm in 2015/2016, 457.0 mm in 2016/2017 and 658.4 mm in 2017/2018.

RESULTS AND DISSCUTIONS

Testing these genotypes for high and stable production was the subject of this scientific paper.
Precipitation during the wheat growing season is expected to have a positive impact on wheat yield. Environmental conditions from the experimentation years were suitable for the examined germplasm, excepting the last.
The choice of varieties resistant to thermal stress and water stress (drought or drought-induced drought) plays an important role in the fight against this wheat plant suffering.
Wheat height is a character for which there is a requirement of both dwarf and tall types. Tested amphidiploids proved to be tall and with no resistance to fall. The registered range of 98.40-126.00 cm for plants height (Table 1) was the maximum values comparative with the other categories, 89.00-108.00 cm for mutant/recombinant (Table 3), respectivelly 80.00-97.00 cm for parental forms (Table 5). 7.23% value for variability coefficient indicate a uniformity of plants as concern height. Other characters, registered medium variability (no. of spikelets/spike, spike length, no. of grains/spike and no. of fertile tillers and yield) to large (grains weight/spike). During the three years the mean plant height value for the amphidiploid germplasm was higher both than mutant/recombinat material and the parental forms.
No. of fertile tillers was of 9.53 in average of three years, a mean closer to the one realised by parental forms (9.68) and higher that mutant/recombinat (8.58).
Knežević et al., (2008) apreciate that one of the most important and promising direction in improvement of grain yield of wheat is the long and fertile spike. In this experiment amphidiploid material presented a mean value of 10.40 cm with range between 8.00 and 13.30 cm and medium variability of 13.49%.
Grains weight/spike varied in large limits because the particularity year of 2017-2018, which presented a drought spring and a very wet summer which leaded to the obtaining of smaller, wrinkled and sprouted grains in

harvesting time. Also, this material have longer vegetation period comparative with mutant/recombinat and parental forms. But, grains of amphidiploids proved to be havier and as Mandea et al. (2016) said seed weight and genetic variability their components (length, width, shape, density) and how they react to environmental conditions, can contribute to increased genetic progress in breeding for yield and yield stability.

Table 1. Values for amphidiploids (2015 – 2018)

Character	Mean ± a	Limits		s%
		Min.	Max.	
Plants height (cm)	108.58 ± 7.85	98.40	126.00	7.23
No. of fertile tillers	9.53 ± 1.73	7.10	13.80	18.16
Spike length (cm)	10.40 ± 1.40	8.00	13.30	13.49
No. of spikelets/spike	19.33 ± 2.42	16.20	28.20	12.50
No. of grains/spike	43.37 ± 6.73	32.10	56.60	15.53
Grains weight /spike (g)	2.88 ± 0.79	1.65	4.80	27.46
Thousand grain weight (g)	48.49 ± 5.61	41.52	53.60	11.58
Yield (Kg/ha)	3122.10 ± 1579,16	843.00	5980.00	50.58

The highest values for coefficient of variation was shown by grain yield (50.58%) fallowed by grains weight/spike (27.46%). Most of this variation was produced by the different conditions of the experimentation years.

Sinthetic amphidiploid germplasm indicate wide variation in field conditions. Most of the amphidiploids possess higher grains size, but well dressed and not easiliy beated. On a study with amphidiploids features, Stoyanov H.P. (2014) reported slight variations in morphological and physiological indicators, which highlights their genetic stability and high level of homozygosity. DH lines may increase the genetic variability of winter wheat and can be selected forms with valuable quantitative characters (Iancu et al., 2016)

Correlation studies provide a better understanding of the association of different characters with grain yield (Yousaf et al., 2008).

The experimented yield components indicate varying trends of association among themselves. In the case of amphidiploids, there were identified negative correlations between thousand grain mass and plants length, number of fertile tillers, spike length, number of spikelets/spike, grains number and weight/spike and yield (Table 2).

Height of plants was in positive correlation with spike lenght, number of spikelets/spike and grains weight/spike. Same type of correlations reported Knežević et al. (2008).

Table 2. Correlation indices for amphidiploid germplasm

	Stem length	No. of fertile tillers	Spike length	No. of spikelets/ spike	No. of grains/ spike	Grains weight/ spike	Yield
No. of fertile tillers	0.620						
Spike length	0.969	0.693					
No. of spikeletes/spike	0.919	0.693	0.985				
No. of grains/spike	0.964	0.695	0.985	0.917			
Grains weght/spike	0.986	0.687	0.985	0.945	0.985		
Yield	0.976	0.692	0.980	0.901	0.984	0.987	
TGW	-0.858	-0.649	-0.912	-0.822	-0.938	-0.889	-0.900

P = 0.349 %

In a similar experiment, of three year and 10 locations, some authors sustain that the most of the correlations among the yield components were negative, illustrating the difficulty of combining in the same cultivar high values of more than one component, because of compensation between yield components (Mandea et al., 2019). Same authors suggest that cultivars showing positive or small negative deviations from the regressions between negatively correlated yield components might be useful in breeding for reducing compensations between yield components. It is well known that grain yield is a complex trait and high y influenced both, by genetic factors and environmental variations. Selection, as a breeding method, to be successful depends upon the information on the genetic variability of characters and the association of the morpho-agronomic traits with grain yield.

The mutant/recombinant germplasm exhibited high production capacity and good resistance to the main climate risk factors in the southern area. The average of 7368.10 kg/ha is a fairly high value for the experiment area. The variation limits were between 5571.00 and 8627.00 kg/ha with medium variability (12.07%) (Table 3). The other characters presented lower variability which means that in time these were uniforms.This material might be suitable for drought tolerance breeding because of the better adaptation, behavior concerning the fluctuation of the timing of rain between vegetation period.

Table 3. Values for mutante/recombinant (2015-2018)

Character	Mean ± a	Limits		s%
		Min.	Max.	
Plants height (cm)	94.37 ± 4.56	89.00	108.00	4.83
No. of fertile tillers	8.58 ± 0.732	7.20	10.20	8.47
Spike length (cm)	10.76 ± 0.70	10.00	12.40	6.52
No. of spikelets/spike	20.05 ± 0.99	18.80	22.60	4.95
No. of grains/spike	61.52 ± 3.74	56.20	70.20	6.07
Grains weight /spike (g)	3.09 ± 0.20	2.85	3.54	6.43
Thousand grain weight (g)	37.75 ± 1.97	33.29	40.35	5.22
Yield (Kg/ha)	7368.10 ± 889.34	5571.00	8627.00	12.07

For the mutant/recombinant germplasm, number of fertile tillers per plant had negative correlation with plant height and spike length. Same type of correlation was identified with the other analysed characters, excepting thousand grain weight. Yield was positively correlated with stem and spike length, no. of spikelets/spike, no. of grains/spike and grains weight/spike (Table 4).

Similar results reported Gholizadeh et al. (2017), but in the salinity-stressed environment. In a study as concern response of some new wheat genotypes to nitrogen fertilization and prospects of yield breeding based on yield elements, Iancu et al., 2019 reported highest positive correlations between yield and grain weight/spike, yield and HW and HW and grain weight/spike.

Table 4. Correlation indices for mutant/recombinant germplasm

	Stem length	No. of fertile tillers	Spike length	No. of spikelets/ spike	No. of grains/ spike	Grains weight/ spike	Yield
No. of fertile tillers	-0,651						
Spike length	0,969	-0,573					
No. of spikeletes/spike	0,981	-0,623	0,975				
No. of grains/spike	0,983	-0,642	0,977	0,987			
Grains weght/spike	0,987	-0,679	0,959	0,980	0,985		
Yield	0,953	-0,619	0,931	0,958	0,968	0,967	
TGW	-0,971	0,605	-0,989	-0,977	-0,979	-0,961	-0,926

P = 0.325%

In average for the three years of experimentation, mutant/recombinant material registered superior values comparative with parental forms as concern almost all characters.

Other authors also consider that mutant/recombinant DH lines of wheat can be considered as an interesting material for breeding programs or an important tool for releasing of new genes sources (Dobre et al., 2018).

Parental forms have a good behavior to the diverse climatic conditions of the years. The results demonstrated stability parameters for the tested characters and indicate the these cultivars are stabile in reaction to environment changes.

Table 5. Values for parental forms (2015-2018)

Character	Mean ± a	Limits		s%
		Min.	Max.	
Plants height (cm)	87.63 ± 5.62	80.00	97.00	6.41
No. of fertile tillers	9.68 ± 0.71	9.10	11.00	7.37
Spike length (cm)	9.18 ± 1.16	8.00	10.80	12.63
No. of spikelets/spike	18.10 ± 1.39	16.10	20.40	7.71
No. of grains/spike	52.30 ± 2.88	48.80	56.60	5.50
Grains weight /spike (g)	2.63 ± 0.32	2.24	3.07	12.37
Thousand grain weight (g)	42.90 ± 4.06	38.70	48.55	9.48
Yield (Kg/ha)	6081,83 ± 1412,551	4150.00	7760.00	23,22

Weather conditions are just one part of a the variation. Germplasm reaction to environmental changes was different, but in favorable year conditions, performed better in this area for studied traits so generally speaking it an appreciate that this material is adapted to South Romania conditions and gave stable yields. As Dimitrijević D. et al., (2002) said, adaptability is a natural reaction of genotype in order to survive and reproduce while for agriculture, stability represents desirable reaction of cultivated genotypes.

Also, Racz et al. (2015) stated that a measure to counter the negative effects caused by climatic changes is the creation of new genotypes with better resistance or tolerance to adverse changes or which avoid stresses by the fact that the critical phases of development do not coincide with these conditions. Some triticale varieties and lines bred also by RICIC Funculea and cultivated many years in different climate pattern then were developed indicate yield variability caused by high temperature, and small amounts of precipitations (Butnaru et al., 2014). Another specie with high genetic yield potential, adapted to local condition, but variable depending on rainfall and moisture is groundnut (Soare et al., 2014).

CONCLUSIONS

Experienced biological material can be considered to have superior agronomic features, is well suited to the experimental area compared to other genotypes existing in culture as well as to the expected climate changes.

Althought environmental conditions have bigger influence upon yield components, it can be made a direct selection for grain yield related traits which increases the chance of improving wheat grain yield using this germplasm.

The mutant/recombinant germplasm with the best values indicate the possibility of selecting them as new genotypes which combine the desirable values for yield components.

REFERENCES

Butnaru, G., Sarac, I., Ciulca, S. (2014). Relationship among yield and plant specific traits on triticale Romanian varieties in Timisoara environment. Communications in *Agricultural and Applied Biological Sciences*, *79*(4), 201–210.

Dimitrijević, M., Knežević, D., Petrović, S., Zečević, V. (2002). Variability and stability of harvest index in wheat (*Triticum aestivum* L.). *Kragujevac J. Sci., 24*, 91–96.

(Dobre) Barbu, S.P., Giura, A., Lazăr, C. (2018). Potential sources of new genetic variability in mutant and mutant/recombinant wheat DH-lines. *Romanian Agricultural Research*, *35*, 81–87.

Giura, A. (2013). Wheat mutagenesis by combining recurrent irradiation, hibridization and DH-technology. *Journal of Horticulture, Forestry and Biotechnology, 17*(4), 114–118.

Gholizadeh, A., Dehghani, H. (2017). Correlation and sequential path analysis between yield and related characters of wheat (*Triticum aestivum* L.) genotypes in non-stressed and salinity-stressed conditions. *Romanian Agricultural Research*, *34*, 37–49.

Iancu, P., Păniţă, O., Soare, M. (2019). Response of some new wheat genotypes to nitrogen fertilization and prospects of yield breeding based on yield elements. *Romanian Agricultural Research*, *36*(1), 1–9.

Iancu, P., Soare, M., Soare, R. (2018). Quality characteristics of whole grains flour of some mutant/ recombinant winter wheat lines. *18th International Multidisciplinary Scientific Geoconference, Nano, Bio, Green and Space Technologies for a Sustainable Future, 18,6*(2), 489–496.

Iancu, P., Soare, M., Bonciu, E. (2016). Experimental results concerning the behavior of some synthetic amphidiploid of wheat in the conditions of Caracal Research Station. *SGEM 16th International Multidisciplinary Scientific Geoconference, 6*(3), 269–276.

Knežević, D., Zečević, V., Đukić, N., Dodig, D. (2008). Genetic and phenotypic variability of grain mass per spike of winter wheat genotypes (*Triticum aestivum* L.). *Kragujevac J. Sci*., *30*. 131–136.

Mandea, V., Mustățea, P., Marinciu, C.M., Şerban, G., Melucă, C., Păunescu, G., Isticioaia, S.F., Dragomir, C., Bunta, G., Filiche, E., Voinea, L., Lobonţiu, I., Domokos, Z., Voica, M., Ittu, G., Săulescu, N.N. (2019). Yield components compensation in winter wheat (*Triticum aestivum* L.) is cultivar dependent. *Romanian Agricultural Research*, *36*, 1–7.

Mandea, V., Mustățea, P., Săulescu, N.N. (2016). Cultivar and environment effects on grain weight and size variation in winter wheat, grown in a semi-continental climate. *Romanian Agricultural Research*, *33*, 23–28.

Mustăţea, P., Săulescu, N.N. (2011). Estimation of genetic trends in yield and agronomic traits of recent Romanian winter wheat (*Triticum aestivum* L.) cultivars, using direct comparisons in multiyear, multi-location yield trials. *Romanian Agricultural Research*, *28*, 17–24.

Racz, I., Kadar Rozalia, Moldovan, V., Haș, I. (2015). Performance and stability of grain yield and yield components in some winter wheat varieties. *Romanian Agricultural Research*, *32*, 11–18.

Singh, R., Tiwari, R., Sharma, D., Tiwari, V., and Sharma, I. (2014). Mutagenesis for wheat improvement in the genomics era. *Journal of Wheat Research*, *6*(2), 120–125.

Soare, M., Iancu, P., Soare, R., Bonciu, E., Panita, O. (2014). Results concerning the productive capacity and chemical composition of groundnuts cultivated on the sandy soils from Southern Romania. *14th Geo Conference on Nano, Bio and Green – Technologies for a Sustainable Future. I.* 479–485.

Stoyanov, H.P. (2014). New amphidiploid wheat species (Nothosp. Nov.) as a result of artificial hybridisation. *Scientific Papers. Series A. Agronomy*, *LVII*, 331–338.

Yousaf, A., Babar, M.A., Javed, A, Monneveux, P., Zahid, L. (2008). Genetic variability, association and diversity studies in wheat (*Triticum aestivum* L.) germplasm. Pak. *J. Bot., 40*(5), 2087–2097.

http://www.mvd.iaea.org/

http://www.incda-fundulea.ro/cercet/ader/ader116.pdf

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF FOLIAR FERTILIZATION ON THE PHENOLOGICAL DEVELOPMENT OF RAPESEED

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Abstract

The experiment is conducted in 2012-2015 in the Training and Experimental Base of the Department of Plant Growing at Agricultural University of Plovdiv. The experiment is set in block mode with four repetitions, with size of the cultivated plot of 20 m^2, with winter rapeseed type - the Visby hybrid. The following leaf fertilizers were tested: Lactofol B - 4000 ml/ha, Litovit® - 2000 g/ha and Fertiactyl Starter - 3000 ml/ha, and untreated control variant. The rapeseed sowing was done 1-10. IX, 1-10. X, 10-20. X, 20-30. X. The leaf fertilization was performed in the 2-4 leaf stages for the first two sowing periods and in 1-2 leaf stages for the second two sowing periods. As a result of the experiment, leaf fertilization has been found to affect the development of the phenophases. The strongest stimulation effect during the three years of the experiment at the different sowing periods is obtained in the variant treated with Fertiactyl Starter - 3000 ml/ha.

Key words: *rapeseed, leaf fertilization, development phases.*

INTRODUCTION

Foliar fertilization is not opposed to soil one but is considered a necessary additional activity in the overall system of optimal mineral nutrition of plants.

While nutrient elements in the soil may have synergistic and antagonistic relationships between them, an important advantage of leaf fertilization is that the problem of elemental antagonism is eliminated. Another advantage of their use is the conservation of the natural ecological parameters of the environment.

The small amount of rainfall during the preparation of the soil and the sowing of rapeseed leads to its later germination and the inability of the plants to enter the optimum phase for wintering. By using foliar fertilizers at different stages of rapeseed development, we can accelerate its development to reach a phase more suitable for wintering.

There are lots of such studies for other agricultural crops, both in the world and in Bulgaria (Coelho H. et al., 2011; Delchev G., 2010; Sanmueang A. et al., 2011; Kolev T. et al., 2012; Kolev T. et al., 2013), but for rapeseed they are insufficient or completely absent.

It is precisely for this purpose that we study the influence of some foliar fertilizers and application phases on the development and productivity of oilseed rape.

In this article we look at one part of the experiment, i.e. the influence of certain foliar fertilizers and application phases on the phenological development of rapeseed.

MATERIALS AND METHODS

The study was conducted in the period 2012-2015 in the area of Training, experimental and implementation base of the Department of Plant Growing at Agricultural University - Plovdiv.

The experiment is based on the block method, repeated 4 times, with a trial plot of 20 m^2, with a Visby hybrid originating in Germany.

Experiment variants:

Factor A - sowing dates

Factor B - foliar fertilizers

Factor C - development phases

I. Date of sowing 1-10.IX

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4 leaf.

- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4 leaf.
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4 leaf.

II. Date of sowing 1-10.X

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4 leaf.
- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4 leaf.
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4 leaf.

III. Date of sowing 10-20.X

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase-1-2 leaf.
- Spraying with Lithovit® - 2000 g/ha - phenophase-1-2 leaf.
- Spraying with Fertiactyl Starter - 3000 ml/ha-1-2 leaf.

IV. Date of sowing 20-30.X

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase-1-2 leaf.
- Spraying with Lithovit® - 2000 g/ha - phenophase-1-2 leaf.
- Spraying with Fertiactyl Starter -3000 ml/ha-1-2 leaf.

The experiment was carried out after a precursor of wheat. Immediately after its harvesting, ploughing was carried out at a depth of 18-20 cm, followed by a two-fold cross-disking and pre-sowing rolling. 100 kg/ha Phosphorus, 80 kg/ha Potassium are inserted into the soil with the main processing of the soil, as well as 170 kg/ha Nitrogen, of which 30 kg/ha in autumn with the pre-sowing cultivation, and the remainder at the earliest opportunity in spring.

Sowing is at 12-15 cm distance between rows and at a sowing rate of 6 kg/ha, providing a density of 60 plants/m^2. Seeds are sown at a depth of 2-3 cm with a seed drill. After sowing, the land is rolled. The phenological development of rapeseed has been monitored in the sown variants.

Regarding the agro-climatic characteristics during the experiment period, the meteorological factors (air temperature and rainfall), their combination and distribution through vegetation, which determine the growth and development of the crop during the years of cultivation, have a major influence.

The data characterizing these factors over the three experimental years in the study area are shown in Figure 1. It can be seen that during the three years of the study, significant deviations from the average monthly temperatures in the experiment area compared to the multiannual period are not observed.

Greater differences are seen in terms of moisture. Over the three years of the experiment, the highest amount of rainfall in the cultivated area during September - July period was reported in 2014-2015 (856.4 mm). They exceed with 422.9 mm those of the multiannual period (433.5 mm).

All three years of study are characterized by sufficient amount of moisture in the critical phases of the development of the crop, with the exception of the sowing-germination period during the first two years.

The absolute minimum temperature during the three years of the study was reported in December -9.8°C in 2012-2013, -10.4°C in 2013-2014, while in 2014-2015 - in January -14.2°C (Figure 2).

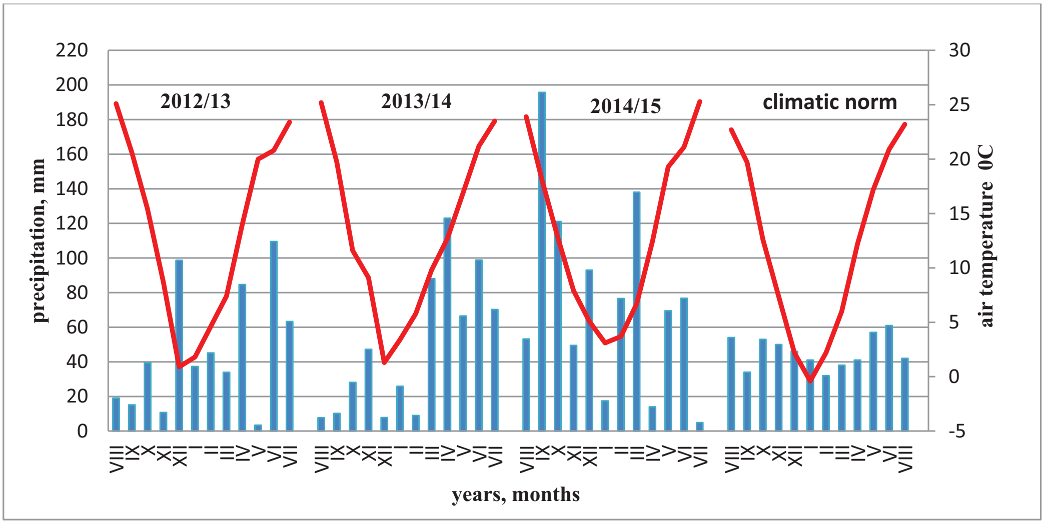

Figure 1. Average monthly temperatures and sum of precipitation during the years of study in the area of the Training, experimental and implementation base

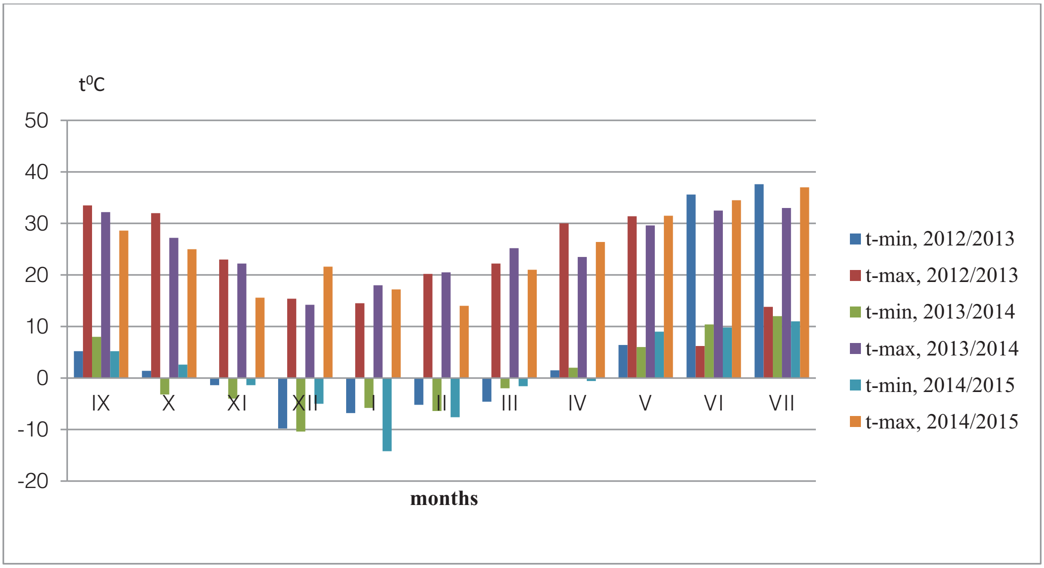

Figure 2. Absolute minimum and maximum temperatures by months 2012/2015

RESULTS AND DISCUSSIONS

Depending on the year, the date of sowing, weather conditions, and used foliar fertilizers phases of development in the area of Training, experimental and implementation base occur at different times (Tables 1, 2, 3 and 4).

The results in the tables show that during the three years of the study, the plants germinate at the earliest at the first sowing date of 1-10.IX, followed by those sown in the periods 1-10.X; 10-20.X and 20-30.X.

Higher temperatures during the germination period at the earliest date of sowing create conditions for the plants to germinate for 21 days in 2012. More balanced distribution of precipitation in 2014 and in 2013 led to the germination of the plants for 16 and 18 days at the first date of sowing, while in the sowing from 10 to 20.X, during the three years of study plants germinate for 18 to 20 days.

During the three years of study, for the remaining sowing dates, the plants germinate for 25 and 18 days at sowing from 01 to 10.X. and for 16, 18 and 14 days at sowing period from 20 to 30.X.

At the first two sowing dates, all tested variants in the three years of study entered the 6-8 leaf stage, about one month after germination.

At sowing in the period of 10 to 20.X, only the Fertiactyl Starter-treated plants in the first year and those treated with Fertiactyl Starter and Lactofol B in the second and third years reached the 6-8 leaf phase.

Late sowing of rapeseed at the end of October (from 20 to 30.X) is the reason for ending the vegetation of plants in all variants at 2-4 leaf phase, with the exception of Fertiactyl Starter-treated variants that reach 4-6 leaf phase in 2013 and 2014.

Phase 6-8 leaf in all sowing dates and over the three years of the experiment were reported for Fertiactyl Starter treated variants with 3 to 6 days earlier than the control variant.

Falling temperatures in December 2012 to minus 9.8°C, and the failure of rapeseed to reach the optimum wintering phase at the last sowing date resulted in plant deaths in Lactofol B, Litovit® treated variants and of the control variants.

The development phases in 2012-2013 were only reported for the Fertiactyl Starter variant.

Temperatures in December 2014 reached minus 5°C, while in 2013 - minus 10.4°C but due to the snow cover and higher average day and night temperatures during this period led to the successful wintering of part of the plants during the two years of study being sown on the last sowing date.

The rise in temperatures at the end of February 2013 to 10.3°C and the 45.1 mm precipitations that had fallen resulted in the earliest resumption of vegetation at all dates and variants of sowing (26.02).

Fallen 76.6 mm precipitation in February of 2015 and a gradual increase in temperatures lead to a later resumption of vegetation (4.03).

The alternation of lower and higher temperatures in February 2014 and less rainfall of 9 mm are a prerequisite for the latest resuming of vegetation (9.03).

Once the vegetation has resumed, the development phases during the three years of the study occur at the earliest at sowing from 1 to 10.IX, followed by sowing in the period 1-10. X, 10-20. X, and 20-30.X.

The rainfall in February in 2013 (45.1 mm) creates a prerequisite for the stem formation phase for all variants and sowing dates to occur at the earliest, i.e. from 14.03 to 6.04.

Later onset of vegetation in 2014 and less rainfall in February (9.0 mm) this year lead to a delay in the stem formation phase. For all tested variants and sowing dates, this phase runs the latest - from 21.03 to 12.04.

The gradual rise in temperature and 76.6 mm precipitation in February 2015 led to a stem formation phase from 17.03 to 9.04.

The earliest phase of stem formation during the three years of study was reported in the Fertiactyl Starter variants (14.03-6.04 in 2013, 21.03-09.04 in 2014, 17.03-6.04 in 2015) followed by the Lactofol B variants (15.03-27.03 in 2013, 22.03-10.04 in 2014, 17.03-7.04 in 2015), Litovit® (15.03-28.03 in 2013, 22.03-11.04 in 2014, 18.03-8.04 in 2015), and control (16.03-29.03 in 2013, 23.03-12.04 in 2014, 19.03-9.04 in 2015).

Plants of all tested variants, during the three years of the experiment, entered budding phase in April.

Of all the variants studied, the earliest budding phase was recorded in Fertiactyl Starter-treated variants followed by Lactofol B, Litovit® treated variants and the control.

Budding phase over the three years of the study occurred from 1 to 4 days earlier in the Fertiactyl Starter treated versus the other variants and the control.

The mass blossoming phase was first reported in Fertiactyl Starter treated plants (15.04 to 4.05 in 2013, 20.04 to 7.05 in 2014, and 16.04 to 5.05 in 2015) followed by those treated with Lactofol B (16.04-27.04 in 2013, 21.04 - 9.05 in 2014, and 17.04 to 6.05 in 2015), Litovit® (17.04-28.04 in 2013, 22.04-9.05 in 2014 and from 18.04 to 07.05 in 2015) and the control variant (17.04-29.04 in 2013, 22.04-10.05 in 2014 and from 19.04 to 8.05 in 2015).

Table 1. Phenological development of rapeseed sown in the period of 1 to 10. IX

Development phases	Sowing			Germination			1-2 leaf			2-4 leaf		
Sowing date	1-10. IX											
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
Control	07.09	06.09	10.09	28.09	24.09	26.09	05.10	02.10	04.10	14.10	11.10	12.10
Fertiactyl Starter	07.09	06.09	10.09	28.09	24.09	26.09	05.10	02.10	04.10	14.10	11.10	12.10
Litovit®	07.09	06.09	10.09	28.09	24.09	26.09	05.10	02.10	04.10	14.10	11.10	12.10
Lactofol B	07.09	06.09	10.09	28.09	24.09	26.09	05.10	02.10	04.10	14.10	11.10	12.10

Development phases	4-6 leaf			6-8 leaf			Vegetation termination			Vegetation resuming		
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2013	2014	2015
Control	24.10	23.10	24.10	02.11	01.11	01.11	13.12	14.12	17.12	26.02	09.03	04.03
Fertiactyl Starter	22.10	20.10	20.10	29.10	27.10	26.10	13.12	14.12	17.12	26.02	09.03	04.03
Litovit®	23.10	22.10	23.10	30.10	29.10	29.10	13.12	14.12	17.12	26.02	09.03	04.03
Lactofol B	23.10	21.10	21.10	30.10	28.10	27.10	13.12	14.12	17.12	26.02	09.03	04.03

Development phases	Stem formation			Budding			Start of blossoming – 10%			Mass blossoming – 75%		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	16.03	23.03	19.03	04.04	08.04	05.04	09.04	14.04	11.04	17.04	22.04	19.04
Fertiactyl Starter	14.03	21.03	17.03	02.04	06.04	02.04	07.04	12.04	08.04	15.04	20.04	16.04
Litovit®	15.03	22.03	18.03	03.04	07.04	04.04	09.04	14.04	10.04	17.04	22.04	18.04
Lactofol B	15.03	22.03	17.03	03.04	07.04	03.04	08.04	13.04	09.04	16.04	21.04	17.04

Development phases	Waxy maturity			Full maturity			Vegetation period		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	04.06	09.06	07.06	18.06	24.06	22.06	263	273	269
Fertiactyl Starter	02.06	07.06	04.06	16.06	22.06	20.06	261	271	267
Litovit®	04.06	09.06	06.06	18.06	24.06	22.06	263	273	269
Lactofol B	03.06	08.06	05.06	17.06	23.06	21.06	262	272	268

Table 2. Phenological development of rapeseed sown in the period of 1 to 10. X

Development phases	Sowing			Germination			1-2 leaf			2-4 leaf		
Sowing date	1-10. X											
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
Control	05.10	07.10	06.10	30.10	25.10	24.10	07.11	02.11	01.11	17.11	13.11	12.11
Fertiactyl Starter	05.10	07.10	06.10	30.10	25.10	24.10	07.11	02.11	01.11	17.11	13.11	12.11
Litovit®	05.10	07.10	06.10	30.10	25.10	24.10	07.11	02.11	01.11	17.11	13.11	12.11
Lactofol B	05.10	07.10	06.10	30.10	25.10	24.10	07.11	02.11	01.11	17.11	13.11	12.11

Development phases	4-6 leaf			6-8 leaf			Vegetation termination			Vegetation resuming		
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2013	2014	2015
Control	27.11	24.11	23.11	05.12	03.12	03.12	13.12	14.12	17.12	26.02	09.03	04.03
Fertiactyl Starter	23.11	20.11	19.11	02.12	30.11	29.11	13.12	14.12	17.12	26.02	09.03	04.03
Litovit®	26.11	23.11	22.11	04.12	02.12	02.12	13.12	14.12	17.12	26.02	09.03	04.03
Lactofol B	25.11	22.11	21.11	03.12	01.12	01.12	13.12	14.12	17.12	26.02	09.03	04.03

Development phases	Stem formation			Budding			Start of blossoming – 10%			Mass blossoming – 75%		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	23.03	30.03	26.03	10.04	16.04	12.04	16.04	22.04	18.04	24.04	30.04	26.04
Fertiactyl Starter	20.03	27.03	23.03	07.04	13.04	09.04	13.04	19.04	15.04	21.04	25.04	21.04
Litovit®	22.03	29.03	25.03	09.04	15.04	11.04	14.04	21.04	17.04	23.04	29.04	25.04
Lactofol B	21.03	29.03	24.03	08.04	14.04	10.04	14.04	20.04	16.04	22.04	26.04	22.04

Development phases	Waxy maturity			Full maturity			Vegetation period		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	14.06	19.06	17.06	28.06	04.07	02.07	241	252	250
Fertiactyl Starter	11.06	16.06	14.06	25.06	01.07	29.06	238	249	247
Litovit®	13.06	18.06	16.06	27.06	03.07	01.07	240	251	249
Lactofol B	12.06	17.06	15.06	26.06	02.07	30.06	239	250	248

Table 3. Phenological development of rapeseed sown in the period of 10 to 20. X

Development phases	Sowing			Germination			1-2 leaf			2-4 leaf		
Sowing date	10-20. X											
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
Control	15.10	15.10	14.10	04.11	02.11	01.11	14.11	12.11	11.11	25.11	24.11	24.11
Fertiactyl Starter	15.10	15.10	14.10	04.11	02.11	01.11	14.11	12.11	11.11	22.11	21.11	20.11
Litovit®	15.10	15.10	14.10	04.11	02.11	01.11	14.11	12.11	11.11	24.11	23.11	23.11
Lactofol B	15.10	15.10	14.10	04.11	02.11	01.11	14.11	12.11	11.11	23.11	22.11	21.11

Development phases	4-6 leaf			6-8 leaf			Vegetation termination			Vegetation resuming		
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2013	2014	2015
Control	05.12	04.12	04.12	-	-	-	13.12	14.12	17.12	26.02	09.03	04.03
Fertiactyl Starter	30.11	29.11	28.11	09.12	08.12	07.12	13.12	14.12	17.12	26.02	09.03	04.03
Litovit®	04.12	03.12	03.12	-	-	-	13.12	14.12	17.12	26.02	09.03	04.03
Lactofol B	03.12	02.12	01.12	-	10.12	09.12	13.12	14.12	17.12	26.02	09.03	04.03

Development phases	Stem formation			Budding			Start of blossoming – 10%			Mass blossoming – 75%		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	29.03	04.04	01.04	15.04	20.04	17.04	21.04	26.04	23.04	29.04	04.05	01.05
Fertiactyl Starter	25.03	01.04	29.03	11.04	16.04	13.04	16.04	21.04	18.04	24.04	29.04	26.04
Litovit®	28.03	03.04	31.03	14.04	18.04	15.04	20.04	24.04	21.04	28.04	02.05	29.04
Lactofol B	27.03	02.04	30.03	13.04	17.04	14.04	19.04	22.04	19.04	27.04	01.05	28.04

Development phases	Waxy maturity			Full maturity			Vegetation period		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	20.06	24.06	23.06	05.07	09.07	08.07	243	249	249
Fertiactyl Starter	15.06	20.06	19.06	01.07	05.07	04.07	239	245	245
Litovit®	19.06	22.06	21.06	04.07	08.07	07.07	242	248	248
Lactofol B	18.06	21.06	20.06	03.07	07.07	06.07	241	247	247

Table 4. Phenological development of rapeseed sown in the period of 20 to 30. X

Development phases	Sowing			Germination			1-2 leaf			2-4 leaf		
Sowing date	20-30. X											
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
Control	25.10	25.10	22.10	10.11	12.11	08.11	21.11	23.11	20.11	07.12	08.12	07.12
Fertiactyl Starter	25.10	25.10	22.10	10.11	12.11	08.11	21.11	23.11	20.11	04.12	05.12	03.12
Litovit®	25.10	25.10	22.10	10.11	12.11	08.11	21.11	23.11	20.11	06.12	07.12	06.12
Lactofol B	25.10	25.10	22.10	10.11	12.11	08.11	21.11	23.11	20.11	05.12	06.12	04.12
Development phases	4-6 leaf			6-8 leaf			Vegetation termination			Vegetation resuming		
Years and variants	2012	2013	2014	2012	2013	2014	2012	2013	2014	2013	2014	2015
Control	-	-		-	-	-	13.12	14.12	17.12	-	09.03	04.03
Fertiactyl Starter	-	14.12	13.12	-	-	-	13.12	14.12	17.12	26.02	09.03	04.03
Litovit®	-	-		-	-	-	13.12	14.12	17.12	-	09.03	04.03
Lactofol B	-	-		-	-	-	13.12	14.12	17.12	-	09.03	04.03
Development phases	Stem formation			Budding			Start of blossoming – 10%			Mass blossoming – 75%		
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015
Control	-	12.04	09.04	-	28.04	26.04	-	04.05	02.05	-	10.05	08.05
Fertiactyl Starter	06.04	9.04	06.04	22.04	25.04	23.04	28.04	01.05	29.04	04.05	07.05	05.05
Litovit®	-	11.04	08.04	-	26.04	25.04	-	02.05	01.05	-	09.05	07.05
Lactofol B	-	10.04	07.04	-	26.04	24.04	-	02.05	30.04	-	09.05	06.05
Development phases	Waxy maturity			Full maturity			Vegetation period					
Years and variants	2013	2014	2015	2013	2014	2015	2013	2014	2015			
Control	-	01.07	30.06	-	17.07	16.07	-	247	250			
Fertiactyl Starter	27.06	28.06	27.06	12.07	15.07	14.07	244	245	248			
Litovit®	-	01.07	29.06	-	17.07	16.07	-	247	250			
Lactofol B	-	29.06	28.06	-	16.07	15.07	-	246	249			

The uneven distribution of rainfall in May and June (3.4; 109.5 mm) in 2013 led to a reduction in the phases of mass blossoming, waxy and full maturity in all tested variants, and they passed through them the fastest and the earliest. Higher rainfall in May and June (66.5, 98.8 mm) in 2014 and (69.5, 76.7 mm) in 2015 in all tested variants lead to the later occurrence of these phases.

During the three years of study and at the different sowing dates, Fertiactyl Starter (3000 ml/ha) treated variants entered these developmental stages 2-3 days earlier than control, whereas Lactofol B-4000 ml/ha and Litovit® 2000 g/ha treated variants reach these phases one day earlier or simultaneously with the control. Comparing the entering into the phase of blossoming, waxy and full maturity by the Lactofol B variant (4000 ml/ha) and Litovit® variant (2000 g/ha), this occurred 1 day earlier in favour of Lactofol B (4000 ml/ha).

The vegetation period for all variants of the experiment at sowing from 1 to 10. X is the longest (from 261 to 263 days, in 2013, from 267 to 269 days, in 2015 and from 271 to 273 days in 2014) for the rest of the sowing dates, it is almost the same from 238 to 244 days in 2013; from 245 to 250 days in 2015 and from 245 to 252 days in 2014.

CONCLUSIONS

On the basis of the experimental work and the results obtained, the following conclusions can be drawn:

1. The phenological observations made during the three years of the experiment show that, at all sowing dates, foliar fertilization contributes to faster developmental phases than the control.
2. The strongest developmental stimulant effect during the three years and at different sowing dates was observed in the Fertiactyl Starter (3000 ml/ha) treated variant followed by Lactofol B (4000 ml/ha) sprayed variant, and Litovit® (2000 g/ha).
3. Phases of development in 2012-2013 at the last sowing date were only reported for the Fertiactyl Starter variant, while the remaining Lactofol B, Litovit® variant and the control plants perished.
4. The vegetation period is the longest in 2015 (248 to 250 days), and the shortest in 2013 (244 days).

Of all foliar fertilizer treated variants with the shortest vegetation period (238 to 271 days) are those treated with Fertiactyl Starter.

REFERENCES

Coelho, H., Grassi Filho, H., Barbosa, R., Romeiro, J. (2011). Pompermayer, G. V.; Lobo, T. Agronomic efficiency of leaf application of nutrients in the soybean crop. *Revista Agrarian*, *4*(11), 73–78.

Delchev, G. (2010). Stability valuation of some mixtures between foliar fertilizers and antibroadleaved herbicides for the grain yield of durum wheat. *Pochvoznanie, Agrokhimiya i Ekologiya*, *44*(2), 41–46.

Kolev, T., Todorov, Zh., Koleva, L. (2012). Influence of the complex foliar fertilizer “Crystallon Special” on the productivity of the hard wheat (*Tr. durum* Desf.). Научные исследования и разработки. Международной научно-практической конференции молодых ученых (19-20 апреля 2012г.), Иркутск, 81–85.

Kolev, T., Todorov, Zh., Koleva, L. (2013). Effect of the liquid fertilizers for leaves on the growth and productivity of durum wheat. Международной научно-практической конференции. Климат, экология, сельское хозяйство евразии (28-30 мая 2013г.), Иркутск, 154-158.

Sanmueang, A., Detpiratmongkol, S., Yoosukyingsataporn, S., Ubolkerd, T. (2011). Influence of foliar application of potassium fertilizers on growth and yield of sweet sorghum. *Proceedings of the 49th Kasetsart University Annual Conference, Kasetsart University, Thailand, 1-4 February. Subject: Plants*, *I*, 458–464.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF GROWTH REGULATORS ON THE DURUM WHEAT YIELD

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Abstract

In a field experiment carried out in the Experimental and Implementation Base of the Department of Plant Growing at the Agricultural University, in non-irrigated conditions, the influence of following growth regulators was tested: Madex Top (600 ml/ha), Flordimex T Extra (750 ml/ha), Seron 480 SL (1000 ml/ha) and Trimax 175 EK (400 g/ha) on the yield of durum wheat varieties Deyana and Zvezditsa. The tested growth regulators have influenced to varying degrees the yield gains of the investigated durum wheat varieties. The treatment of Deyana durum wheat variety with the growth regulator Seron 480 SL (1000 ml/ha) results in an increase of the yield with 450 kg/ha (11.5%) on average over the three-year study period compared to the control. The higher grain yield of the durum wheat variety Zvezditsa with the same growth regulator is 350 kg/ha (9.8%). With second results, for both tested varieties is the variant treated with Medax Top (600 ml/ha), where the average increase of grain yield is respectively: for Deyana variety 280 kg/ha (8.7%) and for Zvezditsa variety 260 kg/ha more than the untreated control. When spraying durum wheat with Trimax 175 EC, the yield gain for Deyana is 280 kg/ha, and 250 kg/ha for Zvezditsa variety. The studied growth regulators have helped to increase the grain mass in one ear for the Deyana variety, while for the Zvezditsa variety - to increase the number of grains in one ear.

***Key words**: growth regulators, durum wheat, yield productivity.*

INTRODUCTION

A key area of cereal crop technology is the use of growth regulators (Delchev et al., 2011; Kolev et al., 2006). A number of scientific publications abroad (Pirasteh Anosheh et al., 2012; Nabti et al., 2010) and in Bulgaria (Mangova et al., 2013; Kolev et al., 2015) also present research results on this issue. A number of leading companies producing agricultural products present new products every year.
The need to investigate the impact of new growth regulators on grain yield and quality of durum wheat has led to the present experiment.

MATERIALS AND METHODS

During the period 2014-2017, a field experiment was conducted in the Training, Experimental and Implementation Centre of the Department of Plant Growing at Agricultural University under non-irrigated conditions, where the influence of some growth regulators on the productivity of durum wheat varieties Deyana and Zvezditsa (selection of the Field Cultures Institute in the town of Chirpan) was tested.
Growth regulators against layering of durum wheat were tested: **Madex Top** (active substance 300 g/l mepiquat chloride + 50 g/l prohexadione calcium) at a dose of 600 ml/ha is administered in BBCH phase 30-39 (start of stem elongation - the flange leaf is completely unfolded); **Floridimex T Extra** (active substance 660 g/l ethephon) at a dose of 750 ml/ha introduced into the BHCH phase 26-30 (end of tillering - start of shooting up); **Seron 480 SL** (active substance 480 g/l ethephon) at a dose of 1000 ml/ha applied in phase BBCH 32-39 (from the phase the second node is at least 2 cm above the first to phase the flange leaf is completely unfolded); **Trimax 175 EK** (active substance 175 g/l trinexapac-ethyl) at a dose of 400 g/ha and phase of administration BBCH 30-39 (beginning of stem elongation to a fully unfolded flange leaf). There was also an untreated control. The experiment is based on the fractional plot method in four iterations

with a plot size of 10 m^2. Durum wheat is grown according to established technology (Yanev et al., 2008). The sowing is carried out in the optimal for Bulgaria period from 20th October to 10th November with a sowing rate of 500 germinating seeds per m^2. The field experiment is carried out on alluvial-meadow soil (Molic Fluvisols by FAO), which is characterized by a medium sand-clayey mechanic composition, a humus content of 1-2% and a pH of 7.7. The nutrient content in the soil layer 0-20 cm is as follows: N - 15.6 mg/1000 g; P_2O_5 - 32 mg/100 g; K_2O - 47 mg/100 g of soil; $CaCO_3$ - 7.4% (Popova et al., 2010).
The area subjected to the experiment was fertilized with 120 kg/ha NH_4NO_3 (ammonium nitrate) and 80 kg/ha P_2O_5 (triple superphosphate - 46% diphosphorus pentoxide) in the active substance. In order to determine the differences between the tested growth regulators and the specific varietal response to them, the following indicators were taken into account: plant height (cm), length of spike (cm), number of spikelets in one spike (pcs), grain yield (t/ha), number of grains in one spike (pcs), grain mass in one spike (g), mass of 1000 grains (g) and specific weight (kg/hl).
The statistical processing of the data obtained on the surveyed indicators was carried out with the BIOSTAT software (Penchev, 1998).
The aim of the experiment carried out is to study the impact of the growth regulators Madex Top, Floridimex T Extra, Seron 480 SL, and Trimax 175 EK on the productivity of the varieties durum wheat Deyana and Zvezditsa in the climatic and soil conditions of Southern Bulgaria.

RESULTS AND DISCUSSIONS

The sum of rainfall during durum wheat vegetation was as follows: 2014/2015 - 655.8 mm/m^2, 2015/2016 - 388.5 mm/m^2, and 2016/2017 - 278.3 mm/m^2 compared to 419.0 mm/m^2 for a thirty year period time (Figures 1, 2).

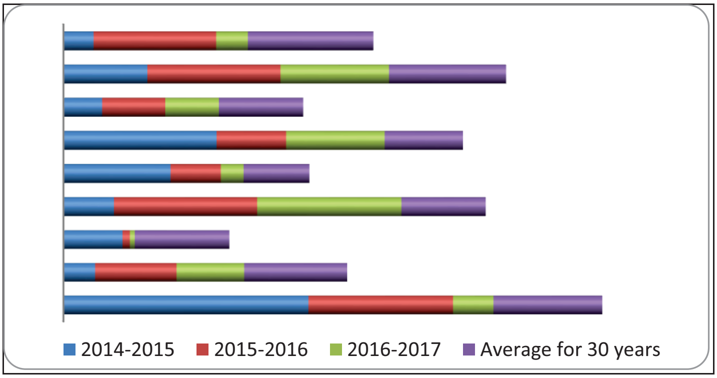
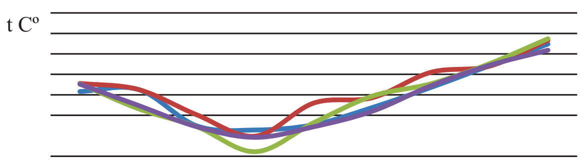


Figure 1. Precipitation by months (sum mm/m^2)



	X	XI	XII	I	II	III	IV	V	VI
2014-2015	10,8	11,3	2,4	1,4	2,7	6,9	11,8	17,1	22,4
2015-2016	12,8	11,3	5,1	-0,1	8	9,3	15,5	17	23,3
2016-2017	12,7	6,6	2,2	-3,9	3,2	9,7	12,7	17,6	23,7
Average for 30 years	12,6	7,4	2,2	-0,4	2,2	6	12,2	17,2	20,9

Figure 2. Monthly temperatures (average)

In the years of the experiment, favourable for the growth and development of durum wheat with a good distribution of rainfall is the harvest year 2017, then higher grain yields were obtained for all tested variants. Despite the large amount of precipitation during vegetation unfavourable for the plant growth is the first year, i.e. 2014/2015 due to a drought in the spring, when in the phases of the end of tillering - the beginning of shooting up, the stages of organogenesis happen, during which the length of the spike, the number of spikelets and blossoms are determined.

Grain yield is the indicator that reflects the effects of growth regulators and other factors studied (Table 1).

From the data presented in the table it is evident that both varieties react differently depending both on the studied growth regulators and on the weather conditions of the year.

In durum wheat of Deyana, the highest increase in grain yield was observed under the effect of the growth regulator Seron 480 SL (1000 ml/ha). In the experimental years, the increase was from 360.0 kg/ha to 530 kg/ha or an average of 450 kg/ha (11.5%) over the three-year study period, compared to the untreated control. In the case of durum wheat variety Zvezditsa with the same growth regulator, the increase in productivity is also high, but compared to the Deyana variety, the values obtained are lower in the range from 350 kg/ha to 380 kg/ha or on average 350 kg/ha (9.8%).

In the second place in both tested varieties is the variant treated with Medax Top (600 ml/ha), where the average increase of grain yield is respectively: for Deyana 280 kg/ha (8.7%) and for Zvezditsa variety 260 kg/ha more than untreated control. The use of the growth regulator Trimax 175 EK results in an increase in productivity for Deyana with 280 kg/ha and 250 kg/ha for the Zvezditsa variety.

The growth regulator Floridimex T Extra (750 ml/ha) affected to the least degree the grain yield increase for both tested varieties, as in both experimental varieties of durum wheat 100 kg/ha of grain more were harvested versus the untreated control.

The productivity of the studied durum wheat varieties results from the effect of the tested growth regulators on some of the structural elements of the yield (Table 2). Plant height decrease was observed in all studied controls compared to untreated control. In the variant treated with the growth regulator Seron 480 SL (1000 ml/ha), the plant height measured for the three-year period of Deyana variety is 4 cm, and of the Zvezditsa variety is 5 cm smaller than that of the control.

Table 1. Grain yield t/ha

Variety	Growth regulators	2015	2016	2017	Average	
		t/ha	t/ha	t/ha	t/ha	%
Deyana	Control untreated	3.70	3.94	4.06	3.90	100,0
	Seron 480 SL	4.17	4.30	4.59	4.35	111,5
	Medaks Top	4.05	4.23	4.45	4.24	108,7
	Flordimeks T Extra	3.86	3.97	4.18	4.00	102,6
	Trimax 175 EK	4.04	4.15	4.35	4.18	107.2
Zvezditsa	Control untreated	3.29	3.57	3.83	3.56	100,0
	Seron 480 SL	3.65	3.92	4.21	3.91	109.8
	Medaks Top	3.51	3.80	4.14	3.82	107,3
	Flordimeks T Extra	3.34	3.68	3.95	3.66	102,8
	Trimax 175 EK	3.45	3.78	4.20	3.81	107,0

	A	B	AxB	A	B	AxB	A	B	AxB
GD 5%	20.2	21.8	24.5	16.3	17.5	19.7	10.1	15.3	18.4

In the spike length indicator, more significant changes were observed when the plants were sprayed with Seron 480 SL (1000 ml/ha), where the decrease in the length of the spike of Deyana variety was by 0.8 cm and for the Zvezditsa variety by 0.5 cm.

The tested growth regulators have helped to increase the grain mass in one spike of Deyana variety, and in the Zvezditsa variety - to increase the number of grains in one spike.

In Deyana variety of durum wheat yield is higher because of the higher grain mass in one spike, by 0.38 g more than the untreated control in the Seron 480 SL (1000 ml/ha) variant, whereas in the Zvezditsa variety there is an increase in the number of grains in one spike with 3.9 pcs. in plants treated with the same growth regulator.

This trend is preserved in the other studied growth regulators. Lower values for the number of grains in a spike, i.e. 34.4 pcs. for Deyana variety, and grain mass of 1.07 g in a spike for Zvezditsa variety were obtained by treatment with Floridimex T Extra (750 ml/ha), but the results obtained were higher than the untreated control, respectively, by 0.9 and 0.03 g.

Table 2 Biometrical data (2015-2017)

Variety	Growth regulators	Height of plants, cm	Length of the spike, cm	Number of the grains per spike	Mass of the grains per spike, g
Deyana	Control untreated	90	7.6	33.5	1.23
	Seron 480 SL	86	6.8	39.7	1.61
	Medaks Top	89	7.2	37.5	1.53
	Flordimeks T Extra	87	7.0	34.4	1.31
	Trimax 175 EK	88	7.1	36.1	1.40
Zvezditsa	Control untreated	94	7.4	38.2	1.04
	Seron 480 SL	89	6.9	42.1	1.26
	Medaks Top	92	7.2	40.5	1.21
	Flordimeks T Extra	93	7.0	39.4	1.07
	Trimax 175 EK	90	7.1	39.8	1.18

CONCLUSIONS

The tested growth regulators have influenced to varying degrees the increase in productivity in the investigated durum wheat varieties.

The treatment of Deyana durum wheat with the growth regulator Seron 480 SL (1000 ml/ha) resulted in an increase in the yield by 450 kg/ha (11.5%) on average over the three-year study period compared to the control.

Higher grain yields of durum wheat variety Zvezditsa treated with the same growth regulator is 350 kg/ha (9.8%).

In the second place, in both tested varieties is the variant treated with Medax Top (600 ml/ha), where the average increase of grain yield is respectively: for Deyana - 280 kg/ha (8.7%), and for Zvezditsa variety 260 kg/ha more than untreated control. When spraying durum wheat with Trimax 175 EK, the yield increase for Deyana is 280 kg/ha, and 250 kg/ha for Zvezditsa variety.

The studied growth regulators have helped to increase the grain mass in one spike in the Deyana variety, while in the Zvezdica variety to increase the number of grains in one spike.

The studied growth regulators improve the physical properties of the grain, mass per 1000 grains, hectolitre mass, and glassiness. For durum wheat varieties, Deyana and Zvezditsa, the highest increase in the values of the physical properties of the grain was observed in the treatment with Seron 480 SL and Medaks Top.

REFERENCES

Delchev, G., Kolev, T., Nenkova, D. (2011). Impact of some mixtures between stimulators and antigrass herbicides on the sowing properties of the durum wheat sowing-seeds. *JCEA, 12*(3), 398–408.

Mangova, M., Menkovska, M., Petrova, I., Kolev, T. (2013). Variety - The Main Factor for Impoving the Quality of Durum wheat (*Triticum durum* Desf.). *Macedonian Journal of Animal Science, 3*(2), 213–217.

Kolev, T., Delchev, G., Ivanova, R., Delibaltova, V. (2006). Effect of several plant growth regulators on the productivity of Durum wheat (*Tr. durum* Desf.). IV Medunarodna ekokonferencija 20-23. IX. Zdravstveno bezbedna hrana. *Safe food. I. Novi Sad*, 363–367.

Kolev, T., Petrova, I, Todorov, Zh., Koleva-Valkova, L. (2015). The influence of natural organic products on the productivity of Durum wheat. Материалы IV международной научно-практической конференции “Климат, экология, сельское хозяйство Евразии”, Иркутск, Част I, 29–33.

Nabti, E., Sahnoune, M., Ghoul, M., Fischer, D.,

Hofmann, A., Rothballer, M., Schmid, M., Hartma, A. (2010). Restoration of Growth of Durum Wheat (*Triticum durum* var. waha) Under Saline Conditions

Due to Inoculation with the Rhizosphere Bacterium Azospirillum brasilense NH and Extracts of the Marine Alga Ulva lactuca. *J. Plant Growth Regul*., *29*, 6–22.

Penchev, E. (1998). Evaluation of productivity and quality indicators of wheat with mathematical models. Dissertation. pp,165.

Pirasteh Anosheh, H., Emam, Y., 2012. Manipulation of Morpho-Physiological Traits in Bread and Durum Wheat by Using Growth Regulators at Different Irrigation Regimes. *Journal of Crop Production and Processing, 2*(5), 29–46.

Popova, R., and Sevov, A. (2010). Soil characteristic of experimental field of Crop Production Department as a result of the cultivation of grain, technical and forage crops. Agricultural University Plovdiv. *Scientific Works, LV*(I), 151–156.

Yanev, Sh., Dechev, D., Lalev, Ts., Saldzhiev, I., Panayotova, G., Delchev, G.,…., Rashev, S. (2008). *Technology for grow*ing *Durum wheat*. Publishing Temko, Stara Zagora.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

GRAIN PROTEIN OF SORGHUM DEPENDING ON NITROGEN RATES

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Abstract

*The response of grain protein concentration and yield of sorghum to nitrogen fertilization in rates 0, 60, 120, 180, 240 and 300 kg N.ha^{-1} was studied in the experimental field of Agricultural University of Plovdiv, Bulgaria in 2017-2018 under non-irrigated conditions. Without nitrogen fertilization sorghum hybrid EC Alize formed grain with 10.98%-11.10% protein. Rate N_{300} significantly increased concentration of grain protein by 15.0% and 21.9%, respectively in 2017 and 2018, compared to N_0 plants. Fertilization N_{60} - N_{300} proven increased grain protein yield over the N_0. Rate N_{180} provided higher grain protein yield of 708 kg.ha^{-1} in 2017 and higher N_{240} and N_{300} rates showed a downward trend in protein yield within limits 677-708 kg.ha^{-1}. In more favourable in terms of rainfall 2018, the highest grain protein yield 907 kg.ha^{-1} was obtained at N_{240}. Application of N_{300} proven reduced by 80 kg.ha^{-1} the protein yield, compared to N_{240}. Rates 0-300 kg N.ha^{-1} highly positively correlated with grain protein concentration (0.864**-0.962**) and protein yield (0.839**-0.874**) of sorghum.*

Key words: *protein, concentration, yield, grain.*

INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) is the fifth leading cereal grain produced worldwide after wheat, corn, rice, and barley (FAO, 2013). In Bulgaria the production of grain sorghum has increased in recent years and sorghum is one of the top ten grown crops in the country. The modern varieties are hybrids with high productivity potential appropriate for application of intensive forage grain production technologies (Kertikov, 2007; Kikindonov et al., 2008). This crop has better ability to tolerate drought stress compared with other crops and is known as an index for drought resistance of agronomic crops (Kebede, 2001; Wenzel, 2001). Sorghum is mainly grown under non-irrigated fields where stressful conditions during grain filling can limit productivity and increase the dependence of the yield of spare assimilations (Kaye et al., 2007). It is a multipurpose crop belonging to the Poaceae family, which are C_4 carbon cycle plants with high photosynthetic efficiency and productivity (Tari et al., 2012).

Soil fertility management is important to facilitate the rapid development of sorghum plants, but many agricultural soils have a limited ability to supply available nitrogen for target yields and nitrogen is the most limiting nutrient for cereal crops including for sorghum production (Gerik et al., 2014). Nitrogen is the main nutrient for C_4 plant productivity (Hao et al., 2014). It plays a critical role in cell division during the plant growth (Stals & Inzé, 2001) and the deficit of soil nitrogen leads to lower sorghum biomass due to reductions in leaf area, chlorophyll index, and photosynthetic rate (Zhao et al., 2005; Hirel et al., 2007; Mahama et al., 2014). Nitrogen fertilizer is known to boost the aboveground biomass yield (Amaducci et al., 2004; Anderson et al., 2013). Nitrogen fertilization had a significant impact on the concentration of protein in the grain and grain protein yield (Anfinrud et al., 2013). Application of nitrogen in rates 120-240 kg N.ha^{-1} increase crude protein content to higher levels to support rapid weight gains and milk yields (Hoffman et al., 2001; Kaufman et al., 2013). The nitrogen doses 50-200 kg. ha^{-1} contributed to an increase in the crude protein together with an increase in dry matter and/or protein concentration and crude protein increased 59.5-312.9% (Melo et al., 2017). The phosphorus and potassium fertilization slightly affected grain protein yield of sorghum grown under good phosphorus and potassium soil availability (Franco et al., 2017).

Improper nitrogen fertilization and excess nitrogen resulted in environmental impacts, such as the pollution by nitrate (NO_3-N) leaching and nitrous oxide emissions (Miller & Cramer, 2004; Ramu et al., 2012), as well as increasing production costs (Marsalis et al., 2010). Thus, coordination of sorghum N demand with N supply is critically important to maximize economic efficiency, optimize biomass quality, and minimize loss of soil NO_3-N and environmental pollution (Schroder et al., 2000; Zhu et al., 2000; Rooney et al., 2007; Cui et al., 2008; Meki et al., 2017).

The objective of this study was to determine the effect of nitrogen fertilization rates on the concentration of protein in the grain and grain protein yield of sorghum grown under non-irrigated conditions.

MATERIALS AND METHODS

The investigation was carried out during the period 2017-2018 on the experimental field of Agricultural University of Plovdiv, Bulgaria, under non-irrigated conditions after wheat as predecessor. The effect of nitrogen fertilization in rates 0, 60, 120, 180, 240 and 300 kg $N.ha^{-1}$ on the concentration of protein in the grain and grain protein yield was studied at grain sorghum hybrid EC Alize. The experimental design consisted of a randomized, complete block design with four replications. The size of individual trial plots was 20 m^2. Total nitrogen as NH_4NO_3 was applied as pre-sowing fertilization on the background $P_{50}K_{50}$ fertilization as triple superphosphate and potassium chloride, respectively. Standard farming practices for the region of Southern Bulgaria were applied.

Table 1. Content of available nitrogen, phosphorus and potassium in the soil

Year	Soil depth, cm	N min, $mg.kg^{-1}$	P_2O_5, mg.100 g^{-1}	K_2O, mg.100 g^{-1}
2017	0-30	27.6	15.8	21,0
	30-60	22.1	13.9	24.0
2018	0-30	33.8	17.3	23.1
	30-60	20.4	14.1	22.9

The soil type of the experimental field is alluvial-meadow *Mollic Fluvisols* (FAO, 2006) with slightly alkaline reaction pH_{H2O} = 7.80. The content of available nutrients in the soil before sowing of the sorghum was determined in soil layers 0-30 and 30-60 cm and pointed out in Table 1. The soil had low content of mineral nitrogen and it was good supplied with available phosphorus (Egner-Ream method) and exchangeable potassium (extracted by 2N HCL).

Meteorological conditions during vegetation period of sorghum were recorded daily in the experimental area and are given in Table 2, together with the long-term average of temperature and precipitations.

Table 2. Hydro-thermal conditions during sorghum vegetation period

Year	April	May	June	July	August
	Temperature (^{o}C)				
2017	12.7	17.6	23.7	25.1	25.4
2018	16.4	19.2	28.8	30.5	24.2
Long-term norm	12.2	17.2	20.9	23.2	22.7
	Precipitation ($l.m^{-1}$)				
2017	26.1	52.7	15.4	29.8	9.2
2018	25	112.3	118.9	94.7	35.1
Long-term norm	45	65	63	49	31

The values of temperature and precipitations during the vegetation period of sorghum characterized hydro-thermal conditions of 2017 as warm and dry. In contrast, the months of May, June and July of 2018 were characterized as extremely humid. The amount of precipitation exceeded nearly twice the values of long-term norm for the region.

The total nitrogen concentration of sorghum grain in maturity was analyzed by Kjeldahl method after wet digestion by H_2SO_4 and H_2O_2 as a catalyst (Tomov et al., 2009). The grain protein concentration was calculated from the percentage of nitrogen in the grain multiplied by a coefficient of 5.6 (Mariotti et al., 2008). Grain protein yield in $kg.ha^{-1}$ was calculated according to the formula:

(Grain yield in $kg.ha^{-1}$ multiplied by the percentage of protein in the grain)/100.

An overall analysis of variance (ANOVA) was performed to evaluate the effect of the experimental treatments on the referred variables. In order to establish the difference among the means Duncan's multiple range test at level of significance $p \leq 0.05$ was used. The regression analysis was done for assessment of the effect of nitrogen fertilization rates on the grain yield, grain protein concentration and grain protein yield of sorghum. Correlation test with significance level reported ($p < 0.05$ or $p< 0.01$) was based on Pearson's correlation coefficient.

RESULTS AND DISCUSSIONS

During the comparatively dry 2017 experimental year, the average grain yield was 5234 $kg.ha^{-1}$, and under the meteorologically favourable year of 2018, average grain yield was 6803 $kg.ha^{-1}$ (Table 3). Regarding the grain yield of sorghum, the strong proven effect of the nitrogen fertilization of dose N_{180} was established in 2017. The yield increase was by 25.8% above the control plants. In 2017, the application of N_{120}, N_{240} and N_{300} resulted in similar grain yields 5355-5455 $kg.ha^{-1}$. Higher grain yields of sorghum in 2018 were found in rates N_{240} and N_{180}, which exceeded the N_0 by 38.4 and 35.7%, respectively.

Table 3. Grain yield of sorghum depending on nitrogen fertilization rates, $kg.ha^{-1}$

Rates	2017 year	% to N_0	2018 year	% to N_0
N_0	4572 ^{d*}	100	5023 ^{d}	100
N_{60}	4897 ^{c}	107.2	5441 ^{c}	108.4
N_{120}	5378 ^{b}	117.7	6090 ^{b}	121.3
N_{180}	5750 ^{a}	125.8	6812 ^{a}	135.7
N_{240}	5455 ^{b}	119.3	6951 ^{a}	138.4
N_{300}	5355 ^{b}	117.3	6180 ^{b}	123.1
Average	*5234*		*6803*	

*Values with identical letters within each column are not significantly different at p<0.05 according to Duncan's multiple range test.

According to various authors, the total nitrogen content in the sorghum grain usually changed from 1% N to 3% N (Singh & Axtell, 1973). The concentration of grain protein ranged from 6% to 18% and the average content was 11% (Lasztity, 1996; de Mesa-Stonestreet et al., 2010). Our results showed the similar values of grain protein in the range of 10.98-13.38% (Table 4). In experimental 2017, the grain protein concentration changed from 11.10% (N_0 control) to 12.77% (N_{300}). The concentration of grain protein in experimental 2018 varied from 10.98% (N_0 control) to 13.38% (N_{300}). The higher average grain protein concentration of all studied nitrogen rates was established in 2018, which characterized as more favourable on precipitation compared to the dry conditions during sorghum vegetation of harvested 2017. In each of the two experimental years, the protein concentration of the grain enhanced with the increase of applied mineral nitrogen. No proven differences of grain protein concentration were found between the control plants and the plants cultivated at low N_{60} rate. A tendency was indicated an increase of the protein concentration of the sorghum grain between the variant of high N_{300} fertilization and N_{180} and N_{240} rates. Application of 300 kg $N.ha^{-1}$ showed a significant higher protein concentration of grain compared to the grain protein percentage of plants cultivated at N_{60} and N_{120} rates. This was observed during both experimental years. Sorghum formed a grain which contained an average of 11.04 % protein when the plants grown without nitrogen fertilization. As the nitrogen fertilization increased to the N_{300} rate, the concentration of grain protein significantly increased up to 12.77% (in 2017) and 13.38% (in 2018). These values were higher by 15.0% and by 21.9%, respectively, compared to N_0 control plants of the two experimental years. The present results confirm the main effect of nitrogen fertilization on the protein concentration in cereals grain and they correspond to the results for grain sorghum of the other researchers (Assefa et al., 2010; Ciampitti et al., 2016; Van Oosterom et al., 2010).

Table 4. Protein concentration of grain of sorghum depending on nitrogen fertilization, %

Rates	2017 year	% to N_0	2018 year	% to N_0
N_0	11.10 ^{c*}	100	10.98 ^{c}	100
N_{60}	11.34 ^{c}	102.2	11.82 ^{bc}	107.7
N_{120}	12.11 ^{b}	109.1	12.38 ^{b}	112.8
N_{180}	12.32 ^{ab}	111.0	12.82 ^{ab}	116.8
N_{240}	12.42 ^{ab}	111.9	13.05 ^{a}	118.9
N_{300}	12.77 ^{a}	115.0	13.38 ^{a}	121.9
Average	*12.01*		*12.40*	

*Values with identical letters within each column are not significantly different at p<0.05 according to Duncan's multiple range test.

The results of grain protein yield of sorghum presented in Table 5 indicated that nitrogen application in N_{60} - N_{300} rates significantly increased grain protein yield over the unfertilized control. This was observed in both experimantal years. The hydro-thermal conditions during sorghum vegetation affected the grain and grain protein yields. The sorghum vegetation period of experimental 2018 was characterized with more rainfall. As a result of that, the average grain protein yield of all studied treatments was 759 kg.ha^{-1} or by 127.8 kg.ha^{-1} higher compared to the obtained average protein yield in harvested 2017. Sorghum had the lowest yields of grain protein when cultivated without nitrogen fertilization. In 2017, the obtained grain protein yield from N_0 variant was 508 kg.ha^{-1}.

The protein yield of the control plants in harvested 2018 was 551 kg.ha^{-1}. Nitrogen fertilization in 60, 120, 180, 240 and 300 kg N.ha^{-1} proven increased yield of grain protein of sorghum in comparison of unfertilized control in both experimental years. The grain protein yields of nitrogen received plants exceeded the yield of N_0 grown plants by 9.3%-39.4% and by 16.7% to 64.6%, respectively, in the harvested 2017 and in 2018 years.

In 2017, grain protein yield increased in a parallel with the levels of applied mineral nitrogen up to the N_{180}. The fertilization rate of 180 kg of N.ha^{-1} provided a high yield of grain protein and the increase was by 39.4% relative to the protein yield of unfertilized control. The results showed a tendency for lower grain protein productivity of sorghum within limits 677-708 kg.ha^{-1} at application of higher N_{240} и N_{300} rates. In 2017, the effect of the higher mineral nitrogen 240 kg N.ha^{-1} and 300 kg N.ha^{-1} on the grain protein yield was mathematically unproven with regard to the obtained grain protein yields of sorghum fertilized with N_{180} and N_{120} rates.

Table 5. Grain protein yield of sorghum depending on nitrogen fertilization, kg.ha^{-1}

Rates	2017 year	% to N_0	2018 year	% to N_0
N_0	508 ^{d*}	100	551 ^{e}	100
N_{60}	555 ^{c}	109.3	643 ^{d}	116.7
N_{120}	651 ^{b}	128.2	754 ^{c}	136.8
N_{180}	708 ^{a}	139.4	873 ^{ab}	158.5
N_{240}	677 ^{ab}	133.3	907 ^{a}	164.6
N_{300}	684 ^{ab}	134.6	827 ^{b}	150.1
Average	*631.2*		*759*	

*Values with identical letters within each column are not significantly different at p<0.05 according to Duncan's multiple range test.

In 2018 harvested yiear, which was more favorable in terms of rainfall, the positive effect of applied nitrogen on the protein yield was observed up to the rate N_{240}. The highest grain protein yield (907 kg grain protein.ha^{-1}) was obtained with 240 kg N.ha^{-1} application, but the difference with the yield at N_{180} fertilization (873 kg grain protein.ha^{-1}) was not proven. Application of the higher N_{300} rate significantly reduced the grain protein yield of sorghum plants by 80 kg.ha^{-1} in comparison with the obtained yield in variant N_{240}.

Table 6. Correlation coefficients of nitrogen fertilization rates with grain yield, grain protein concentration and grain protein yield of sorghum

Year	Grain yield	Grain protein concentration	Grain protein yield
2017	0.724*	0.864**	0.839**
2018	0.770*	0.962**	0.874**

*Correlation is significant at the 0.05 level
**Correlation is significant at the 0.01 level

Positive correlations between examined parameters of sorghum were recorded in the study (Table 6). The results indicated that nitrogen fertilization in rates 0-300 kg N.ha^{-1} was highly positively correlated with the grain yield, grain protein concentration (r = 0.864**) and protein yield (r = 0.839**) in experimental 2017. Also, a positive significant correlation was achieved between the nitrogen fertilization rates and yields of grain and protein, and protein concentration of grain (r = 0.962**) in harvested 2018.

Table 7. Regression models of sorghum grain yield, grain protein concentration and grain protein yield depending on nitrogen fertilization rates.

Parameter	Equation	R^2
2017		
Grain yield	$y = -0.026x^2 + 10.6x + 4497$	0.923
Protein concentration	$y = -1E\text{-}05x^2 + 0.01x + 11.0$	0.959
Grain protein yield	$y = -0.004x^2 + 1.68x + 494$	0.940
2018		
Grain yield	$y = -0.04x^2 + 17.1x + 4816$	0.884
Protein concentration	$y = -2E\text{-}05x^2 + 0.014x + 11.0$	0.995
Grain protein yield	$y = -0.006x^2 + 2.83x + 525$	0.947

Regression analysis for the dependencies between the resulting parameters (grain yield, grain protein concentration, and grain protein yield) and the factor nitrogen fertilization on sorghum hybrid EC Alize indicated that correlations were represented by equations of the second degree (Table 7). High values of coefficients of determination ($R^2 > 0.850$) were found for the studied parameters of sorghum productivity in dependence of nitrogen fertilization. The regression model indicated that the grain yield increased with the raise of applied nitrogen rate up to N_{180} and the grain protein concentration increased up to N_{300} fertilization of in both experimental years.

CONCLUSIONS

Without nitrogen fertilization sorghum hybrid EC Alize formed grain with 10.98%-11.10% protein. Applied mineral nitrogen enhanced the protein concentration of sorghum grain, but effect of N_{60} was not proven compared to the N_0 control. Rate 300 kg $N.ha^{-1}$ significantly increased concentration of grain protein by 15.0% and 21.9%, respectively in 2017 and 2018, compared to N_0 plants.

Fertilization N_{60} - N_{300} significantly increased grain protein yield over the unfertilized control. The N_{180} rate provided higher grain protein yield of 708 $kg.ha^{-1}$ in 2017. The increase was by 39.4% relative to the protein yield of N_0 control. Application of higher N_{240} and N_{300} rates showed a downward trend in protein yield within limits 677-708 $kg.ha^{-1}$. In more favourable in terms of rainfall 2018 harvested yiear, the highest grain protein yield 907 $kg.ha^{-1}$ was obtained at fertilization 240 kg $N.ha^{-1}$. Application of N_{300} significantly reduced by 80 $kg.ha^{-1}$ the grain protein yield of sorghum, compared to N_{240}. The fertilization in rates 0–300 kg $N.ha^{-1}$ highly positively correlated with grain protein concentration (0.864**-0.962**) and protein yield (0.839**-0.874**).

REFERENCES

Amaducci, S., Monti, A., Venturi, G., (2004). Non-structural carbohydrates and fibre components in sweet and fibre sorghum as affected by low and normal input techniques. Ind. *Crop Prod*., *20*. 111–118.

Anderson, E.K., Parrish, A.S., Voigt, T.B., Owens, V.N., Hong, C., Lee, D.K., (2013). Nitrogen fertility and harvest management of switch grass for sustainable bioenergy feedstock production in Illinois. *Ind. Crop Prod*., *48*. 19–27.

Anfinrud, R., Cihacek, L., Johnson, B.L., Ji, Y., Berti, M.T., (2013). Sorghum and kenaf biomass yield and quality response to nitrogen fertilization in the Northern Great Plains of the USA. *Ind. Crop Prod*., *50*, 159–165.

Assefa, Y., and Staggenborg, S.A. (2010). Grain sorghum yield with hybrid advancement and changes in agronomic practices from 1957 through 2008. *Agronomy Journal*, *102*, 703–706.

Ciampitti, I.A., and Prasad, P.V. (2016). Historical Synthesis-Analysis of Changes in Grain Nitrogen Dynamics in Sorghum. *Front. Plant Sci*., *7*, 275.

Cui, Z., Zhang, F., Chen, X., Miao, Y., Li, J., Shi, L., Xu, J., Ye, Y., Liu, C., Yang, Z., Zhang, Q., Huang, S., Bao, D., (2008). On-farm evaluation of an in-season nitrogen management strategy based on soil Nmin test. *Field Crop Res*., *105*, 48–55.

de Mesa-Stonestreet, N.J., Alavi, A.S., Bean, S.R. (2010). Sorghum Proteins: The Concentration, Isolation, Modification, and Food Applications of Kafirins. *Journal of Food Science*, *75*(5), 90–104.

FAO, 2013. FAOSTAT online database: http://faostat3.fao.org/download/Q/QC/E.

Franco, A. (2011). Marcha de absorção e acúmulo de nutrientes na cultura do sorgo. Dissertação (Mestrado em Produção Vegetal), *Universidade Estadual de Montes Claros*, Beasil, 13–19.

Gerik, T., Bean, B. & Vanderlip, R. (2014). Sorghum Growth and Development, Productivity Rotations on farms in Texas, *7*(3), 2–7.

Hao, B., Xue, Q., Bean, B.W., Rooney, W.L., Becker, J.D., (2014). Biomass production, water and nitrogen use efficiency in photoperiod-sensitive sorghum in the Texas High Plains. *Biomass Bioenergy, 62*, 108–116.

Hirel, B., Le Gouis, J., Ney, B., Gallais, A. (2007). The challenge of improving nitrogen use efficiency in crop plants: towards a more central role for genetic variability and quantitative genetics within integrated approaches. *J. Exp. Bot*., *58*, 2369–2387.

Hoffman, P.C., Esser, N.M., Bauman, L.M., Denzine, S.L., Engstrom, M., Chester-Jones, H., (2001). Short communication: effect of dietary protein on growth and nitrogen balance of Holstein Heifers1. *J. Dairy Sci*., *84*, 843–847.

Kaye, N.M., Mason, S.C., Jackson, D.S., Galusha, T.D. (2007). Crop Rotation and Soil Amendment Alters Sorghum Grain Quality. *Crop Science, 47*, 722–729.

Kaufman, R.C., Wilson. J.D., Bean, S.R., Presley, D.R., Blanco-Canqui, H., Mikha, M. (2013). Effect of Nitrogen Fertilization and Cover Cropping Systems on Sorghum Grain Characteristics, *Journal of Agricultural and Food Chemistry*, *61*, 5715–5719.

Kebede, H., Subudhi, P.K., Rosenow, D.T., Nguyen, H.T, (2001). Quantitative trait loci influencing drought tolerance in grain sorghum (*Sorghum bicolor* L. Moench). *Springer-Verla. Theor Appl Genet, 103*. 266–276.

Kertikov, T. (2007). Study of productivity capacities for production of forage, crude protein and chemical composition in sorghum hybrids. *Bulgarian Journal of Agricultural Science, 13*(3), 281–289.

Kikindonov, Ts., Slanev, K., Kikindonov, G. (2008). Green mass productivity of sorghum origins. *Journal* *of Mountain Agriculture on the Balkans*, *11*(3), 503–511.

Lasztity, R. (1996). Sorghum proteins. In: Lasztity R, editor. *The chemistry of cereal proteins*. 2nd ed. Boca Raton, Fla.: *CRC Press*, 227–248.

Mahama, G.Y., Prasad, P.V.V., Mengel, D.B., Tesso, T.T., (2014). Influence of nitrogen fertilizer on growth and yield of grain sorghum hybrids and inbred lines. *Agron. J*., *106*, 1623–1630.

Mariotti, F., Tomé, D., Mirand, P. (2008). Converting Nitrogen into Protein - Beyond 6.25 and Jones' Factors. *Critical Reviews in Food Science and Nutrition*, *48*(2), 177–184.

Marsalis, M.A., Angadi, S.V., Contreras-Govea, F.E., (2010). Dry matter yield and nutritive value of corn, forage sorghum, and BMR forage sorghum at different plant populations and nitrogen rates. *Field Crop Res*., *116*, 52–57.

Meki, M.N., Ogoshi, R.M., Kiniry, J.R., Crow, S.E., Youkhana, A.H., Nakahata, M.H., Littlejohn, K., (2017). Performance evaluation of biomass sorghum in Hawaii and Texas. *Ind. Crop Prod., 103*. 257–266.

Melo, D., Orlando Da Ros, C., Ferreira da Silva, R., José Coldebella, I. (2017). Fffect of N, P or K doses on the dry matter and crude protein yield in maize and sorghum, *Pesquisa Agropecuária Tropical*, *47*(1), 21–43.

Miller, A.J., Cramer, M.D., (2004). Root nitrogen acquisition and assimilation. *Plant Soil, 274*. 1–36.

Ramu, K., Watanabe, T., Uchino, H., Sahrawat, K.L., Wani, S.P., Ito, O., (2012). Fertilizer induced nitrous oxide emissions from Vertisols and Alfisols during sweet sorghum cultivation in the Indian semi-arid tropics. *Sci. Total Environ*., *438*, 9–14.

Rooney, W.L., Blumenthal, J., Bean, B., Mullet, J.E., (2007). Designing sorghum as a dedicated bioenergy feedstock. *Biofuel Bioprod. Biorefin*., *2*, 147–157.

Schroder, J., Neeteson, J., Oenema, O., Struik, P., (2000). Does the crop or the soil indicate how to save nitrogen in maize production? Reviewing the state of the art. *Field Crop Res*., *68*, 151–164.

Stals, H., Inzé, D., (2001). When plant cells decide to divide. *Trends Plant Sci*., *6*, 359–364.

Tari, I., Laskay, G., Takács, Z., Poor, P. (2012). Responses of Sorghum to Abiotic Stresses: A Review. *Journal of Agronomy Crop Science*, *199*(4), 264–274.

Tomov, T., Rachovski, G., Kostadinova, S., Manolov, I. (2009). Handbook of Agrochemistry. *Academic publisher of Agricultural University of Plovdiv*, Bulgaria.

Van Oosterom, E.J., Chapman, S.C., Borrell, A.K., Broad, I.J., and Hammer, G.L. (2010). Functional dynamics of the nitrogen balance of sorghum. II. Grain filling period. *Field Crops Res*., *115*, 29–38.

Wenzel, W.G., and Van Rooyen, P.J., (2001). Moisture Stress and Potential Sorghum Yield. *ISMN*, *42*, 28–29.

Zhao, D., Reddy, K.R., Kakani, V.G., Reddy, V.R., (2005). Nitrogen deficiency effects on plant growth, leaf photosynthesis, and hyperspectral reflectance properties of sorghum. *Eur. J. Agron., 22*. 391–403.

Zhu, Z.L., Chen, D.L., (2002). Nitrogen fertilizer use in China contributions to food production, impacts on the environment and best management strategies. *Nutr. Cycl. Agroecosyst*., *63*, 117–127.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

IMPACT OF PRECEDING CROP AND VARIETY ON GRAIN YIELD AND QUALITY PARAMETERS IN WINTER WHEAT CULTIVATED IN DÂMBOVIȚA COUNTY

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Abstract

The experimental research developed during 2017-2018 in Morteni Commune, Dâmbovița County using Exotic and Solehio wheat varieties was based on the investigation of the variability of (i) yield components (yield, number of spikes/m^2, hectolitre mass HLM, thousand kernel weight TKW) and of (ii) quality parameters (wet gluten, crude protein content) influenced by preceding crops and by wheat variety in correlation with different agrophysiological crop parameters (plant height, spike length). The experimental scheme was composed from 4 plots, as it follows: plots 1 and 3 with Exotic wheat variety and plots 2 and 4 with Solehio wheat variety. The crop rotation sequence for plots 1 and 2 was maize (2016-2017), sunflower (2015-2016), wheat (2014-2015) and for plots 3 and 4 was sunflower (2016-2017), maize (2015-2016), wheat (2014-2015). The results of the research indicated that yield, spike length, number of spikes/m^2, HLM and TKW parameters were higher after sunflower as preceding crop, meanwhile the plant height, gluten and protein contents were higher after maize as preceding crop for both wheat varieties.

Key words*: crop rotations, gluten, protein, wheat, yield components.*

INTRODUCTION

Experimental trials conducted on wheat culture demonstrated that balanced fertilization ensure high yields, nitrogen being considered as the most influential factor for good quality of grains, protein content and bread-making quality. Reviewing the literature data, it was found many papers dealing with importance of fertilization on yield components and quality parameters (Basso et al., 2010; Bunta et al., 2011; Hlisnikovsky et al., 2016; Panayotova et al., 2017; Starodub et al., 2017; Madjar et al., 2018). Some studies sustain use of liquid fertilizers to obtain high yields (Watson et al., 1992; Madjar et al., 2018). Accordingly, a research paper (Madjar et al., 2018) evidenced that splitting the nitrogen dose applied as liquid fertilizer on wheat, led to higher yields and influenced positively some yield components and quality parameters.

But to reduce the use of nitrogen and therefore to minimize its environmental effects, some practices are adopted, one of them being crop rotation. Beside yield increase, crop rotation is an important strategy to manage properly insects, weeds, to improve soil properties (Roth, 1996).

Wheat produce higher yields when is grown after unrelated species, the importance of rotation being recognized since ancient times.

The inclusion of oilseed crops in crop rotation has positive agronomic impact, this being evidenced in a study (Litke et al., 2017) which revealed in the case of wheat, the increase of yield and TKW parameter when preceding crop was oilseed rape.

Some authors (Lopez-Bellido Garrido et al., 2001) developed a field study to investigate the effects of crop rotation (wheat-sunflower, wheat-chickpea, wheat-faba bean, wheat-fallow and continuous wheat) and N fertilizer (rates 50, 100, 150 kg N·ha^{-1}) on wheat yield and it evidenced that chickpea rotation on yield was significantly smaller than of faba bean rotation and closer to sunflower rotation.

For wheat as main crop, other authors (Dogan et al., 2008) found that the most suitable crop rotation systems for Southern Marmara region, Turkey are sunflower-rapeseed-wheat, rapeseed-fodder pea + sunflower-wheat and rapeseed-common vetch + sunflower-wheat.

The investigation of influence of crop rotation (winter wheat-monocrop, winter wheat-maize, pea-winter wheat-maize, pea-winter wheat-maize-maize) and fertilization (N_0P_0, $N_{120}P_{80}$,

$N_{120}P_{80}$ + manure 10 t·ha^{-1}, applied for every crop) on biomass and yield evidenced the smallest values of subjected parameters in winter wheat monocrop case (Ardelean & Bandici, 2013).

Having in view above mentioned information, the paper presents the results obtained on the basis of an experimental research developed during 2017-2018 in Morteni Commune, Dambovita County.

The aim of the research was to investigate in the case of Exotic and Solehio wheat varieties, the variability of **(i)yield components** (yield, number of spikes/m^2, hectolitre mass HLM, thousand kernel weight TKW) and of **(ii)quality parameters** (wet gluten, crude protein content) influenced by preceding crops and by wheat variety in correlation with different agrophysiological crop parameters (plant height, spike length).

MATERIALS AND METHODS

Experimental site

Experimental research was carried out in Morteni Commune, Dâmbovița County (Figure 1).

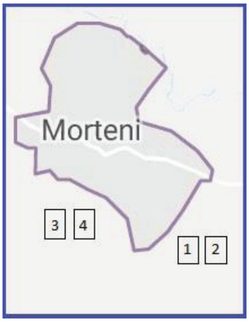

Figure 1. The position of experimental plots on Morteni Commune map

Wheat varieties

For the experiment were chosen Exotic and Solehio wheat varieties from KWS.

Fertilizers

In the experiment were used NPK 18: 46: 0, calcium ammonium nitrate (CAN) with 27% N and urea with 46% N.

Experimental design

The experimental scheme was composed from 4 plots, as it follows: plots 1 and 3 with Exotic wheat variety and plots 2 and 4 with Solehio wheat variety (Table 1). It was developed a bifactorial experiment where **a** factor was wheat variety (Exotic, Solehio) and **b** factor was preceding crop.

Table 1. Crop rotation sequence during 2014-2018

Year	Plot 1	Plot 2	Plot 3	Plot 4
2017-2018	Wheat (Exotic)	Wheat (Solehio)	Wheat (Exotic)	Wheat (Solehio)
2016-2017	Maize	Maize	Sunflower	Sunflower
2015-2016	Sunflower	Sunflower	Maize	Maize
2014-2015	Wheat	Wheat	Wheat	Wheat

Soil and plant analyses

A presentation of performed analyses, methods and apparatus are synthesized in Table 2.

Table 2. Analyses, methods and instrumentation

Analyses	Method	Apparatus
Soil		
pH_{H2O} (1: 2.5)	potentiometry	Hanna pH-meter
Total soluble salts (1: 5), TSS	conductometry	HACH sensION 7
Potassium (mobile form), K_{AL}	flame emission spectrometry	Sherwood 410
Phosphorus (mobile form), P_{AL}	spectrophotometry	CECIL 2041 spectrophotometer
Humus content, H	Walkley-Black-Gogoașă	-
Plant		
Wet gluten	manual method	-
Crude protein content (on the basis of total nitrogen content)	Kjeldahl method	HACH Digesdahl

P_{AL} - mobile form of phosphorus using for extraction ammonium acetate-lactate (AL)
K_{AL} - mobile form of potassium using for extraction ammonium acetate-lactate (AL)

Fertilization and applied treatments

For both wheat varieties was adopted the same technology and phytosanitary treatments (Table 3). The sowing was done in October, first decade and harvesting in June, the third decade.

Table 3. Fertilization and phytosanitary treatments scheme for Exotic and Solehio wheat varieties

Period of time	Fertilizer and phytosanitary treatments	Dose
September, IIIrd decade	NPK 18:46:0	200 kg/ha
February, IIIrd decade	Urea	200 kg/ha
March, IInd decade	amidosulfuron100 g/L + iodosulfuron-methyl-sodium 25 g/L + mefenpyr diethyl 250 g/L (Sekator Progress OD)	0.15 l/ha
	cyproconazole 80 g/L + propiconazole 250 g/L (Artea)	0.40 l/ha
April, Ist decade	calcium ammonium nitrate (CAN)	150 kg/ha
May, Istdecade	tebuconazole 200 g/L + trifloxystrobin 100 g/L (Nativo Pro SC 325)	0.8 l/ha

RESULTS AND DISCUSSIONS

Agrochemical soil analysis (Table 4) indicated that soil reaction was moderately acidic for all plots, excepting plot 2 which is weak acidic. TSS values indicate a non-saline soil.

Humus contents correspond to medium level for all plots. Mobile form of phosphorus, P_{AL}, is very low for plots 3 and 4, meanwhile for plots 1 and 2 correspond to low and very low levels, respectively.

In the case of potassium content, K_{AL}, the found concentrations correspond to middle content for plots 1-3 and normal content for plot 4.

Table 4. Soil agrochemical analysis

Plot	Wheat variety	Preceding crop	pH	TSS, %	H, %	P_{AL}	K_{AL}
						mg/kg	
1	Exotic	Maize	5.73	0.0141	2.37	18.66	90
2	Solehio	Maize	6.11	0.0117	3.24	28	82
3	Exotic	Sunflower	5.66	0.0122	2.49	5.84	96
4	Solehio	Sunflower	5.60	0.0118	2.99	6.33	148

1. Results concerning yield related to preceding crop and wheat variety

The yields for both wheat varieties grown after sunflower are higher, this meaning 114.41% for Exotic and 107.25% for Solehio in comparison with those obtained using maize as preceding crop. The variance analysis concerning the influence of preceding crop on wheat yield indicates significant differences for both wheat varieties (Table 5). The influence of wheat variety on yield presents significant differences for Exotic and Solehio, with superior values given by Solehio.

Table 5. Influence of wheat variety (a factor) and of preceding crop (b factor) on yield (kg/ha)

Yield, kg/ha		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a7700a	a6730b
a2= Solehio	b6950a	b6480b
B constant A variable: LSD 5% = 154* kg/ha; LSD 1% = 297 kg/ha; LSD 0.1% = 749 kg/ha		
A constant B variable: LSD 5% = 167* kg/ha; LSD 1% = 278 kg/ha; LSD0.1% = 519 kg/ha		

There were made interpretations by LSD 5% indicated in the table by *

2. Results concerning plant height related to preceding crop and wheat variety

Wheat grown after maize presents for both varieties the highest values for plant height in comparison with those obtained using sunflower as preceding crop (Table 6). Solehio variety presented the highest values of plant height of 77 cm after maize and 76 cm after sunflower in comparison with Exotic variety.

The variance analysis regarding the influence of preceding crop on plant height indicates significant differences for Exotic, meanwhile for Solehio the differences are no significant. The influence of variety presents significant differences between Exotic and Solehio, with superior values given by Solehio.

Table 6. Influence of wheat variety (a factor) and of preceding crop (b factor) on plant height (cm)

Plant height, cm		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	b67b	b70a
a2= Solehio	a76a	a77a
B constant A variable: LSD 5% = 2.19* cm; LSD 1% = 4.70cm; LSD 0.1% = 13.81cm		
A constant B variable: LSD 5% = 1.60* cm; LSD 1% = 2.65cm; LSD 0.1% = 4.97 cm		

There were made interpretations by LSD 5% indicated in the table by *

3. Results concerning spike length related to preceding crop and wheat variety

Spike lengths for both wheat varieties grown after sunflower as preceding crop are higher than those obtained after maize, this meaning differences of 14.28% (1.5 cm) for Exotic and 7.52% (0.7 cm) for Solehio (Table 7).

The variance analysis concerning the influence of preceding crop on spike length indicates significant differences for Exotic and Solehio. The influence of wheat variety present significant differences between Exotic and Solehio, with higher values given by Exotic.

Table 7. Influence of wheat variety (a factor) and of preceding crop (b factor) on spike length (cm)

Spike length		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a12a	a10.5b
a2= Solehio	b10a	b9.3b
B constant A variable: LSD 5% = 0.92* cm; LSD 1% = 2.02cm; LSD 0.1% = 6.11 cm		
A constant B variable: LSD 5% = 0.56* cm; LSD 1% = 0.93 cm; LSD 0.1% = 1.75 cm		

There were made interpretations by LSD 5% indicated in the table by *

4. Results concerning number of spikes/m^2 related to preceding crop and wheat variety

The number of spikes/m^2 in the case of both wheat varieties grown after sunflower presents superior values in comparison with those obtained after maize, the difference being identical 2.94% (15 spikes/m^2) (Table 8). The highest values were obtained for Solehio variety after sunflower and maize as well, in comparison with Exotic.

The variance analysis regarding the influence of number of spikes/m^2 indicates no significant differences for Solehio and Exotic, as well. The influence of variety presents no significant differences between Solehio and Exotic.

Table 8. Influence of wheat variety (a factor) and of preceding crop (b factor) on no of spikes/m^2

Number of spikes/m^2		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a525a	a510a
a2= Solehio	a535a	a520a
B constant A variable: LSD 5% = 15.30* no. spikes/m^2; LSD 1% = 26.43 no. spikes/m^2; LSD 0.1% = 54.37 no. spikes/m^2		
A constant B variable: LSD 5% = 20.43* no. spikes/m^2; LSD 1% = 33.88 no. spikes/m^2; LSD 0.1% = 63.37 no. spikes/m^2		

There were made interpretations by LSD 5% indicated in the table by *

5. Results concerning hectolitre mass (HLM) related to preceding crop and wheat variety

The values of HLM for wheat grown after sunflower are higher for Exotic and Solehio with differences of 2.75% (2.1 kg/hl) and 3.34% (2.5 kg/hl), respectively in comparison with those obtained after maize (Table 9).

The variance analysis concerning the influence of preceding crop indicates significant differences for both wheat varieties. The influence of wheat variety on HLM presents significant differences between Solehio and Exotic after Maize, with superior values for Exotic variety. After sunflower as preceding crop, the influence on HLM present no significant differences between analyzed wheat varieties.

Table 9. Influence of wheat variety (a factor) and of preceding crop (b factor) on HLM (kg/hl)

HLM, kg/hl		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a78.2a	a76.1b
a2= Solehio	a77.3a	b74.8b
B constant A variable: LSD 5% = 1.04* kg/hl; LSD 1% = 2.03 kg/hl; LSD 0.1% = 5.19 kg/hl		
A constant B variable: LSD 5% = 1.11* kg/hl; LSD 1% = 1.85 kg/hl; LSD 0.1% = 3.46 kg/hl		

There were made interpretations by LSD 5% indicated in the table by *

6. Results concerning thousand kernel weight (TKW) related to preceding crop and wheat variety

The highest values of TKW were obtained for both wheat varieties grown after sunflower, with differences of 14.58% (7 g) for Exotic and 11.11% (5 g) for Solehio in comparison with those obtained when preceding crop was maize (Table 10).

The variance analysis concerning preceding crop on TKW indicates significant differences for Exotic and Solehio varieties. The influence of wheat variety on TKW presents significant differences between Exotic and Solehio, with superior values given by Exotic variety.

Table 10. Influence of wheat variety (a factor) and of preceding crop (b factor) on TKW (g)

TKW, g		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a55a	a48b
a2= Solehio	b50a	b45b
B constant A variable: LSD 5% = 1.25* g; LSD 1% = 2.56 g; LSD 0.1% = 7.10 g		
A constant B variable: LSD 5% = 1.13* g; LSD 1% = 1.87 g; LSD 0.1% = 3.51 g		

There were made interpretations by LSD 5% indicated in the table by *

7. Results concerning wet gluten related to preceding crop and wheat variety

The values found for wet gluten parameter in the case of both wheat varieties were higher when preceding crop was maize, with differences of 1.3% for Exotic and 0.4% for Solehio in comparison with values obtained when preceding crop was sunflower (Table 11).

The variance analysis concerning the influence of preceding crop of wet gluten content indicates no significant differences for Solehio and significant differences for Exotic variety. The influence of wheat variety on wet gluten content presents no significant differences for both wheat varieties.

Table 11. Influence of wheat variety (a factor) and of preceding crop (b factor) on wet gluten (%)

Wet gluten, %		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a26.7b	a28.0a
a2= Solehio	a27.0a	a27.4a
B constant A variable: LSD 5% = 0.74* % ; LSD 1% = 1.52%; LSD 0.1% = 4.25%		
A constant B variable: LSD 5% = 0.66* % ; LSD 1% = 1.09%; LSD 0.1% = 2.04%		

There were made interpretations by LSD 5% indicated in the table by *

8. Results concerning crude protein related to preceding crop and wheat variety

The highest values of crude protein contents were found for both wheat varieties grown after maize but with no significant differences of

0.7% for Exotic and 0.1% for Solehio in comparison with those found for wheat grown after sunflower (Table 12).

The variance analysis concerning the influence of preceding crop on protein content presents no significant differences for Exotic and Solehio varieties. Also, the influence of wheat variety on protein content presents no significant differences between Solehio and Exotic with close values between wheat varieties.

Table 12. Influence of wheat variety (a factor) and of preceding crop (b factor) on crude protein (%)

Crude protein, %		
b= preceding crop / **a**= wheat variety	**b1** sunflower	**b2** maize
a1= Exotic	a12.5a	a13.2a
a2= Solehio	a12.9a	a13.0a
B constant A variable: LSD 5% = 0.69* %; LSD 1% = 1.23%; LSD 0.1% = 2.70%		
A constant B variable: LSD 5% = 0.88* %; LSD 1% = 1.46%; LSD 0.1% = 2.74%		

There were made interpretations by LSD 5% indicated in the table by *

CONCLUSIONS

The research carried out during 2017-2018 in Morteni Commune, Dambovita County with the purpose of investigation for Exotic and Solehio wheat varieties the variability of yield components and quality parameters influenced by preceding crops and by wheat variety generated the conclusions presented below.

1. A significant influence upon the wheat yield, spike lengths, number of spikes/m^2, HLM and TKW parameters was registered under sunflower as preceding crop.

2. Among the two preceding crops, maize determined higher values upon the wheat quality components, wet gluten and crude protein.

3. The highest yield, 7700 kg/ha, was recorded for Exotic after sunflower, and the lowest, 6480 kg/ha, was found for Solehio after maize.

4. Solehio variety presented the highest values of plant height of 77 cm after maize and 76 cm after sunflower in comparison with Exotic variety.

5. Spike lengths for both wheat varieties grown after sunflower as preceding crop are higher than those obtained after maize, this meaning differences of 14.28% (1.5 cm) for Exotic and 7.52% (0.7 cm) for Solehio.

6. The highest values of number of spikes/m^2 were obtained for Solehio variety after sunflower and maize as well, in comparison with Exotic (535 and 520, respectively).

7. The highest HLM value, 78.2kg/hl, was found for Exotic variety grown after sunflower and the lowest was recorded for Solehio after maize, 74.8 kg/hl.

8. The highest values of TKW were obtained for both wheat varieties grown after sunflower: 55 and 50 g, respectively with differences of 7 and 5 g, respectively, in comparison with maize as preceding crop.

9. Wet gluten and crude protein contents were higher for both wheat varieties in the case of maize as preceding crop with close differences between varieties.

ACKNOWLEDGEMENTS

This article was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Ardelean, I., Bandici, Gh. (2013). The influence of crop rotation and nutrition regime on the growth progress of biomass and production in winter wheat. *Natural Resources and Sustainable Development*. Available at SSRN: https://ssrn.com/abstract=2700094

Basso, B., Cammarano, D., Troccoli, A., Chen, D., Ritchie, J. (2010). Long-term wheat response to nitrogen in a rainfed Mediterranean environment: Field data and simulation analysis. *European Journal of Agronomy*, *33*. 132–138.

Bunta, Gh., Bucurean, E. (2011). Researches regarding the yield and quality of some winter wheat varieties in interactions with nitrogen fertilization. *Research Journal of Agricultural Science*, *43*(1), 9–18.

Dogan, R., Goksoy, T.A., Turan, M.Z. (2008). Comparison of the effects of different crop rotation systems on winter wheat and sunflower under rain-fed conditions. *African Journal of Biotechnology*, *7*(22), 4076–4082.

Hlisnikovsky, L., Kunzova, E., Mensik., L. (2016). Winter wheat: results of long term fertilizer experiment in Prague-Ruzyne over the last 60 years. *Plant Soil Environment*, *62*(3), 105–113.

Litke, L., Gaile, Z., Ruža, A. (2017). Nitrogen fertilizer influence on winter wheat yield and yield components depending on soil tillage and forecrop. *Research for Rural Development*, *2*, 54–61.

Lopez-Bellido Garrido, R., Lopez-Bellido, L. (2001). Effects of crop rotation and nitrogen fertilization on soil nitrate and wheat yield under rainfed Mediterranean conditions. *Agronomie*, *21*(6-7), 509–516.

Madjar, R.M., Scăețeanu Vasile, G., Anton, A. (2018). Improve of grain yield and quality of winter wheat by nitrogen inputs. *Scientific Papers, Series A, Agronomy*, *LXI*(I), 310‒315.

Panayotova, G., Kostadinova, S., Valkova, N. (2017). Grain quality of durum wheat as affected by phosphorus and combined nitrogen-phosphorus fertilization. *Scientific Papers, Series A, Agronomy*, *LX*, 356‒363.

Roth, G. (1996). https://extension.psu.edu/crop-rotations-and-conservation-tillage.

Starodub, V., Tabacari, R. (2017). Assessment of the quantitative and qualitative yield of the winter wheat variety "Select" in polyfactorial field experiments. *Scientific Papers, Series A, Agronomy*, *LXI*, 387‒391.

Watson, C.J., Stevens, R.J., Laughlin, R.J., Poland, P. (1992). Volatilization of ammonia from solid and liquid urea surface applied to perennial ryegrass. *The Journal of Agricultural Science*, *119*(2), 223‒226.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

NEW EDITION ON SUNFLOWER CROP - ROMANIAN TECHNOLOGY UNDER CLIMATE CHANGE CONDITIONS IN DOBROGEA

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Abstract

In Dobrogea, about 200,000 ha of sunflower (Helianthus anuus L.) are cultivated, accounting for 20% of the total area from Romania. SC FIRST GRAIN SRL-Amzacea organized in 2018 two polyfactorial experiences in the Amzacea and Fantanele fields in order to improve the technology for the sunflower culture in Dobrogea under climate change. In this paper, the behavior of 5 sunflower hybrids was observed at the attack of the main pests (pathogens: Phomopsis helianthi Munt.-Cvet. et al., Sclerotinia sclerotiorum (Lib.) de Bary, Alternaria helianthi (Hansf.) Tubaki & Nishihara, and the parasite Orobanche cumana Wallr.) and 5 experimental models for weeds and parasite control. The phytosanitary status and the yields obtained in both localities are presented. The two phytosanitary treatments during the vegetation period controlled the pathogens attack. The four ways of herbicide tested have differentiated the attack of weeds and especially the broomrape attack. The highest yield was obtained for Katana hybrid when Listego Plus was applied at 4-6 leaves 4555 kg/ha and 4480 kg/ha in Amzacea and Fantanele, respectively.

Key words*: sunflower hybrids, major pest control, yield.*

INTRODUCTION

In Romania, sunflower crop is the 3rd agricultural crop after maize and wheat. In 2018, around 1.000.000 ha were cultivated with sunflower of which 20% in Dobrogea area. One of the most dangerous plant parasites in Dobrogea area is broomrape (Păcureanu et al., 1998) due to an improper implementation of crop rotation plan. Sunflower crop has shown a significant extension, especially in the south and south-eastern area of Romania (Parker, 1994; Vrânceanu & Păcureanu, 1995). On sunflower crops, the losses can reach 30-70% due to diseases and broomrape (Iliescu et al., 1995).

Farmers can choose the sunflower hybrids from a vastly offer of new foreign hybrids. Therefore, it is required to know their behavior in the presence of main pathogens and their yields under particular abiotic (Ion et al., 2010) and biotic conditions.

Experiments were carried out to improve the technology of sunflower cultivation under climate changes by modifying the seed period about 30 days earlier to avoid the droughts periods of June-August, to develop more vigorous plants and to prevent the attack of broomrape (Manole et al., 2018). Experiments were carried out in order to control weeds that are closely related to the crop itself and some main parasitic weeds using imidazolinone herbicides (Jinga et al., 2016).

The aim of this paper is to present the five experimental plots with different weed control methods and the behaviour of five sunflower hybrids in the presence of main pathogens: white mold (*S. sclerotiorum*), stem canker (*P. helianthi*), alternaria blight (*A. helianthi*) and root parasite plant (*O. cumana*) attack. The yields are presented.

MATERIALS AND METHODS

The hybrids taken into account were imazamox-resistant: Diamantis CL, Bacardi CLP, Neostar CLP, Katana, Odessa.
The experience has been organised at SC FIRST GRAIN SRL-Amzacea, and Fantanele fields - Constanta county on demonstrative plots (Figure 8).
The soil was represented by cambic chernoziom with a profile deeper than other chernozioms, a blackish-brown soil of 40-50 cm thickness with medium texture (Demeter, 2009). The content of nutrients was: mobile P index -72; N index -4; K index -200; humus - 3.11%; neutral pH -7.2. Quantity of precipitations during the vegetation period was presented in Table 1.
The surface of each plot was 1612 m^2. The planting density was 65000 plants per hectare.
In 2017 autumn the field was ploughed at 23-25 cm deep and after that, when the weeds emerged, there was applied glyphosate. Sowing was performed on April 12. The preceding crop was wheat. The seed treatment with fludioxonil 2g/l + metalaxil M 9,7 g/l (MAXIM XL 5l/t) was performed. Concurrent with sowing was applied 190 kg/ha of complex fertilizer 20.10.10+10S. Sunflower had come back on this field after four years. During the crop vegetation a mechanic hoeing was realised. At the same time 200 kg/ha of complex fertilizer (40 N + 13 SO_3) was applied. The pathogens were controlled with two fungicides applications with procloraz (Mirage 1 l/ha) and boscalid + dimoxystrobin (Pictor 0,5 l/ha), respectively.
The attack rate (AR) was calculated with the formula AR = F x I/100 (F% -frequency of the attacked organs, I % -intensity of organs attack). Observations on phytosanitary status of sunflower hybrids were made on July 16 and August 7, 2018 the last being displayed. The yields realised by the five hybrids in the five experimental plots are presented.
The five variants for weed and broomrape control were: V1 - S-metalaclor + terbutilazin applied pre-emergent (Gardoprim Plus 4 l/ha), and imazamox 25 g/l (Listego Plus 1.6 l/ha) applied post-emergent at 6-8 leaves. V2 - Control, V3 - imazamox 25 g/l (Listego Plus 1.6 l/ha) applied post-emergent at 4-6 leaves (Figure 1), V4 - S-metalaclor + terbutilazin applied pre-emergent (Gardoprim Plus 4 l/ha) and IMI 1 l/ha applied post-emergent at 8 leaves, V5-IMI 0.5 l/ha at 2-4 leaves and IMI 0.5 l/ha at 6-8 leaves.

Figure 1. Application of imazamox 25 g/l at 4-6 leaves

Table 1. Precipitation during 2018 growing season of sunflower (Valu lui Traian Station, Constanta, Romania)

	Month								
	Jan.	Feb.	March	Apr	May	June	July	Aug.	
Days	The growing season 2018: Precipitation (mm) for 10-day periods								Sum
1-10	0	9	6	2	64	35	98	0	214
11-20	44	31	37	0	28	0	2	0	142
21-31	19	80	26	0	0	41	47	0	213
Sum	63	120	69	2	92	76	147	0	569
Days	Average 1961-1990 : monthly values of precipitation (mm)								Sum
1-31	27.7	24.0	29.1	31.8	37.7	47.1	38.9	37.4	464.0

RESULTS AND DISCUSSIONS

Precipitation was atypical in June and July representing 223 mm, making possible a high attack of the main pathogens of sunflower as it is shown in the results.

Observations on phytosanitary status of sunflower plots were made on July 16 and August 7, 2018 the last being presented in the tables.

In August, due to the abundant rainfall in July (147 mm), the occurrence of *S. sclerotiorum*, *P. helianthi* and *A. helianthi* pathogenic attack on the studied hybrids was observed.

In the first location, Amzacea, in V1 *S. sclerotiorum* showed AR between 2-8%, *P. helianthi* between 3-8.75%, *A. helianthi* between 17.5-51% and *O. cumana* between 1-8.5% (Table 2).

In V2 *S. sclerotiorum* had shown an AR between 5-8%, *P. helianthi* between 3.75-7.5%, *A. helianthi* between 8.25-38% and *O. cumana* under 1.5-9.5% (Table 3).

Table 2. Phytosanitary status in V1 - Amzacea

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	7	100	7	20	15	3	60	60	36	25	10	2.5
KATANA	5	100	5	20	15	3	50	35	17.5	15	10	1.5
NEOSTAR CLP	2	100	2	25	20	5	80	25	20	100	10	10
BACARDI CLP	8	100	8	30	25	7.5	85	60	51	95	9	8.55
DIAMANTIS CL	7	100	7	35	25	8.75	90	25	22.5	80	6	4.8

Table 3. Phytosanitary status in V2 - Amzacea

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	3	100	3	95	35	33.25	50	25	12.5	25	10	2.5
KATANA	3	100	3	55	15	8.25	65	35	22.75	15	10	1.5
NEOSTAR CLP	5	100	5	60	25	15	70	35	24.5	100	10	10
BACARDI CLP	3	100	3	95	40	38	65	25	16.25	95	10	9.5
DIAMANTIS CL	7	100	7	85	15	12.75	75	30	22.5	80	10	8.0

In V3 *S. sclerotiorum* had shown an AR between 5-8%, *P. helianthi* between 5-16.25%, *A. helianthi* between 15-25.5% and *O. cumana* under 1% (Table 4).

In V4 *S. sclerotiorum* had shown an AR between 1-6%, *P. helianthi* between 3-9%, *A. helianthi* between 11.25-24% and *O. Cumana* under 4.5% (Table 5).

Table 4. Phytosanitary status in V3 - Amzacea

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	8	100	8	45	25	11.25	75	20	15	0	0	0
KATANA	5	100	5	65	25	16.25	85	30	25.5	0	0	0
NEOSTAR CLP	7	100	7	25	20	5	80	25	20	10	3	0.3
BACARDI CLP	5	100	5	30	25	7.5	85	20	17	20	5	1
DIAMANTIS CL	5	100	5	25	20	5	70	25	17.5	10	5	0.5

Table 5. Phytosanitary status in V4 - Amzacea

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	2	100	2	20	15	3	80	30	24	0	0	0
KATANA	1	100	1	25	20	5	75	15	11.25	0	0	0
NEOSTAR CLP	6	100	6	30	20	6	65	35	22.75	20	5	1
BACARDI CLP	5	100	5	45	20	9	65	30	19.5	70	5	3.5
DIAMANTIS CL	5	100	5	25	20	5	68	25	17	75	6	4.5

In V5, *S. sclerotiorum* showed an AR between 2-4%, *P. helianthi* between 3-10.5%, *A. helianthi* between 7.5-25.5% and *O. cumana* under 0.3% (Table 6).
In Fantanele field in V1, *S. sclerotiorum* hadn't shown any AR, *P. helianthi* had shown an AR between 7-13.5%, *A. helianthi* between 24-38% and *O. cumana* under 2% (Table 7).
In V2, *S. sclerotiorum* hadn't shown any AR, *P. helianthi* between 14.4-20%, *A. helianthi* between 27-33.25% and *O. cumana* between 0.1-6% (Table 8).
In V3, *S. sclerotiorum* hadn't shown any AR, *P. helianthi* between 12.4-25.5%, *A. helianthi* between 21.25-40% and *O. cumana* did not show any AR (Table 9).
In V4, *S. sclerotiorum* hadn't shown any AR, *P. helianthi* between 10-19.5%, *A. helianthi* between 27-38% and *O. cumana* hadn't shown any AR (Table 10).
In V5, *S. sclerotiorum* and *O. cumana* did not show AR, *P. helianthi* showed values between 2.5-9% and *A. helianthi* between 25.5-38% (Table11).

Table 6. Phytosanitary status in V5 - Amzacea

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	2	100	2	15	20	3	30	25	7.5	0	0	0
KATANA	4	100	4	25	15	3.75	75	20	15	0	0	0
NEOSTAR CLP	2	100	2	28	30	8.4	45	50	22.5	0	0	0
BACARDI CLP	2	100	2	35	30	10.5	75	20	15	0	0	0
DIAMANTIS CL	4	100	4	25	20	5	85	30	25.5	10	3	0.3

Table 7. Phytosanitary status in V1 - Fantanele

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	0	0	0	35	20	7	80	30	24	0	0	0
KATANA	0	0	0	25	15	3.75	85	30	25.5	0	0	0
NEOSTAR CLP	0	0	0	45	30	13.5	85	35	29.75	20	3	0.6
BACARDI CLP	0	0	0	45	25	11.25	90	30	27	25	5	1.25
DIAMANTIS CL	0	0	0	45	30	13.5	95	40	38	40	5	2

Table 8. Phytosanitary status in V2 - Fantanele

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	0	0	0	75	20	15	92	30	27.6	20	10	2.0
KATANA	0	0	0	80	18	14.4	95	35	33.25	1	10	0.1
NEOSTAR CLP	0	0	0	95	18	17.1	98	30	29.4	5	10	0.5
BACARDI CLP	0	0	0	80	25	20	95	35	33.25	30	20	6
DIAMANTIS CL	0	0	0	95	20	19	90	30	27	40	10	4

Table 9. Phytosanitary status in V3 - Fantanele

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	0	0	0	80	15	12	85	25	21,25	0	0	0
KATANA	0	0	0	85	30	25,5	98	35	34,3	0	0	0
NEOSTAR CLP	0	0	0	85	25	21,25	100	40	40	0	0	0
BACARDI CLP	0	0	0	75	25	18,75	100	30	30	0	0	0
DIAMANTIS CL	0	0	0	80	25	20	95	30	28,5	0	0	0

Table 10. Phytosanitary status in V4 - Fantanele

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	0	0	0	55	30	16,5	80	40	32	0	0	0
KATANA	0	0	0	65	30	19,5	90	30	27	0	0	0
NEOSTAR CLP	0	0	0	65	20	13	95	40	38	0	0	0
BACARDI CLP	0	0	0	55	20	11	95	35	33,25	0	0	0
DIAMANTIS CL	0	0	0	50	20	10	95	40	38	20	3	0,6

Table 11. Phytosanitary status in V5 - Fantanele

HYBRID	Pathogens and parasite											
	Sclerotinia sclerotiorum			*Phomopsis helianthi*			*Alternaria helianthi*			*Orobanche cumana*		
	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)	F (%)	I (%)	AR (%)
ODESSA	0	0	0	25	15	3,75	85	30	25,5	0	0	0
KATANA	0	0	0	35	10	3,5	95	35	33,25	0	0	0
NEOSTAR CLP	0	0	0	45	20	9	95	40	38	0	0	0
BACARDI CLP	0	0	0	25	10	2,5	95	40	38	0	0	0
DIAMANTIS CL	0	0	0	30	15	4,5	85	30	25,5	0	0	0

The yields obtained in the first location had values between 2805-3030 kg/ha at V2, 3710-4222 V5, 3780-4555 kg/ha at V3, 3810-4480 at V4 and 3900-4410 kg/ha at V1 (Figure 2).

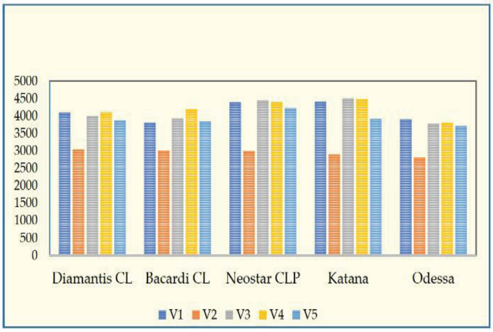

Figure 2. Yields (kg/ha) obtained in Amzacea plots

The yields obtained in the second location had values between 2810-3100 kg/ha at V2, 3810-4250 at V1, 3850-4480 kg/ha at V3, 3780-4300 at V4 and 3880-4300 kg/ha at V5 (Figure 3).

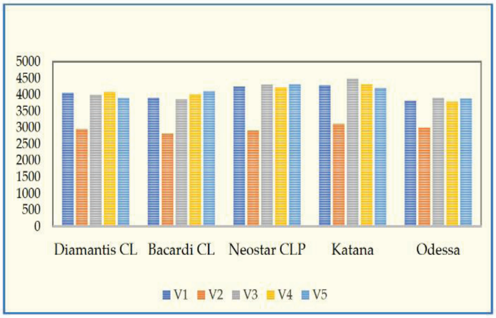

Figure 3. Yields (kg/ha) obtained in Fantanele plots

As a result of these experiments is recommended the following technological elements: a pre-emergent herbicide with glyphosate in autumn and one with S-metalaclor + terbutilazin in spring, during the vegetation an IMI herbicide applied at 4-6 leaves, using a competitive hybrids of new generation which be able to ensure considerable yields.

Weeds problem in sunflower crops in Amzacea and Fantanele fields were: *Chenopodium album*, *Amaranthus blitoides*, *Convolvulus arvensis*, and *Echinochloa crus-galli*.

Weed species, which had a density of 3-5 plots, became a problem for diseases and plants per square meters in sunflower cultivated yields in untreated plots (Figures 4, 6, 7).

Figure 4. *P. helianthi* attack

Figure 5. *A. helianthi* attack

Figure 6. *S. sclerotiorum* attack

Figure 7. *O. cumana* attack

Figure 8. Overview of the experimental field

CONCLUSIONS

The 2018 was an atypical year for Dobrogea, with precipitations over limits in June and July, which favoured the attack of the pathogens.

The two phytosanitary treatments during the vegetation period controlled the pathogens attack.

The four ways of herbicide tested have differentiated the attack of weeds and especially the broomrape attack. The production obtained in the 5 herbicide blocks and 5 experienced hybrids were clearly differentiated. V4 determined constant high yields throughout all hybrids. The highest yields were obtained for V3 at the Neostar and Katana hybrids 4444 and 4555 kg/ha, respectively in Amzacea and 4300-4480 kg/ha, respectively in Fantanele.

ACKNOWLEDGEMENTS

This research work was carried out within the Contract No. 694 – April 16 2018 - SC SPORT AGRA SRL, Amzacea.

REFERENCES

Iliescu, H., Popescu, A., Ionita, A., Csep, N., Jinga V., Iordache, E. (1995). Integrated control measures for sunflower crop as elements included in sustainable agriculture. *Breeding and cultivation of wheat, sunflower and legume crops in the Balkan countries*. Albena, Bulgaria, 509–515.

Ion, V., Ştefan, V., Dumbravă, M., Ion, N., Băşa, A.Gh. (2010). Yield results obtained from an assortment of sunflower hybrids cultivated at Moara Domnească research farm in the period 2006-2008. *Scientific Papers*, *Series A, Agronomy*, *53*, 364–370.

Demeter, T. (2009). *Pedologie generala*. Editura Credis, Bucharest, p.174.

Jinga, V., Dudoiu, R., Lupu, C., Manole, D., Giumba, A. M. (2016). Research regarding the improvement of sunflower crop technology in south-eastern Dobrogea in the context of current climate changes. *Romanian Journal for Plant Protection*, *9*, 16–21.

Jinga, V., Paica, A., Dudoiu, R., Gradila, M., Manole, D., Giumba, A.M. (2016a). The behaviour of some sunflower cultivars to the major pest agents in the south-eastern area of Romania. *Proceedings of the 19th International Sunflower Conference*, 808–815.

Manole, D, Jinga, V., Giumba, A.M., Dudoiu, R., Cristea, S. (2018). Researches regarding new and improved technologies for sunflower and sorghum crops in the context of climate changes in Dobrogea region. *Proceedings Agriculture for Life, Life for Agriculture Conference*, *1*(1), 79–85.

Parker, C. (1994). The present state of the *Orobanche* problem. *Proceedings of the Third International Workshop on Orobanche and realted Striga research, Amsterdam, the Netherlands*, 17–26.

Păcureanu-Joița, M., Vrânceanu, Al.V., Soare, G., Marinescu, A., Sandu, I. (1998). The evaluation of the parasite-host interaction in the system *Helianthus annus* L. - *Orobanche cumana* Wallr. in Romania. *Proccedings of 2nd Balkan Symposium of Field Crops*, *Novi Sad, Yugoslavia*, 153–155.

Vrânceanu, A.V., Păcureanu-Joita, M. (1995). Evaluation of an international set of sunflower hybrids in relation to broomrape *Orobanche cumana* Wallr. resistance. *Romanian Agricultural Research*, *3*, 19–24.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

YIELD AND NUTRIENT USE EFFICIENCY OF FIELD CROPS GROWN IN THE REGION OF SADIEVO, SOUTH BULGARIA

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Abstract

Fertilizer experiment in four field's crop rotation (wheat, barley, maize, and sunflower) was carried out in the region of Sadievo, Bulgaria, during the period 2009-2012. The aim of the research was to study the effect of fertilization on crop productivity and nutrient use efficiency (NUE) of the mentioned four field crops. The fertilizer treatments were: Control; N; P; K; NP; NK; PK; NPK. Agronomic Efficiency (AE), Partial nutrient balance (PNB) and Recovery efficiency (RE) were the indicators calculated for assessing the NUE. The fertilization was very effective practice – NPK treatment ensured 71% higher yields for maize, 64% for wheat, 60% for barley and 51% for sunflower in comparison with control, on average for 4 years period. In spite of the relatively good fertilization effect, AE data was lower than reported as typical levels for cereals crops which showed relatively low productivity of unit nutrient input. PNB values for N showed that removed nitrogen amounts were higher and the deficit was less pronounced in wheat. In maize and sunflower PNB values for N were high and showed a severe nitrogen balance deficit. PNB and RE values for P showed that the fertilization rates could be decreased, while PNB and RE for K indicated that for maintaining the soil K content, the fertilization with K was recommendable in spite of the relatively low agronomic efficiency.

Key words: *Nutrient use efficiency, NPK, wheat, barley, maize, sunflower.*

INTRODUCTION

One of the main crop management practices affecting field's crop yields is the application of mineral fertilizers. The nutrient which strongly affects yields is nitrogen (Lopez-Bellido et al., 2005; Kismányoky & Tóth, 2010; Boukef et al., 2013). In most of the cases unbalanced application and utilization of nitrogen, phosphorus and potassium fertilizers is a reason for low efficiency of fertilization (Romheld, 2006; Nikolova, 2010). The current nitrogen application strategies for field crops cultivation are extremely inefficient, with nitrogen efficiency ranging from 14% to 59% (Melaj et al., 2003; Loppez-Bellido et al., 2005). Baligar et al. (2001) summarizing results from different publications estimates that overall efficiency of applied fertilizers has been below 50% for N, less than 10% for P, and about 40% for K. The lower the level of soil fertility is, the higher the agronomic efficiency of applied phosphorus and potassium is (Fixen, 2009). One modern method for assessing the effectiveness of fertilization is by determinating of Nutrient Use Efficiency (NUE) indicators (Rao, 2007). These indicators represent the ability of plants to translate the uptaken nutrients into economic yield-grains (Delogu et al., 1998). The aim of the efficient use of nutrients is to increase productivity of crops, to minimize nutrient losses and to maintain fertility of the soil (Mikkelsen et al., 2012; Fixen et al., 2015). Grain yield and NUE depend on many factors such as the soil, climate conditions, environment, grown cultivars etc. (Delogu et al., 1998; Noulas et al., 2010; Zhu et al., 2011). Asplund et al. (2016) made connection between the amount of chlorophyll in the plants leafs, its correlation with N content and NUE indicators in the pot and field experiments with more than 50 spring and winter varieties of wheat. NUE can also be expressed through agronomic, physiological and economic indicators (Rao, 2007). The optimization of fertilization of crops should be determined by conducting precise local experiments. The aim of the present research was to study the effect of fertilization on crop productivity of field crops grown in the region of Sadievo, South Bulgaria, and to estimate some NUE indicators.

MATERIALS AND METHODS

Omission plot trials in four field crop rotation - wheat, maize, barley, and sunflower was conducted in region of Sadievo, Bulgaria (42°31'38.1"N 26°05'39.3"E), during the period 2009-2012. Soil type was clayed Eutric Vertisols with clay content of 56.13%. The trial included eight fertilizing variants with nutrient addition and omission: control; N; P; K; NP; NK; PK; NPK. Fertilization rates based on soil diagnostic are presented on Table 1.

Table 1. Applied fertilizer rates for every crop kg ha^{-1}

Nutrients	Wheat	Barley	Maize	Sunflower
N	100	80	140	80
P_2O	120	120	120	120
K_2O	80	80	80	80

The used fertilizers in the experiment were ammonium nitrate (N - 33.4%), triple superphosphate (P_2O_5 - 46%), and potassium chloride (K_2O - 60%). The obtained data was processed for calculating of three agronomic indexes indicating the nutrient use efficiency:
AE - Agronomic Efficiency of applied nutrient (kg kg^{-1}) = ($yield_{nutrient}$ - $yield_{control}$)/nutrient rate; PNB - Partial nutrient balance (kg kg^{-1}) = removed nutrient/fertilizing rate;
RE - Recovery efficiency of applied nutrient (kg kg^{-1}) = (removed nutrient at fertilizing treatment - removed nutrient at control)/fertilizing rate.

RESULTS AND DISCUSSIONS

The fertilization was an effective practice at all grown crops (Figure 1 and Table 2). The most effective nutrient was nitrogen. Single application of nitrogen increased yields from winter cereal crops (wheat and barley) with 41% on average. The increase of the yields for maize and sunflower was 36 %. Double nutrient applications - NP and NK also ensured formation of higher yields especially at wheat, barley and maize.

The average yield increase of these crops was from 45 to 62% higher than the control. The increase in the same treatments was lower at sunflower, 26-28%, more than in the control treatment. Single addition of P and K and their double combination (PK), because of N omission had weak effect on yield, especially at sunflower. The application of the three nutrients (balanced NPK variant) had the best results but the increased yields at wheat, barley and maize was comparatively small in comparison with NP and NK variants. NPK fertilization demonstrates the highest yield increase at sunflower - 51% in comparison with control variant. The effect of other fertilizing treatments on the yield of sunflower was comparatively small (Table 2).

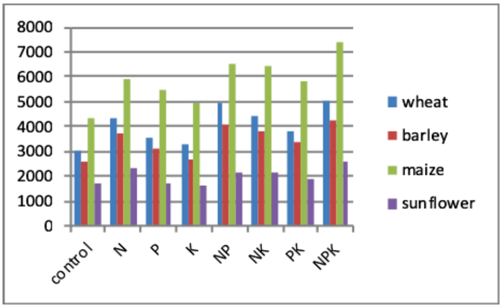

Figure 1. Yields of field crops
(average for period 2009-2012) (kg ha^{-1})

Table 2. Relative yields of field crops (average for period 2009-2012) (%)

Variants	Wheat	Barley	Maize	Sunflower
control	100	100	100	100
N	141.0	141.9	136.1	136.3
P	116.3	117.7	127.2	103.4
K	106.9	101.8	114.1	98.1
NP	161.4	156.3	150.6	129.0
NK	145.6	144.8	149.8	126.3
PK	123.7	129.6	134.7	111.6
NPK	163.8	160.6	171.3	151.2

AE is index representing the ability of the crops to increase yield in response to applied fertilizers. According to Fixen et al. (2015) and Liu et al. (2011) AE_N at wheat ranges from 10-30 kg kg^{-1}. The data for wheat grown in Sadievo region were in the same range for all variants with nitrogen application - from 12 to 14 kg kg^{-1} (Table 3). These figures were close to the lower value for this indicator according to the above-mentioned authors. Delogu et al. (1998), Baligar et al. (2001) and Zhu et al. (2011) found out that the values for AE_N for wheat decreased with increasing of the nitrogen rate. According to the same authors, the index is also dependent on the time of application of nitrogen as pre–sowing or top–dressing.

Another factor which influences the AE indicator is grown cultivar (Noulas et al., 2010). Agronomic efficiency (AE) of phosphorus and potassium application was less than AE for nitrogen fertilizers. The PNB_N indicator was the lowest at variant with single N addition, 0.96 kg kg^{-1}. It was increased at double nutrient addition (NK and NP). The highest value for this indicator was determined at balanced nutrient combination (NPK). The values for double and triple fertilizer combinations were above 1, which indicated that the wheat had removed higher amount of nitrogen from the soil than the amount of the element applied with the fertilizers. This suggests negative N balance, better manifested at the most productive NPK treatment. Nitrogen fertilizer rates for wheat grown in Sadievo region should be increased when balanced with P and K crop fertilization is foreseen. The published data about PNB_N index by Zhu et al. (2011) showed considerable variation of the index depending on the rate of N fertilizers and time of their application. The data for PNB in Sadievo indicated that applied amounts of P and K fertilizers cover the crop needs of both elements (Table 3) and exceeded the P and K uptakes from the soil. The data for PNB_P (0.2-0.3 kg kg^{-1}) indicated strongly positive P balance which showed that the P rate could be reduced in order to increase AE_P. K balance was also positive but the higher PNB_K data (0.6-0.9 kg kg^{-1}) indicated that at most productive practice of balanced NPK fertilization, K application is recommendable in order to maintain the soil fertility.

The RE indicator represents the actual amount of removed nitrogen by crop from the applied fertilizers (in comparison with the control treatment). According to Mosier et al. (2001) and Fixen et al. (2015), the RE values of nitrogen for wheat range between 0.4-0.7 kg kg^{-1}. The highest values for RE_N in our trials were found for NP and NPK variants. The RE_P and RE_K were low for all treatments. Only RE_K from NPK variant showed higher value - 0.34 kg kg^{-1}.

Table 3. Nutrient use efficiency for wheat (average for the period 2009-2012)

variant	AE kg kg^{-1}			PNB kg kg^{-1}			RE kg kg^{-1}		
	N	P	K	N	P	K	N	P	K
N	13			0.96			0.33		
P		6			0.2			0.04	
K			4			0.6			0.01
NP	14	6		1.20	0.3		0.48	0.11	
NK	12		4	1.07		0.8	0.32		0.19
PK		5	5		0.2	0.7		0.04	0.06
NPK	12	5	3	1.26	0.3	0.9	0.48	0.10	0.34

Fixen et al. (2015), determined values in the range 15-30 kg kg^{-1} as typical AE_N index for barley. The same like at wheat tendency for AE_N indicator was found out by Delogu et al. (1998). The values for AE decreased with increasing of nitrogen rate. AE_N index for barley in the region of Sadievo was below this range (Table 4). This suggest for low efficiency of applied nitrogen fertilizers in the region. The lowest AE_N index was found at NPK treatment - 10 kg kg^{-1}. Delogu et al (1998) found that *A*E mean values of barley and wheat were similar in experiment in Italy and ranged between 8.7 and 9.2 kg kg^{-1}. The data for region of Sadievo about AE_N was also similar for both crops but were higher in comparison with the cited study (Tables 3 and 4) and ranged between 10 and 14 kg kg^{-1}. This suggests that both species respond equally to N fertilization.

The data for PNB_N from the similar experiment conducted in the region of Pomorie, Bulgaria, were in the range between 0.86-0.94 (Manolov et al., 2018). The results from Sadievo were almost the same in all treatments (1.14–1.17 kg kg^{-1}) but higher than in the Pomorie region. They were also higher compared to the typical values for this index according to Fixen et al. (2015) (0.7-0.9 kg kg^{-1}). This indicated that applied fertilizer rate can not cover the needs of nitrogen for grown barley in the region of Sadievo and should be increased. Considerable part of applied P was not uptaken by crop

(PNB_P - 0.2 kg kg^{-1}). This should lead to increasing of soil phosphorus reserves. A bigger part of the applied K with fertilizers was absorbed by crop (PNB_P - 0.6-0.9 kg kg^{-1}). The results indicated that part of applied K remains in the soil and increased its potassium reserves. The barley adsorbed less than the half of applied N rate (RE_N - in the range 0.35-0.44 kg kg^{-1}), while the adsorbed P from fertilizers was very low (0.03-0.09 kg kg^{-1}). The RE_K for single application of the element was very low 0.07 kg kg^{-1}. The combine application of K with N and P increased considerably the data for RE_K. The highest amount of absorbed K from fertilizers was found at variant NPK (0.31 kg kg^{-1})

Table 4. Nutrient use efficiency for barley (average for the period 2009-2012)

Variant	AE kg kg^{-1}			PNB kg kg^{-1}			RE kg kg^{-1}		
	N	P	K	N	P	K	N	P	K
N	14			1.17			0.44		
P		5			0.2			0.03	
K			1			0.6			0.07
NP	13	4		1.16	0.3		0.38	0.10	
NK	14		4	1.14		0.8	0.39		0.28
PK		6	6		0.2	0.7		0.09	0.20
NPK	10	4	2	1.17	0.2	0.9	0.35	0.09	0.31

The AE_N data for maize was similar at different treatments (9-11 kg kg^{-1}) (Table 5) and was lower than typical ones for this crop (15-30 kg kg^{-1}) (Fixen et al., 2015). Fixen et al. (2015) indicated values ≥ 7 kg kg^{-1} as typical for AEp for maize. Walsh et al. (2012) found out decreasing of NUE_N with the increasing of the nitrogen rates at maize. Such values of 8 kg kg^{-1} for experiment in Sadievo were found at single P addition and NPK variants. The AE_K was also the highest at NPK treatment reaching 11 kg kg^{-1}.

The PNB_N values for the trial in Sadievo exceeded 1 for all treatments and were higher than the typical values (0.7-0.9 kg kg^{-1}) (Fixen et al., 2015). Therefore, the quantity of applied nitrogen was less than the quantity of N removed by maize. There were no big differences between the results for PNB_P (Table 5). The results were very low which means that considerable part of the applied P remained in the soil and increased its phosphorus reserves. Because of high K removal by maize, a large amount of potassium was taken from the soil reserves (PNB_K was in the range 1.4-2.0 kg kg^{-1}). This means that fertilizer K rates should be increased in order to maintain soil potassium reserves at optimal level.

For the maize, grown in the region of Sadievo, the values of RE for the three nutrients were highest at the NPK treatment. At the treatments with single nutrient addition or nutrient omission the RE values were much lower. The optimal values in NPK variant could be explained by the higher yield when balanced nutrition is ensured. The higher biomass formation and respective higher nutrient uptake lead to better utilization of applied nutrients.

Table 5. Nutrient use efficiency for maize (average for the period 2009-2012)

Variant	AE kg kg^{-1}			PNB kg kg^{-1}			RE kg kg^{-1}		
	N	P	K	N	P	K	N	P	K
N	11			1.05			0.35		
P		8			0.3			0.12	
K			9			1.5			0.29
NP	9	3		1.24	0.4		0.39	0.17	
NK	11		3	1.22		2.0	0.43		0.49
PK		4	3		0.4	1.4		0.13	0.23
NPK	11	8	11	1.31	0.5	1.6	0.92	0.30	0.46

AE for N, P and K for sunflower was comparatively low (Table 6). The lowest AE values were registered for P. For the three nutrients AE was the highest at the balanced NPK treatment, reaching 8 kg kg^{-1} for N, 5 kg kg^{-1} for K and 4 kg kg^{-1} for P. Similar was the agronomic efficiency at the treatment with P omission (NK) and could be concluded that at the conditions of Sadievo phosphorus was the nutrient with lower efficiency for sunflower.

Table 6. Nutrient use efficiency for sunflower (average for the period 2009-2012)

Variant	AE kg kg^{-1}			PNB kg kg^{-1}			RE kg kg^{-1}		
	N	P	K	N	P	K	N	P	K
N	8			1.29			0.42		
P		1			0.2			0.02	
K			1			1.0			0.02
NP	5	4		1.29	0.3		0.37	0.09	
NK	7		6	1.28		1.4	0.48		0.43
PK		0	3		0.2	1.1		0.08	0.15
NPK	8	3	5	1.49	0.3	1.8	0.57	0.12	0.84

PNB for N and K were much higher than 1 for all variants, but low in comparison with data received for the region of Pomorie (Manolov et al. 2018) (Table 6). The results showed that the crop uptakes much more N and K from the soil than was applied with fertilizers. This indicated that the fertilization rates for both nutrients should be increased. PNB_P kept the tendency for low values found at the other crops included in the experiment. Considerable amount of applied P by fertilizers remained in the soil. This was confirmed by the values for the indicator RE_P which were very low especially for treatment P (Table 6). RE_N varies slightly between the treatments and was in the range 0.37-0.57. RE_K varies significantly (0.02-0.84) due to the big yield differences depending on addition or omission of nitrogen. For the three nutrients highest RE values were recorded at NPK treatment indicating that balanced fertilization led to formation of the highest yield, respectively the highest removal of nutrients and better utilization of applied nutrients with fertilizers.

CONCLUSIONS

The fertilization of all studied field crops in the region of Sadievo was a very effective practice. The highest efficiency of fertilization was registered at maize, followed by winter cereals (wheat and barley) and sunflower. The nitrogen was the main limiting nutrients in the region of Sadievo. The balanced NPK fertilization demonstrated the highest efficiency. Similar to NPK effect was obtained under NP and NK treatments for wheat and barley. The values for Agronomic Efficiency showed relatively low efficiency of unit nutrient input in spite of the relatively good fertilization effect. Partial Nutrient Balance and Recovery Efficiency for phosphorus indicated that the fertilization rates could be decreased in order to increase the Agronomic efficiency. Both indices for potassium showed that for maintaining the soil K content, fertilization with K is recommended despite the relatively low agronomic efficiency of the element.

ACKNOWLEDGEMENTS

The study was realized thanks to the financial and methodical support of International Plant Nutrition Institute (IPNI), USA.

REFERENCES

Asplund, L., Bergkvist, G., Weih, M. (2016). Functional traits associated with nitrogen use efficiency in wheat. *Acta Agriculturae Scandinavica, Section B-Soil & Plant Science*, *66*(2), 153–169.

Baligar, V.C., Fageria, N.K., He, Z.L. (2001). Nutrient use efficiency in plants. *Communications in Soil Science and Plant Analysis*, *32*(7-8), 921–950.

Boukef, S., Karmous, C., Trifa, Y., Rezgui, S. (2013). Durum wheat grain quality traits as affected by nitrogen fertilization sources under Mediterranean rainfed conditions. *Journal of Agriculture and Sustainability*, *4*(1), 99–114.

Delogu, G., Cattivelli, L., Pecchioni, N. De Falcis, D., Maggiore, T., Stanca, A.M. (1998). Uptake and agronomic effciency of nitrogen in winter barley and winter wheat. *European Journal of Agronomy*, *9*(1), 11‒20.

Fixen, P.E. (2009). Nutrient Use Efficiency in the Context of Sustainable Agriculture. In: Espinosa, J,. García F. (eds.), *Nutrient Use Efficiency*, International Plant Nutrition Institute (IPNI), USA, 1‒10.

Fixen, P.E., Brentrup, F., Bruulsema, T.W., Garcia, F., Norton R., Zingore S. (2015). *Nutrient/fertilizer use efficiency: Measurement, current situation and trends*. International Fertilizer Industry Association (IFA), International Water Management Institute (IWMI), International Plant Nutrition Institute (IPNI), and International Potash Institute (IPI), Paris, France. Available from: http://www.academia.edu/24038256

Kismányoky, T., Tóth, Z., (2010). Effect of mineral and organic fertilization on soil fertility as well as on the biomass production and N utilization of winter wheat *Ttriticum aestivum* L.) In a long-term cereal crop rotation experiment (IOSDV). *Archives of Agronomy and Soil Science*, *56*(4), 473‒479.

Lopez-Bellido, L., Lopez-Bellido, R.J., Redondo, R. (2005). Nitrogen efficiency in wheat under rainfed Mediterranean conditions as affected by split nitrogen application. *Field Crops Research*, *94*(1), 86‒97.

Liu, X., He, P., Jin, J. (2011). A Long-term Analysis of Factors to Improve Nutrient Management for Winter Wheat Production in China. *Better Crops*, *95*(3), 16‒18.

Manolov, I., Valeva, N., Stamenov, J., Kostadinova, S., Nikolova, M. (2018). Estimating nutrient use efficiency in field crops grown on Pomorie region, Bulgaria. *Proceedings of the 53rd Croatian and 13th International Symposium on Agriculture*, 18-23 February, Vodice, Croatia, 309‒3013.

Melaj, M.A., Echeverria, H.E., Lopez, S.C., Studdert, G., Andrade, F., Barbaro, N.O. (2003). Timing of nitrogen fertilization in wheat under conventional and no-tillage system. *Agronomy Journal*, *95*(6), 1525‒1531.

Mikkelsen, R., Jensen, T.L., Snyder, C., Bruulsema, T.W. (2012). Nutrient Management Planning and Accountability. Chapter 9. In 4R Plant Nutrition: *A Manual for Improving the Management of Plant Nutrition*. Bruulsema T.W., Fixen P.E., Sulewski G.D. (ed.), International Plant Nutrition Institute, Norcross, GA, USA.

Mosier, A.R., Bleken, M.A., Chaiwanakupt, P., Ellis, E.C., Freney, J.R., Howarth, R.B., Matson, P.A., Minami, K., Naylor, R., Weeks, K.N., Zhu, Z.L. (2001). Policy implications of human-accelerated nitrogen cycling. *Biogeochemistry*, *52*(3), 281‒320.

Noulas, C., Liedgens, M., Stamp, P., Alexiou, I., Herrera, J. (2010). Subsoil root growth of field grown spring wheat genotypes (*Triticum aestivum* L.) Differing in itrogen use efficiency parameters. *Journal of Plant Nutrition, 33*(13), 1887‒1903.

Nikolova, M. (2010). *Potassium - an important nutrient for crop yields and quality*, pp. 86, IPI *Research Topics, No 18*, International Potash Institute, Horgen, Switzerland.

Romheld, V. (2006). Balanced fertilization for crop sustainability: the neglect of potassium. In *The Balanced Fertilization for Sustaining Crop Productivity,* Benbi D.K., Brar M.S., Bansal S.K. (eds.), 205‒218. Ludhiana, India, Swami Printers.

Rao, T.N. (2007). Improving Nutrient Use Efficiency: The Role of Beneficial Management Practices. *Better Crops – India*, *1*(1), 6‒7.

Walsh, O., Raun, W., Klatt, A., Solie, J. (2012). Effect of delayed nitrogen fertilization on maize (*Zea mays* L.) grain yields and nitrogen use efficiency. *Journal of Plant Nutrition, 35*(4), 538–555.

Zhu, X., Guo, W., Ding, J., Li, C., Feng, C., Peng, Y. (2011). Enhancing nitrogen use efficiency by combinations of nitrogen application amount and time in wheat. *Journal of Plant Nutrition, 34*(12), 1747‒1761.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESEARCHES ABOUT THE INFLUENCE OF THE HYBRID AND THE IRRIGATION REGIME ON THE SIZE FEATURES OF THE MAIZE COB

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Abstract

This paper presents the results of a study conducted at ARDS Marculesti in 2017 and 2018 on the influence of 5 new maize hybrids under different irrigation conditions on the size features of the maize cobs, namely the number of rows per cob and the number of grains per row. The experiment was set up using the two-factor subdivided plot method, the A-factor was the maize hybrid and B factor-irrigation regime. The subdivisions of A factor: a1- P9175 (FAO 330), a2 - KWS BELLAVISTA (FAO 330), a3 - KWS SMARAGD (FAO 350), a4 - KWS KASHMIR (FAO 370) and a5 - KWS DURANGO (FAO 480). The subdivisions of B factor: b1 – unirrigated; b2 – stressed at and after flowering; b3 – stressed before flowering and b4 – full irrigated throughout the vegetation period. Regarding the number of rows on the cob, the best results were obtained by the hybrids KWS Bellavista and KWS Smaragd and the lowest, by the hybrid P9175.

Key words*: maize hybrids, maize cob size features, irrigation, water stress.*

INTRODUCTION

In the current context, when the global climate is constantly changing and tends to reach air temperature values that have not yet been observed since meteorological measurements, the maize crop encounters challenges related particularly on pollen grain fertility (Jonghan K. & Piccinni G., 2009; Lizaso J.I. et al., 2018; Năescu V. & Eliana Alionte, 2008; NeSmith D.S. & Ritchie J.T., 1992; Niaz Ahmad & Rameshwar S. Kanwar, 1991; Oşvat I.M., 2015).
Maize is the main staple crop at national level. In the climatic conditions present during the last 15 years there have been problems with adapting the new hybrids to the warmer climate conditions and to the lower atmospheric humidity recorded by the meteorological stations in the major agricultural areas of the country.
The hydric stress encountered by the maize plant during the formation of the floral primordia is of great importance in the perspective of obtaining cobs with a high number of rows and grains in a row (Schoper John B. et al., 1987; Setter Tim L. et al., 2001; Stegman E.C., 1982).
In this context, we considered to set up an experiment that takes into account the stress that the maize plant undergoes before, during and after flowering. It was also taken into account the duration in days of coincidence of female and male flowers in order to ensure pollination.

MATERIALS AND METHODS

The researches have been done in the experimental field of Agricultural Research and Development Station (ARDS) Marculesti, on a clayey vermic loamy soil and it had 2 factors, the factor A - maize hybrid and the factor B - irrigation regime. The setting up method was randomized blocks, with three replications. The investigated hybrids (factor A) were: a1- P9175 (FAO 330), a2 - KWS BELLAVISTA (FAO 330), a3 - KWS SMARAGD (FAO 350), a4 - KWS KASHMIR (FAO 370) and a5 - KWS DURANGO (FAO 480). The B factor (irrigation regime and period) had 4 graduations, namely: b1 - unirrigated; b2 - stressed at and after flowering; b3 - stressed before flowering and b4 - full irrigated throughout the vegetation period. The irrigation method was drip to drip and there were automatic sensors to determine the actual soil moisture. Sowing and harvesting were done mechanically using the seed drilling machine and BAURAL SP2100 combine harvester. The production obtained on each experimental plot, the humidity of the grains at harvesting and the hectolitric mass were determined automatically with the equipment installed on the combine.

The mass of a thousand grains was calculated using a photocell device for counting, along with a precision balance.

Along with these determinations, measurements were made on the height of the cob's insertion and the height of the plant. The statistical interpretation was made by variance analysis.

RESULTS AND DISCUSSIONS

The climatic conditions of the experimental years, 2017 and 2018, in comparison with the multiannual average, are presented in Figures 1 and 2.

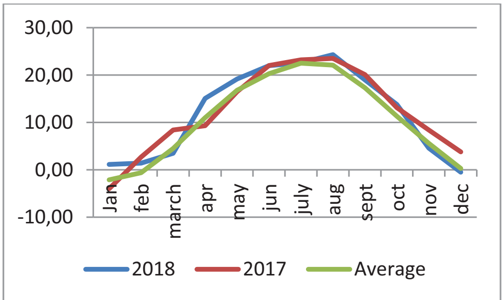

Figure 1. The temperature of 2017 and 2018 years in comparison with the multiannual average, at ARDS Marculesti

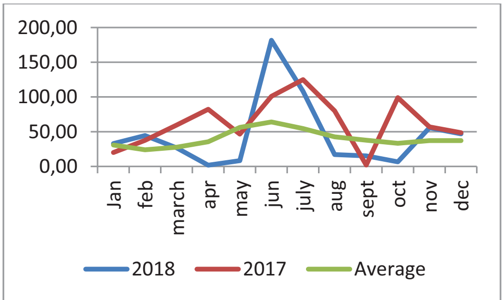

Figure 2. The rainfall of 2017 and 2018 years, in comparison with multiannual average, at ARDS Marculesti

As far as rainfall is concerned, we can see from this data that the year 2017 recorded a very rainy period in April, June, July and August, the quantities registered being more than double than the monthly multiannual average. The highest values were registered in June and July, over 100 and 120 mm respectively. September was very dry, with only 4 mm of precipitation, but this did not affect the maize vegetation.

Due to the rainfall recorded during this year's vegetation period, we can consider very favorable conditions for the growth and development of maize, as evidenced by the yields obtained.

In terms of air temperature, in 2017 there were values close to the multiannual average of the area.

The year 2018 recorded a very droughty period in the spring, in April and May, when extremely small rainfall was recorded, only 10 mm in two months. This has greatly affected germination and growth in the early stages of maize growth. The situation changed radically in the summer months, June and July, when record rainfall was 180 mm in June and 108 mm in July. The extremely abundant precipitation of this period favored the growth and development of maize plants under optimal conditions. August and September were droughty but this did not affect the production of maize.

The temperatures recorded in 2018 were generally lower than the multiannual average during June and July, due to the extremely abundant rainfall recorded. These climatic conditions have led to a very good viability and fertility of maize pollen.

a. The influence of hybrid and irrigation regime on the number of rows per cob.

The first table comprises the number of rows per cob in 2017. From this data it can be seen that with the irrigated regime the largest number of rows per cob was recorded at KWS Bellavista and KWS Smaragd hybrids and the smallest, with the hybrid KWS Durango (Table1).

With the case of irrigation before flowering, the largest number of rows of kernels on the cob was recorded with the hybrids KWS Bellavista and KWS Smaragd, too, with positive significant differences for hybrid KWS

Bellavista and significantly positive for hybrid KWS Smaragd, as compared to hybrid P9175 taken as control (Table 1).

When irrigation was done after flowering, the KWS Bellavista hybrid recorded 18.67 rows per ear and KWS Durango hybrid produced 15.33 rows per ear.

With the irrigation throughout the vegetation period, the best results were also obtained at the KWS Bellavista hybrid, with a difference of two more lines, distinctly significant, compared to the P9175 control (Table 1).

As regard the differences between hybrids at the average of irrigation regimes, the KWS Bellavista hybrid showed a very significant positive difference from the control, the P9175 hybrid and the KWS Durango hybrid recorded a significantly negative difference compared to the same control variant (Table 1).

As concerned on the irrigation regime (Table 1), the stressed before and after flowering have given positive significant difference over the unirrigated treatment, taken as control as well as over the irrigated treatment.

Table 1. The number of rows of grains per cob in function of maize hybrid and irrigation regime in 2017 at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3 – stressed before flowering			
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign
a1-P9175	16.00	100	Mt	-	16.00	100	Mt	-	16.00	100	Mt	-
a2-KWS Bellavista	17.33	108.3	1.33	*	20.00	125	4.00	***	18.67	116.6	2.67	***
a3-KWS Smaragd	15.33	95.8	-0.6	-	15.33	95.8	-0.67	-	16.00	100	0.00	-
a4-KWS Kashmir	17.33	108.3	1.33	*	17.33	108.3	1.33	*	18.00	112.5	2.00	**
a5-KWS Durango	14.67	91.6	-1.3	0	15.33	95.8	-0.67	-	15.33	95.8	-0.6	-
LSD5%=1.21; LSD 1%=1.66; LSD 0.1%=2.25												

Hybrid	b4 –full time irrigated				Average of irrigation regimes				Irr. Regime	Number of rows	%	Diff.	Sign
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign					
a1-P9175	16.00	100	Mt	-	16	100	Mt	-	b1–unirrigated	16,13	100	Ctrl	-
a2-KWS Bellavista	18.00	112.5	2.00	**	18.5	115.6	2.5	***					
a3-KWS Smaragd	16.00	100	0.00	-	15.6	97.9	-0.33	-	b2 – stressed during and after flowering	16,80	103	0,67	*
a4-KWS Kashmir	17.33	108.3	1.33	*	17.5	109.3	1.5	**	b3 – stressed before flowering	16,80	103	0,67	*
a5-KWS Durango	14.67	91.6	-1.33	0	15	93.7	-1	0	b4 –full time irrigated	16,40	101	0,27	-
LSD 5%=1.21; LSD 1%=1.66; LSD 0.1%=2.25					LSD 5%=0.82; LSD 1%=1.20; LSD 0.1%=1.80				LSD 5%=0.64; LSD 1%=0.87; LSD 0.1%=1.15				

The feature of the number of rows on the cob, in 2018, is shown in (Table 2).

This year, the smallest number of rows on the maize cob was recorded with hybrid P9175, i.e., 15.6 rows. The differences between KWS Bellavista and KWS Smaragd hybrids compared to P9175 were significantly positive and those between the KWS Kashmir and KWS Durango hybrids to the control were insignificant (Table 2).

Regarding the irrigation regimes, the recorded data does not lead to a clear conclusion on the influence of the experimental treatments, probably due to the extreme rainfall quantities, too little at the beginning of the vegetation and too high during the growing and fructification period. Thus, the P9175 hybrid recorded the same number of rows per year on all irrigation regimens. With the KWS Bellavista hybrid, the best results were given by irrigation before flowering, and with the KWS Smaragd hybrid, the highest number of rows on the cob were counted with the irrigation after flowering treatment. Overall, unirrigated treatment gave the lowest results (Table 2).

The irrigation on the whole vegetation period has given the best results on the number of rows per cob in 2018. Nevertheless, the irrigation after flowering (stressed before flowering) has recorded no significance over the unirrigated control because during that period there were recorded lots of rainfall and the irrigation regime has had less influence on the formation of the rows on the cob (Table 2)

Table 2. The number of rows of grains per cob in function of maize hybrid and irrigation regime in 2018 at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3 – stressed before flowering			
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign
a1-P9175	15.33	100	Mt	-	16	100	Mt	-	15.33	100	Mt	-
a2-KWS Bellavista	17.33	113.0	2.00	**	19.33	120.8	3.33	***	18.66	121.7	3.33	***
a3-KWS Smaragd	16	104.3	0.67	-	16.66	104.1	0.67	-	16	104.3	0.67	-
a4-KWS Kashmir	17.33	113.0	2.00	**	18	112.5	2.00	**	16.66	108.6	1.33	*
a5-KWS Durango	16	104.3	0.67	-	16.66	104.1	0.67		15.33	100	0.00	-
LSD 5%=1.21; LSD 1%=1.65; LSD 0,1%=2.25												

Hybrid	b4 –full time irrigated				Average of irrigation regimes			
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign
a1-P9175	16	100	Mt	-	15.66	100	Mt	-
a2-KWS Bellavista	20	125	4.00	***	18.83	120.2	3.16	***
a3-KWS Smaragd	16.66	104.16	0.67	-	16.33	104.2	0.66	-
a4-KWS Kashmir	18	112.5	2.00	**	17.5	111.7	1.83	***
a5-KWS Durango	16.66	104.16	0.67	-	16.16	103.1	0.5	-
LSD 5%=1.21; LSD 1%=1.65; LSD 0.1%=2.25					LSD 5%=0.77; LSD 1%=1.13; LSD 0.1%=1.70			

Irr. Regime	Number of rows	%	Diff.	Sign
b1–unirrigated	16,4	100	Ctrl	-
b2 – stressed during and after flowering	17,3	105	0,9	*
b3 – stressed before flowering	16,4	100	-	-
b4 –full time irrigated	17,4	106	1,0	**
LSD 5%=0.68; LSD 1%=0.92; LSD 0.1%=1.22				

The number of rows per cob as average of the two experimental years is presented in the (Table 3).

The average values of the two years of trials show very and distinct significant differences between hybrids KWS Bellavista and KWS Smaragd over P9175 hybrid taken as control (Table 3). There, also, can be seen that full irrigated and stressed after flowering gave higher values of the number of rows per cob (Table 3).

Table 3. The number of rows of grains per cob in function of maize hybrid and irrigation regime as average of 2017 and 2018 years, at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3 – stressed before flowering			
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign
a1-P9175	15.6	100	Mt	-	16	100	Mt	-	15.6	100	Mt	-
a2-KWS Bellavista	17.3	110.6	1.67	**	19,6	122.9	3.67	***	18.6	119.1	3.00	***
a3-KWS Smaragd	17.3	110.6	1.67	**	17,6	110.4	1.67	**	17.3	110.6	1.67	**
a4-KWS Kashmir	15.6	100	0.00	-	16	100	0.00	-	16	102.1	0.33	-
a5-KWS Durango	15.3	97.8	-0.3	-	16	100	0.00	-	15.3	97.8	-0.3	-
LSD 5%=1.21; LSD 1%=1.66; LSD 0.1%=2.25												

Hybrid	b4 –full time irrigated				Average of irrigation regimes			
	Number of rows	%	Diff.	Sign	Number of rows	%	Diff.	Sign
a1-P9175	16	100	Mt	-	15.8	100	Mt	-
a2-KWS Bellavista	19	118.7	3,00	***	18.6	117.8	2.8	***
a3-KWS Smaragd	17.6	110.4	1,67	**	17.5	110.5	1.6	**
a4-KWS Kashmir	16.3	102.0	0,33	-	16	101.0	0.1	-
a5-KWS Durango	15.6	97.9	-0,33	-	15.5	98.4	-0.2	-
LSD 5%=1.21; LSD 1%=1.66; LSD 0.1%=2.25					LSD 5%=0.79; LSD 1%=1.16; LSD 0.1%=1.75			

The number of grains per row, in 2017 and the statistical interpretation are shown in table 4. Regarding the number of grains per row, in the year 2017, the P9175 hybrid, which is taken as control, is very clear emphasized. Thus, in the unirrigated condition, the KWS Bellavista hybrid recorded very significant negative differences compared to the control P9175 and the other 3 tried hybrids (KWS Kashmir, KWS Smaragd and KWS Durango) recorded distinctly significant differences. These differences were maintained in the other irrigation regimes but more mildly (Table 4).

Regarding the irrigation regimes, the worst results were obtained with rainfed treatment, where there were 30.9 grains in a row and the highest number of grains was recorded in full time irrigated treatment, i.e. 35.8 grains in a row. The difference between these two irrigation regimes was very significant. All three irrigation treatments gave very significant positive differences as compared with the unirrigated control treatment (Table 4)

Table 4. The number of grains in a row in function of maize hybrid and irrigation regime, in 2017, at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3			
	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign
a1-P9175	33.00	100	Mt	-	35.33	100	Mt	-	36.67	100	Mt	-
a2-KWS Bellavista	29.67	89.8	-3.3	000	31.00	87.7	-4.33	000	32.33	88.1	-4.3	000
a3-KWS Smaragd	30.67	92.9	-2.3	00	34.67	98.1	-0.67	-	35.33	96.3	-1.3	-
a4-KWS Kashmir	30.67	92.9	-2.3	00	35.00	99.0	-0.33	-	35.33	96.3	-1.3	-
a5-KWS Durango	30.67	92.9	-2.3	00	32.33	91.5	-3.00	000	35.67	97.2	-1.0	-
LSD 5%=1.34; LSD 1%=1.83; LSD 0.1%=2.49												

Hybrid	b4 –full time irrigated				Average of irrigation regimes				Irr. Regime	Number of rows	%	Diff.	Sign
	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign					
a1-P9175	37.33	100	Mt	-	35.53	100	Mt	-	b1–unirrigated	30,93	100	Ctrl	-
a2-KWS Bellavista	33.00	88.3	-4.33	000	31.5	88.52	-4.08	000					
a3-KWS Smaragd	36.00	96.4	-1.33	-	34.16	96.01	-1.41	00	b2 – stressed during and after flowering	33,66	108	2,73	***
a4-KWS Kashmir	36.67	98.2	-0.67	-	34.41	96.72	-1.16	00	b3 – stressed before flowering	35,06	113	4,13	***
a5-KWS Durango	36.00	96.4	-1.33	-	33.66	94.61	-1.91	000	b4 –full time irrigated	35,80	115	4,87	***
LSD 5%=1.34; LSD 1%=1.83; LSD 0.1%=2.49					LSD 5%=0.76; LSD 1%=1.10; LSD 0.1%=1.66				LSD 5%=1,01; LSD 1%=1,36; LSD 0.1%=1.80				

The number of grains per row in 2018 is shown in (Table 5). In 2018, the highest number of grains were recorded by the hybrids KWS Smaragd and KWS Durango, but the value recorded by these two hybrids is very close to that recorded by hybrid P9175 (37.6 vs. 37.1). The lowest values of this character were recorded by the KWS Bellavista hybrid, with 33.3 grains per row, the difference from the P9175 being very significant negative (Table 5).

Irrigation regimes have influenced the number of grains in a row, from unirrigated to irrigated throughout the vegetation period. The treatments: irrigated before and after flowering yielded intermediate results. However, hybrids P9175 and KWS Bellavista recorded very significant positive differences between the unirrigated variant, taken as a control, and the one irrigated during the entire period of maize vegetation (Table 5).

In general, the influence of the irrigation regime in 2018 year was not concludent because of high rainfall which were recorded during the vegetation period of maize (Table 5).

Table 5. The number of grains in a row in function of maize hybrid and irrigation regime, in 2018, at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3 – stressed before flowering			
	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign
a1-P9175	36.66	100	Mt	-	37.33	100	Mt	-	36.66	100	Mt	-
a2-KWS Bellavista	32.67	89.0	-4.00	000	34.00	91.07	-3.33	000	32.67	89.0	-4.00	000
a3-KWS Smaragd	35.33	96.3	-1.33	-	36.66	98.2	-0.67	-	35.33	96.3	-1.33	-
a4-KWS Kashmir	36.66	100	0.00	-	38.66	103.5	1.33	-	36.66	100	0.00	-
a5-KWS Durango	36.67	100	0.00	-	38.00	101.7	0.67	-	37.33	101.8	0.67	-
LSD 5%=1.62; LSD 1%=2.22; LSD 0.1%=3.01												

Hybrid	b4 –full time irrigated				Average of irrigation regimes				Irr. Regime	Number of rows	%	Diff.	Sign
	Number of grains in a row	%	Diff.	Sign	Number of grains in a row	%	Diff.	Sign					
a1-P9175	38	100	Mt	-	37.16	100	Mt	-	b1–unirrigated	35,6	100	Ctrl	-
a2-KWS Bellavista	34.00	89.4	-4.00	000	33.33	89.6	-3.83	000					
a3-KWS Smaragd	37.33	98.2	-0.67	-	36.16	97.3	-1	-	b2 – stressed during and after flowering	36,9	103	1,3	**
a4-KWS Kashmir	38.66	101.7	0.67	-	37.66	101.3	0.5	-	b3 – stressed before flowering	35,7	100	0,1	-
a5-KWS Durango	38.67	101.7	0.67	-	37.66	101.3	0.5	-	b4 –full time irrigated	37,3	104	1,7	***
LSD 5%=1.62; LSD 1%=2.22; LSD 0.1%=3.01					LSD 5%=1.73;LSD 1%=2.51; LSD 0.1%=3.77				LSD 5%=0,93; LSD 1%=1,26; LSD 0.1%=1.67				

The average values of the two years of trials on the number of grains in a row in function on the hybrid and the irrigation regime are presented in the (Table 6). These values show in very clear way that the KWS Bellavista hybrid recorded very significant negative differences over the control P9175 (Table 6). This fact can be explained by shorter period of vegetation of this hybrid (FAO 330)

Table 6. The number of grains in a row in function of maize hybrid and irrigation regime (average of 2017 and 2018), at ARDS Marculesti

Hybrid	b1 –unirrigated				b2 – stressed during and after flowering				b3 – stressed before flowering			
	Number of grains per row	%	Diff.	Sign	Number of grains per row	%	Diff.	Sign	Number of grains per row	%	Diff.	Sign
a1-P9175	34.8	100	Mt		36.3	100	Mt		36.6	100	Mt	
a2-KWS Bellavista	31.1	89.4	-3.6	000	32.5	89.4	-3,83	000	32.5	88.6	-4.1	000
a3-KWS Smaragd	33.6	96.6	-1.1	-	36.8	101.3	0,50	-	36	98.1	-0.6	-
a4-KWS Kashmir	33	94.7	-1.8	0	35.6	98.1	-0.67	-	35.3	96.3	-1.3	-
a5-KWS Durango	33.6	96.6	-1.1	-	35.1	96.7	-1.17	-	36.5	99.5	-0.1	-
LSD 5%=1.48; LSD 1%=2.02; LSD 0.1%=2.75												

Hybrid	b4 –full time irrigated				Average of irrigation regimes			
	Number of grains per row	%	Diff.	Sign	Number of grains per row	%	Diff.	Sign
a1-P9175	37.6	100	Mt	-	36.3	100	Mt	-
a2-KWS Bellavista	33.5	88.9	-4.17	000	32.4	89.1	-3.9	000
a3-KWS Smaragd	37.6	100	0.00	-	36.0	99.0	-0.3	-
a4-KWS Kashmir	36.6	97.3	-1.00	-	35.1	96.6	-1.2	0
a5-KWS Durango	37.3	99.1	-0.33	-	35.6	98.0	-0.7	-
LSD 5%=1.48; LSD 1%=2.02; LSD 0.1%=2,75					LSD5%=0.74; LSD 1%=1.80; LSD 0.1%=2.72			

CONCLUSIONS

Maize is a plant that responds positively to irrigation. The water consumption of the maize plant differs according to the phenophase in which it is and it can influences the final production, no matter when it occurs.

There were very significant differences in both the number of rows per cob and the number of grains per row, depending on the moment of the application of irrigation as well as the time of precipitation (19 vs. 16 rows and 33 vs. 37 grains per row)

If the water deficit was between the emergence and the blossom time, in the 5-6 leaf stage, there was recorded a decrease in the number of rows per cob because this is the time to differentiate the primordia of the maize cob and the number of fertile flowers on the ears.

The second situation when the water deficit was after flowering shows that the number of grains, in turn, decreased significantly in treatments with a lower water intake.

The vegetation period of the investigated hybrids significantly influenced both the number of rows on the ears and the number of grains per row in the sense that with tardy hybrids these values were higher.

As regard the number of rows of kernels on the cob the best results were given by the hybrids KWS Bellavista (18.6) and KWS Smaragd (17.5), with distinct and very significant positive differences over the P 9175 hybrid (15.8) which was taken as control.

In return, the KWS Bellavista hybrid recorded the lowest results about the number of grains in a row (32.4) with very significant negative difference over the control, P 9175 hybrid (36.3).

REFERENCES

Jonghan, K., Piccinni, G. (2009). Maize yield responses under crop evapotranspiration-based irrigation management. *Agricultural Water Management, 209*, 65–84.

Lizaso, J.I., Ruiz-Ramos, M., Rodríguez, L., Gabaldon-Leal, C. (2018). Impact of high temperatures in maize: Phenology and yield components. *Field Crops Research, 216*, 129–140.

Năescu, V., Alionte, E. (2008). Importanţa irigării, la principalele culturi de câmp, în condiţiile unui an cu secetă accentuată. *Analele INCDA Fundulea, LXXVI.*

NeSmith, D.S., Ritchie, J.T. (1992). Effects of soil water-deficits during tassel emergence on development and yield component of maize (*Zea mays*). *Field Crops Research, 28*(3), 251–256.

Niaz A., and Rameshwar S.K. (1991). Effect of Different Moisture Stress Levels on Maize Growth in Field Lysimeters. *Agricultural and biosystems engineering publications,* Iowa State University.

Oşvat, I.M., (2015). Water use in maize crop on sloping soils from N-W region of Romania. *Fundamental Economic Research in the Field of Agriculture, Food Safety and Security. Timisoara, Vol. 2*.

Schoper, J., Robert, B., Lambert, J., Bruce, L., Vasilas, M., Westgate, E. (1987). The Influence of Silk, Pollen, and Ear-Leaf Water Status and Tassel Heat Treatment at Pollination. *Plant physiology*, *8*, 83–121.

Setter Tim, L., Brian, A. F., Jeff, M. (2001). Loss of Kernel Set Due to Water Deficit and Shade in Maize. *Crop Science*, *41*(5), 1530–1540.

Stegman, E.C. (1982). Maize grain yield as influenced by timing of evapotranspiration deficits. *Irrigation Science, 3*(2), 75–87.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

HERBICIDE CONTROL OF THE WEEDS IN MAIZE (*Zea mays* L.)

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Abstract

During the period of 2017-2018 a field study with maize (Zea mays L.) was conducted. The experiment was situated on the experimental field of the Agricultural University of Plovdiv, Bulgaria. Variants of the trial were as follows: 1. Untreated control; 2. Equip OD (75 g/l mesotrione + 30 g/l nicosulfuron) - 2500 ml/ha; 3. Elumis OD (75 g/l mesotrione + 30 g/l nicosulfuron) - 2000 ml/ha; 4. Arigo WG (360 g/kg mesotrione + 120 g/kg nicosulfuron + 30 g/kg rimsulfuron) + Trend 90 - 330 g/ha + 1000 ml/ha; 5. Samson Extra 6 OD (60 g/l nicosulfuron) - 750 ml/ha; 6. Samson 4 OD (40 g/l nicosulfuron) - 1250 ml/ha; 7. Principal Plus WG (552 g/kg dicamba + 92 g/kg nicosulfuron + 23 g/kg rimsulfuron) + Trend 90 - 440 g/ha + 1000 ml/ha; 8. Capreno SC (345 g/l tembotrione + 68 g/l thiencarbazone-methyl + 134 g/l isoxadifen-ethyl) + Mero - 290 ml/ha + 2000 ml/ha. The grown maize hybrid was "P 9241". The efficacy of the studied herbicide products against the weeds was evaluated. The highest yield was achieved after the application of Principal Plus WG – 1.53 t/ha average for both years.

Key words*: maize, herbicides, efficacy, yields.*

INTRODUCTION

Maize (*Zea mays* L.) is main grain-forage crop with adaptive ability to different geographical and climatic conditions. That is the reason for the successful growing of this culture in many regions around the globe. In Bulgaria it is strategical field crop. Maize has the highest energy value in comparison to the others forage crops (Tomov & Yordanov, 1984).

One of the main negative factors for agricultural production is the weeds. They decrease the yields and the quality of maize grain (Spasov, 1995; Masqood et al., 1999; Tonev, 2000; Changsaluk et al., 2007).

In Bulgaria, economically the most important weeds at this crop are *Amaranthus retroflexus* L., *Datura stramonium* L., *Xanthium strumarium* L., *Solanum nigrum* L., *Chenopodium album* L., *Abutilon theophrasti* L., *Sinapis arvensis* L., *Echinochloa crus-gali* L., *Setaria glauca* L., *Sorghum halepense* L., *Convolvulus arvensis* L., *Cynodon dactilon* L. and *Cirsium arvense* L. (Hristova et al., 2012; Kalinova et al., 2012).

The maize grain yield can decrease from 24% to 96.7% (Mukherjee and Debnath, 2013; Oerke & Dehne, 2004; Zhalnov & Raikov, 1996).

The monoculture growing of maize can lead to increase of the population of *S. halepense, C. arvense*, *C. arvensis* L., *C. dactilon* and other perennial weed species.

The most efficient and economically most effective and environmentally safest is integrated weed control. It includes applicayion of different weed control means - mechanical, chemical, cultural, biological etc. (Tonev, 2013).

The chemical method is the most often used by the farmers. The method is highly effective, fast and easy to apply. The proper herbicide application reduces the weed management costs up to 60%. The fuel cost as well the soil erosion are also decreased (Valcheva, 2011).

The aim of the study is to evaluate the possibilities for efficient chemical weed control in maize.

MATERIALS AND METHODS

During the 2017 and 2017 a field experiment was carried out in the experimental base of the Department of Agriculture and Herbology of the Agricultural University of Plovdiv, Bulgaria. The studied maize (*Zea mays* L.) hybrid was "P 9241". The trial was conducted by the randomized block design in 4 replications. The size of the harvesting plot was 28 m^2.

The variants of the trial were: 1. Untreated control; 2. Equip OD (75 g/l mesotrione + 30 g/l nicosulfuron) - 2500 ml/ha; 3. Elumis OD (75 g/l mesotrione + 30 g/l nicosulfuron) - 2000 ml/ha; 4. Arigo WG (360 g/kg mesotrione + 120 g/kg nicosulfuron + 30 g/kg rimsulfuron) + Trend 90 (adjuvant) - 330 g/ha + 1000 ml/ha; 5. Samson Extra 6 OD (60 g/l nicosulfuron) - 750 ml/ha; 6. Samson 4 OD (40 g/l nicosulfuron) - 1250 ml/ha; 7. Principal Plus WG (552 g/kg dicamba + 92 g/kg nicosulfuron + 23 g/kg rimsulfuron) + Trend 90 - 440 g/ha + 1000 ml/ha; 8. Capreno SK (345 g/l tembotrione + 68 g/l thiencarbazone-methyl + 134 g/l isoxadifen-ethyl) + Mero (adjuvant) - 290 ml/ha + 2000 ml/ha.

The herbicide products were applied in BBCH 14-15. The volume of the spraying solution was 250 l/ha.

The efficacy of the studied herbicides against the weeds was performed by the 10 score scale of EWRS (European Weed Research Society) on the 14th, 28th and on the 56th day after application.

The selectivity of the herbicides was evaluated by the 9 score scale of EWRS.

The yield was recorded by harvesting the whole experimental plot of every repetition from each treatment.

Statistical analysis of collected data was performed by using Duncan's multiple range test of SPSS 17 program. Statistical differences were considered significant at $p < 0.05$.

RESULTS AND DISCUSSIONS

The existing weeds on the experaimental field were *Setaria viridis* L., *Echinochloa crus-gali* L., *Sorghum halepense* Pers. developed from seeds, *Chenopodium album* L., *Amaranthus retroflexus* L., *Xanthium strumarium* L., *Abutilon theophrasti* Medic., *Datura stramonium* L., *Solanum nigrum* L., *Portulaca oleracea* L., *S. halepense* developed from rhizomes and *Cynodon dactylon* L.

On Table 1 is shown the dynamics considering the efficacy of the studied herbicide products against *S. viridis*. In both experimental years, on the 14th day after the herbicide application the efficacy is not satisfactory, but the weed is depressed, growth retardation is also observed. On the next evaluation dates very high efficacy is reported. The efficacy ranges from 85 to 100% on the 56th day after treatments.

Table 1. Efficacy of the studied herbicide products against *S. viridis,* average for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	75	85	90
3. Elumis OD	75	85	90
4. Arigo WG + Trend 90	85	95	95
5. Samson Extra 6 OD	85	100	100
6. Samson 4 OD	85	95	100
7. Principal Plus WG +Trend 90	80	90	100
8. Capreno SK + Mero	60	75	85

The efficacy against *E. crus-gali* is on Table 2. The data from the reports showed efficacy percentage close to those of *S. viridis*.

On the first reporting date the results are low and increasing in the next evaluations. The lowest efficacy is recorded to be for treatment 8 (Capreno SK + Mero - 290 ml/ha + 2000 ml/ha).

In previous study Mitkov et al. (2018) observed the highest efficacy against *Echinochloa crus-galli* L. for Merlin Duo at rate of 2.00 l/ha.

Table 2. Efficacy of the studied herbicide products against *E. crus-gali,* average for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	80	90	95
3. Elumis OD	80	90	95
4. Arigo WG + Trend 90	85	95	95
5. Samson Extra 6 OD	85	95	100
6. Samson 4 OD	80	90	100
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	60	75	85

The efficacy against *S. halepense* developed from seeds is on Table 3.

All studied herbicide products sucseffully control *S. halepense* Pers. developed from seeds. On the last evaluation date, the weed is 100% controlled independently the herbicide product.

Table 3. Efficacy of the studied herbicide products against *S. halepense* developed from seeds for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	90	100	100
3. Elumis OD	80	90	100
4. Arigo WG + Trend 90	85	95	100
5. Samson Extra 6 OD	85	100	100
6. Samson 4 OD	80	95	100
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	80	90	100

The efficacy data against *Ch. album* for 2017-2018 is shown on table 4.

Against this weed the efficacy of Arigo WG + Trend 90 and Principal Plus WG + Trend 90 was excellent - 100% on the 3rd evaluation date 56 days after the herbicide treatments. The efficacy of Equip OD, Elumis OD and Capreno SK + Mero is lower and raging from 85 to 90%. Unsatisfactory efficacy from the treatments with Samson 4 OD and Samson Extra 6 OD was recorded - fromт 35 to 40%. If there is high infestation with *Chenopodium album* L. tank mixture of Mustang® 306.25 SC + Nishin® 4 OD at rates of 600 ml/ha + 1300 ml/ha can be successfully applied (Tonev et al., 2016).

Table 4. Efficacy of the studied herbicide products against *Ch. album* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	70	85	85
3. Elumis OD	70	80	90
4. Arigo WG + Trend 90	80	90	100
5. Samson Extra 6 OD	80	60	40
6. Samson 4 OD	75	55	35
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	80	90	90

Independently the applied herbicide product, on the last reporting date the efficacy reached 100% for all treatments (Table 5). It was observed for bot experimental years.

Damalas et al. (2018) report excellent efficacy against *A. retroflexus* after treatment with herbicide mixtures based on tembotrione + rimsulfuron, nicosulfuron or foramsulfuron (label rates for weed control in maize).

Zhao et al. (2017) observed excellent efficacy against *A. retroflexus* after application of Isoxaflutole, Mesotrione and Isoxaflutole + acetochlor in both experimental years of their research.

Table 5. Efficacy of the studied herbicide products against *A. retroflexus* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	85	95	100
3. Elumis OD	80	90	100
4. Arigo WG + Trend 90	85	95	100
5. Samson Extra 6 OD	80	90	100
6. Samson 4 OD	75	95	100
7. Principal Plus WG +Trend 90	90	95	100
8. Capreno SK + Mero	80	90	100

From all annual dicotyledonous weeds present in the study, *Xa. strumarium* was the most difficult to control. The obtained results are shown on table 6.

For none of the studied herbicide products, the efficacy reached satisfactory results. The efficacy of Equip OD, Principal Plus WG + Trend 90 and Capreno SK + Mero the average efficacy for both trial years was fom 85 to 90%. On the first evaluation date the efficacy of Arigo WG + Trend 90 was 70%, but the weed recovered to some extend and on the last reporting date the efficacy decreased to 50%. From the rest three herbicides the efficacy was 0% on last reporting date.

Saflufenacil and saflufenacil + dimethenamid-p applied pre-emergence and dicamba, dicamba + diflufenzopyr, dicamba + atrazine and mesotrione + atrazine applied post-emergence had the potential to provide from good to excellent control of common cocklebur in corn (Soltani et al., 2010).

Table 6. Efficacy of the studied herbicide products against *Xa. strumarium* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	70	80	85
3. Elumis OD	40	10	0
4. Arigo WG + Trend 90	70	60	50
5. Samson Extra 6 OD	20	5	0
6. Samson 4 OD	15	5	0
7. Principal Plus WG +Trend 90	70	85	90
8. Capreno SK + Mero	75	85	90

On Table 7 is shown that the average efficacy of Equip OD, Principal Plus WG + Trend 90 and Capreno SK + Mero for 2017-2018 was 100% on the last reporting date.
The efficacy of the other treatments was with very low differences and also was excellent – from 90 to 95%.
The weed *Abuthilon theophrasti* L. can also be controlled by application of Merlin® Duo (Mitkov et al., 2018)

Table 7. Efficacy of the studied herbicide products against *A. theophrasti* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	80	90	100
3. Elumis OD	80	90	95
4. Arigo WG + Trend 90	75	90	95
5. Samson Extra 6 OD	75	90	90
6. Samson 4 OD	75	85	90
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	80	95	100

After the application of Equip OD, Arigo WG + Trend 90, Principal Plus WG + Trend 90 and Capreno SK + Mero the recorded efficacy against *D. stramonium* on the 56th day after treatments reached 100%.
The efficacy Elumis OD, Samson Extra 6 OD and Samson 4 OD was also excellent reaching from 90 to 95% (Table 8).
The application of nicosulfuron + dicamba + bentazon; nicosulfuron + dicamba + bentazon; nicosulfuron + tritosulfuron + icamba; nicosulfuron + mesotrione; nicosulfuron + mesotrione + atrazine; etc. can have very good results for controlling *D. stramonium* (Torma et al., 2006).

Table 8. Efficacy of the studied herbicide products against *D. stramonium* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	85	95	100
3. Elumis OD	85	95	95
4. Arigo WG + Trend 90	80	95	100
5. Samson Extra 6 OD	75	85	90
6. Samson 4 OD	75	85	90
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	80	95	100

All herbicide products successfully control the weed *S. nigrum* from the first evaluation date to the 56th after the treatments average for both trial years (Table 9).
According to Pannacci and Covarelli (2009), in order to obtain 95% of efficacy against *S. nigrum*, mesotrione could be used at 1/6 of the maximum labelled dose (150 g a.i./ ha).
According to Mitkov et al. (2018) *Solanum nigrum* can be also controlled successfully with Merlin® Duo, Adengo® 465 SC and Lumax® 538 SC.

Table 9. Efficacy of the studied herbicide products against *S. nigrum* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	90	100	100
3. Elumis OD	85	95	100
4. Arigo WG + Trend 90	90	95	100
5. Samson Extra 6 OD	80	90	100
6. Samson 4 OD	80	95	100
7. Principal Plus WG +Trend 90	95	100	100
8. Capreno SK + Mero	85	95	100

The efficacy results against the weed *P. oleracea* average for the period are shown on Table 10. Dogan et al. (2005) reported 90% efficacy against *Portulaca oleracea* when nicosulfuron (Samson) and 2.4 D amine salt (Di Amin) was applied.
In our trial all evaluated herbicide products achieved excellent control of this weed average for both years. The efficacy was very high from the 14th day after application till the last reporting date.

Table 10. Efficacy of the studied herbicide products against *P. oleracea* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	90	95	100
3. Elumis OD	85	95	100
4. Arigo WG + Trend 90	80	90	100
5. Samson Extra 6 OD	85	95	100
6. Samson 4 OD	80	95	100
7. Principal Plus WG +Trend 90	85	95	100
8. Capreno SK + Mero	80	95	100

The efficacy against the *S. halepense* developed from rhizomes is shown Table 11.

None of the evaluated herbicides showed 100% efficacy against this weed average for the period of the experiment.

The highest efficacy is achieved after the application of Equip OD starting from 70% on the 14th day after the herbicide application and reaching 95% on the 56th day. The efficacy of nicosulfuron-containing herbicide products - Samson Extra 6 OD, Samson 4 OD, Principal Plus WG and Arigo WG was lower but good. Very poor efficacy was reported for the products Elumis OD Capreno SK – 40% on the 56th after treatments.

In field experiments were carried out in Greece from by Eleftherohorinos and Kotoula-Syka (1995) the authors also observed satisfactory efficacy against *S. halepense* from rhizomes after application of nicosulfuron.

No efficacy (0%) was found against *S. halepense* after the application of Merlin® Duo, Adengo® 465 SC and Lumax® 538 SC (Mitkov et al., 2018).

Table 11. Efficacy of the studied herbicide products against *S. halepense* developed from rhizomes for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	70	85	95
3. Elumis OD	20	30	40
4. Arigo WG + Trend 90	65	75	85
5. Samson Extra 6 OD	65	85	90
6. Samson 4 OD	60	80	90
7. Principal Plus WG +Trend 90	65	80	85
8. Capreno SK + Mero	20	30	40

On the three evaluation dates the efficacy against *C. dactylon* was 0% independently the studied herbicide product (Table 12).

Table 12. Efficacy of the studied herbicide products against *C. dactylon* for 2017-2018, %

Treatments	Days after application		
	14th	28th	56th
1. Untreated control	-	-	-
2. Equip OD	0	0	0
3. Elumis OD	0	0	0
4. Arigo WG + Trend 90	0	0	0
5. Samson Extra 6 OD	0	0	0
6. Samson 4 OD	0	0	0
7. Principal Plus WG +Trend 90	0	0	0
8. Capreno SK + Mero	0	0	0

No visible signs of phytotoxicity were reported for any of the treatments.

The weeds decrease the yields and the quality of maize grain (Masqood et al., 1999). The results of the comparative analysis of the yield per hectare showed that during the two years of the study, significant differences in the benefit of the individual treated variants compared to the untreated control were demonstrated (Figure 1).

The highest grain yield was recorded for the treatment with Principal Plus WG + Trend 90 – 1.53 t/ha average for both years of the research. The result was with proven difference according to Duncan's multiple range test ($p < 0.05$). The yield from the variants treated with Equip OD, Arigo WG + Trend 90 and Capreno SK + Mero were with lower yields - 1.02, 0.99 and 0.95 t/ha respectively. The treatments with Elumis OD, Samson Extra 6 OD and Samson 4 OD had lower yields. The lowest yield from the study was reported for the untreated control – 0.57 t/ha.

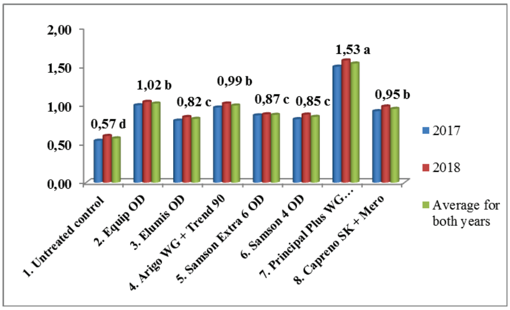

Figure 1. Maize grain yield, t/ha

Columns with different letters are with a proved difference by Duncan's multiple range test ($p < 0.05$).

CONCLUSIONS

The herbicide products Principal Plus WG, Samson Extra 6 OD and Samson 4 OD had the highest efficacy against *S. viridis* and *E. crus-galli* L.

The best herbicide effect is against *Ch. album* L. for Principal Plus WG and Arigo WG was observed.

Xa. strumarium was the most difficult to control dicotyledonous weed from the treatments with Arigo WG, Elumis OD, Samson Extra 6 OD and Samson 4 OD.

All herbicide products showed excellent efficacy against weeds *A. theophrasti* and *D. stramonium*.

The lowest herbicide efficacy against *S. halepense* developed prom rhizomes for Elumis OD and Capreno SK was reported.

The most difficult-to-control-weed present in the field was *C. dactylon* L. The efficacy against this weed was 0 % independently the studied herbicide product and evaluation date.

The highest grain yield was recorded for the treatment with Principal Plus WG + Trend 90, and the lowest yield for the untreated control was recorded.

ACKNOWLEDGEMENTS

This research work was carried out with the support of The Center for Biological Examination of Products for Plant Protection at the Agricultural University of Plovdiv, Bulgaria.

REFERENCES

Changsaluk, S., Pornprom, T., Waramitr, N., Suwanmakkha, R., Pathom, N., Lim-aroon, S. (2007). Effect of weed densities of fresh corn yield. Proceedings of the 45th Kasetsart University, Annual. Conference, Bangok, Thailand, 30 January-2 February, on CD.

Damalas, Ch., Gitsopoulos, T., Koutroubas, S., Alexoudis, Ch., Georgoulas, I. (2018). Weed control and selectivity in maize (*Zea mays* L.) with tembotrione mixtures. *International Journal of Pest Management, 64*(1), 11–18, DOI: 10.1080/09670874.2017.1293307

Dogan, M., Boz, O., Unay, A. (2005). Efficacies of Reduced Herbicide Rates for Weed Control in Maize (*Zea mays* L.) During Critical Period. *Journal of Agronomy*, *4*, 44–48. DOI: 10.3923/ja.2005.44.48.

Eleftherohorinos, I., and Kotoula-Syka, E. (1995). Influence of herbicide application rate and timings for post-emergence control of *Sorghum halepense* (L.) Pers. Maize. *Weed Research*., *35*(2), 99–103.

Hristova, S., Nankov, M., Georgiva, I., Tonev, T., Kalinova, Sht. (2012). Influence of Wild mustard (*Sinapis arvensis* L.) on the growth and productivity of maize hybrid KH-613. *Collection of Reports of the 9th Scientific and Technical Conference with International Participation „Ecology and health"*, 277–282.

Kalinova, Sht., Zhalnov, I., Yanchev, I. (2000). Influence of the combined action of Stomp 33 EK and Mistral 4 SK on the weeds in maize. *Journal of Mountain Agriculture on the Balkans, 3*(6), 705–712.

Khan, M., Marwat, K., Khan, N. (2003). Efficacy of different herbicides on the yield and yield components of maize. *Asian J. Plant Sci*., *2*(3), 300–304.

Maqsood, M., Akbar, M., Yousaf, N., Mahmood, M., Ahmed, S. (1999). Studies on weed–crop competition in maize. *International Journal of Agriculture & Biology, 4*, 270–272.

Mitkov, A., Yanev, M., Neshev, N., Tonev, T. (2018). Biological Efficacy of Some Soil Herbicides at Maize (*Zea mays* L.). *Scientific Papers. Series A. Agronomy, LXI*(1), 340–345. ISSN 2285-5785.

Mukherjee, P., and Debnath, P. (2013). Weed control practices in maize (*Zea mays* L.) under conventional and conservation tillage practices. The role of weed science in supporting food security by 2020. *Proceedings of the 24th Asian-Pacific Weed Science Society Conference, Bandung, Indonesia, October 22-25, Bandung: Weed Science Society of Indonesia*, 302–311.

Oerke, E., Dehne, H. (2004). Safequarding production–losses in major crops and the role of crop protection. *Crop Prot*., *23*, 275–285.

Pannacci, E., and Covarelli, G. (2009). Efficacy of mesotrione used at reduced doses for post-emergence weed control in maize (*Zea mays* L.). *Crop Protection*, *28*(1), 57–61.

Soltani, N., Shropshire, C., Sikkema, P. (2010). Control of common cocklebur (*Xanthium strumarium* L.) in corn. *Canadian Journal of Plant Science, 90*(6), 933–938. https://doi.org/10.4141/cjps10065

Spasov, V. (1995). Habilitation for the academic title „Professor". (In Bulgarian).

Tomov, N., and Yordanov, Yo. (1984). The maize in Bulgaria Publisher: "Zemizdat", Sofia (pp. 315) (Book in Bulgarian).

Tonev, T. (2000). *Integrated weed management and proficiency of agriculture*. Publisher "VSI Plovdiv". (pp. 221–222) (In Bulgarian).

Tonev, T., Tityanov, M., Mitkov, A., Yanev, M., Neshev, N., 2016. Control of highly blended weeding at maize (*Zea mays* L.). *Book of Proceedings, VII International Scientific Agriculture Symposium "Agrosym 2016", Jahorina*, October 06th - 09th, 1256–1262.

Tonev, T. (2013). Complexed control of the weeds. *Plant Protection*, *2*, 18.

Torma, M., Kazinczi, G., HÓDI, L. (2006). Postemergence herbicide treatments in maize against difficult to control weeds. *Hungary Journal of Plant Diseases and Protection*, *XX*, 781–786. ISSN 1861-4051.

Valcheva, A. (2011). Efficient control of the weeds in sunflower and maize. *Practical Agriculture*, *2*(4), 13.

Zhao, N., Zuo, L., Li, W., Guo, W., Liu, W., Wang, J. (2017). Greenhouse and field evaluation of isoxaflutole for weed control in maize in China. Scientific Reports 7: 12690, 1–9. DOI:10.1038/s41598-017-12696-7.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

QUALITY PARAMETERS AND GRAIN YIELDS OF SOYBEAN VARIETIES PRODUCED AT ARDS TURDA: A CHEMOMETRIC APPROACH

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Abstract

*The main purpose of this study was to determine the quality parameters and grain yields of twenty soybean varieties created at Agricultural Research and Development Station Turda (ARDS)**:** Diamant, Perla, Safir, Granat, Eugen, Onix, Felix, Darina TD, Cristina TD, Mălina TD, Carla TD, Larisa, Caro TD, Ilinca TD, Bia TD, Ada TD, Teo TD, Miruna TD, Nicola TD and Felicia TD. Field trials were carried out in the experimental field of the ARDS Turda, based on a randomized complete block design with three replications, using plots of 10 m^2. The varieties were characterized by: dry weight, ash, proteins, lipids, total carbohydrates, total carotenoids, plants' height, insertion height of basal pods, grain yield and thousand kernels' weight. Analytical determinations were performed using oven drying (for moisture), furnace ashing (for ash), Soxhlet extraction (for total lipids), Kjeldahl method (for total proteins), UV-VIS spectrophotometry (for total carotenoids); the total carbohydrate content was assessed by difference. A chemometric evaluation of the obtained results was carried out, highlighting similarities between the studied varieties and pointing up those ones with the best scores for quality parameters.*

Key words*: soybean seeds, quality, chemometry.*

INTRODUCTION

The current interest in soybean (*Glycine max* (L.) Merr.) is motivated by its high nutritional value, especially with respect to their protein and amino acids' content (Mello Filho et al., 2004; Specht et al., 1999). Soybean is a leading crop, grown worldwide as the most important source of vegetable protein known to mankind (30-49%) and oil (~20%); it is one of the cheapest and convenient sources of protein available, particularly in developing countries (Idrisa et al., 2010; Jiang et al., 2018). Soybean seeds contain proteins that include all the essential amino acids, besides valuable fatty acids (Clemente & Cahoon, 2009; Goldflus et al., 2006; Karr-Lilienthal et al., 2004; Liu, 1997; Lombardi et al, 2013).
The soybean breeding program from the Agricultural Research and Development Station (ARDS) Turda had as a priority the creation of early, productive varieties with good yield stability, well adapted to the pedo-climatic conditions of the area. Particular attention was paid to increased resistance to the main stress factors, increased availability to mechanized harvesting and quality (Mureşanu & Mărginean 2011; Mureşanu & Mărginean 2011a).
The results of the works carried out within the Soybean Breeding Program at ARDS Turda have been materialized by recording 20 early and very early soybean varieties during 1987-2017; most of the varieties were created and registered after 2000, 13 out of them being registered in the last 7 years (Mureşanu et al, 2017). Seventeen of these are registered in The Romanian Official Catalogue of Crop Plant Varieties.
From a morphological point of view, Turda soybean varieties are characterized by gray or red pubescence, violet or white flower, and different colors of hilum (grey, yellow, black and different shade of brown). Cristina TD, Caro TD, Larisa, Ilinca TD, Ada TD, Darina TD are varieties with high yield and also a very good stability of the yield. Until now, Cristina TD is considered the most valuable creation regarding yield; in the trial organized at Agricost Brăila, Insula Mare a Brăilei it

reached 4779 kg/ha. It should note the production potential of the Ada TD variety, which in irrigation conditions reached 5703 kg/ha in Mircea Vodă testing center, in 2015 (Mureşanu et al, 2016). All the Turda soybean varieties but especially Ilinca TD, Miruna TD, Nicola TD, Darina TD, Ada TD and Teo TD have high insertion of the basal pods (over 19 cm), being suitable for mechanized harvest.

In Europe there is an increasing interest for conventional soybean (Dima, 2018). At Freising, in 2013, the European soybean breeders established new direction for their breeding programs namely the creation of new varieties with specific destination: human consumption, animals feeding and oil derivatives. Selection of the appropriate variety is a very important task and needs to be taken seriously since it directly influences yields and also the seed quality.

In this context, the quality parameters of plant raw material are important issues for both breeding programs and food processors, due to their impact on the quality of the end-products; hence, they are important criteria for selection of appropriate genotypes for specific consumer's preferences. Since in the last years, consumers are becoming increasingly aware of the health benefits of soybean, there is a growing interest in breeding programs directed towards the improvement of taste, functional characteristics and health benefits (Gandhi, 2009; Lagos & Stein, 2017; Mihalache, 2006).

The main purpose of this study was to assess the quality parameters and grain yields of the soybean varieties produced at ARDS Turda in the conditions of the year 2018, then to perform a chemometric evaluation of the obtained results in order to highlight similarities between the studied varieties.

MATERIALS AND METHODS

Biological material

The seeds used in this study were produced at ARDS Turda in 2018, twenty early and very early soybean varieties being evaluated: Diamant, Perla, Safir, Granat, Eugen, Onix, Felix, Darina TD, Cristina TD, Mălina TD, Carla TD, Larisa, Caro TD, Ilinca TD, Bia TD, Ada TD, Teo TD, Miruna TD, Nicola TD and Felicia TD. Field trials were based on a randomized block design with three replications with the harvested plot of 10 m^2; each soybean variety was sowed on two rows of 12 m length and 50 cm distance between rows. At the end of the growing period 10 plants/ variety were analyzed for height and for insertion of the basal pods. The plots were harvested at the end of September 2018 with a Wintersteiger Classic Plot Combine (Wintersteiger AG, Austria); the thousand kernel weight (TKW) was determined using a Sadkiewicz Seed Counter (Sadkiewicz Instruments, Bydgoszcz, Poland) and a KERN 573-34 NM analytical balance (Kern & Sohn GmbH, Bahlingen-Frommern, Germany); the results are the average of 100 seeds counted and weighed in 8 replications.

Sampling and sample processing

Average representative samples of soybean seeds were collected from each variety; they were milled using a WZ-1 laboratory mill (Sadkiewicz Instruments, Bydgoszcz, Poland) the resulted powder being used for next analytical steps after a weighing step, using a Kern ABT-220-5DM analytical scale (Kern & Sohn GmbH, Bahlingen-Frommern, Germany).

Analytical determinations

Proximate composition of seeds was accomplished according to AOAC methods (Latimer, 2012); all determinations were accomplished in triplicates and mean values were reported. The dry matter content was determined by drying using a SLW 53 programmable forced air convection oven (POL-EKO-Aparatura, Wodzisław Śląski, Poland). The ash content was established by calcination at 550°C in a Nabertherm B180 muffle furnace (Nabertherm GmbH, Lilienthal, Germany). The protein content was determined using the Kjeldahl method: about 0.5 g of samples were weighed and transferred into 250 mL digestion tubes, where 20 mL of concentrated sulfuric acid (Chempur, Piekary Śląskie, Poland) and two tablets of Kjeltabs CX catalysts (each containing 5 g K_2SO_4 and 0.5 g $CuSO_4$. $5H_2O$ - Gerhardt, Koenigswinter, Germany) were added, digestion being therefore achieved for three hours, at 400°C using a Turbotherm TT 265 digestion unit connected with a Turbosorg Tur/K scrub unit (both from Gerhardt (Koenigswinter, Germany). The digested samples were treated with 32% sodium hydroxide (Chempur, Piekary Śląskie, Poland) and the resulted ammonia was distilled in a Vapodest 30S device (Gerhardt, Koenigswinter, Germany) in a known volume of standardized 0.1 N sulfuric acid. The nitrogen content was determined by titration with a standardized 0.1 N NaOH solution using a classic burette; the titration's endpoint was established using as indicator a mixture of five parts of bromocresol green and one part of methyl red (both from Sigma Aldrich). The crude protein content was estimated by multiplying the nitrogen content with the conversion factor 6.25. The total fat content was determined using the Soxhlet method, in a Det Gras N6p system (JP Selecta, Barcelona, Spain); after the recovery stage, the aluminium bakers were transferred in an SLW 53 drying oven (POL-EKO-Aparatura, Wodzisław Śląski, Poland), where they were kept at 105°C for one hour; after cooling, the final weighing was carried out on a Kern ABT-220-5DM analytical scale (Kern&Sohn GmbH, Bahlingen-Frommern, Germany). The total carbohydrates' content was assessed by difference: (dry matter - protein - fat - ash), this including the polyglucides from starch and fibres.

The total carotenoid content was determined by UV-VIS spectrophotometry, after extraction of about 0.5 g sample with 15 ml ethanol (Merck KGaA Darmstadt, Germany), on an Arex magnetic stirrer (Velp Scientifica, Italy), at 40°C, for 30 min, at 150 rpm. The resulted suspensions were transferred in 15 ml Falcon centrifuge tubes, and then centrifuged at 5000 rpm for 10 minutes on a Hettich Universal 320 (Hettich, Germany). The volumes of supernatants were measured and the absorbances of the extracts were measured at 450 nm using a T80+ UV/VIS Spectrophotometer (PG Instruments Ltd, Leicestershire, UK). The total carotenoid content was calculated based on the formula:

$$\mathrm{TC} = \frac{A.V.10}{2500.m} \quad \text{[mg/kg]}$$

where: A - recorded absorbance; V - volume [ml]; m - sample weight [g] (Britton et al., 1995).

The solutions were prepared using bidistilled water, obtained in a GFL 2104 system (GFL Gesellschaft fur Labortechnik, Burgwedel, Germany).

Data analysis

The data matrix was prepared and processed in Excel (Microsoft, USA), then chemometric analysis was performed using MatLab (The Mathworks, USA) after mean center preprocessing.

RESULTS AND DISCUSSIONS

The studied soybean varieties are tall or very tall, with high insertion of the basal pods (Table 1). In natural conditions, they had a good or very good tolerance to specific diseases and pests. Agat, Safir, Eugen, Onix, Felix, Carla TD and Bia TD are typical early soybean varieties, while the other eleven tend to approach the maturity group 0. Onix and Felix varieties are the most cultivated, being preferred by farmers for high ecological plasticity; Felix and Cristina TD are recommended for food industry, while Caro TD is the new control variety used by Romanian State Institute for Variety Testing and Registration.

The experimental data are summarized in Table 1, highlighting genotypes with the maximum values for the studied parameters: Perla - with maximum values for dry matter (94.29%), lipids (18.38 g/100 g DW) and yield (3467 kg/ha), Safir - with the highest protein content (42.76 g/100 g DW), Nicola TD - with the maximum total carotenoid content (16.19 mg/100 g DW) and total carbohydrate content (36.51 g/100 g DW), Darina TD - with the highest ash content (5.80 g/100 g DW) and height (157 cm) and Teo TD with the maximum insertion (23 cm).

Principal component analysis (PCA) was based on seven variables (concentrations of proteins, carotenoids, carbohydrates, lipids, dry matter and ash), leading to a model which expose a close correlation between the yield (P) and the content of lipids (Lip), while explaining 57.39% of variance (Figure 1).

The scores' plot of the PCA model reveals three classes (Figure 2), confirmed by cluster analysis (Figure 3): one consisting from genotypes with high protein content (the blue one, with Onix, Felix, Eugen and Bia TD), a second joining genotypes higher in carbohydrates (the orange one, with Miruna TD, Nicola TD, Felicia TD, Ilinca TD and Mălina TD) and a third one containing the biggest number of the studied genotypes, characterized by intermediate values for the studied parameters (the red one, with Diamant, Agat, Darina TD, Cristina TD, Larisa, Caro TD, Bia TD, Ada TD and Teo TD).

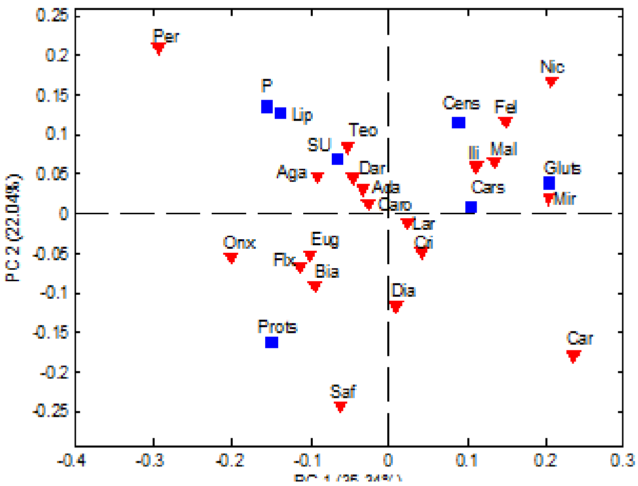

Figure 1. Biplot for the experimental dataset (P - yield, Lip - lipids, SU - dry matter, Prots - protein content, Cens - ash, Cars- total carotenoids, Gluts - total carbohydrates)

Several genotypes are much dissimilar from the described clusters: thus, Carla TD has the lowest lipid content from all, Safir has both the highest protein content and the smallest ash content, while Perla is an outlier of this model, being outside the border corresponding to the 95% confidence interval due to its exceptional yield, combined with the maximum dry weight and maximum lipid content.

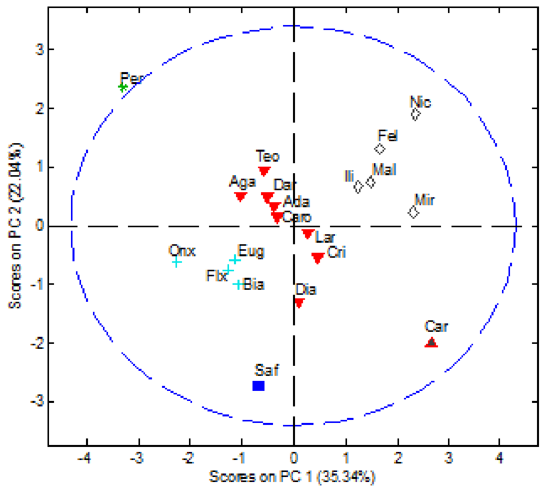

Figure 2. Score's plot for the PCA model

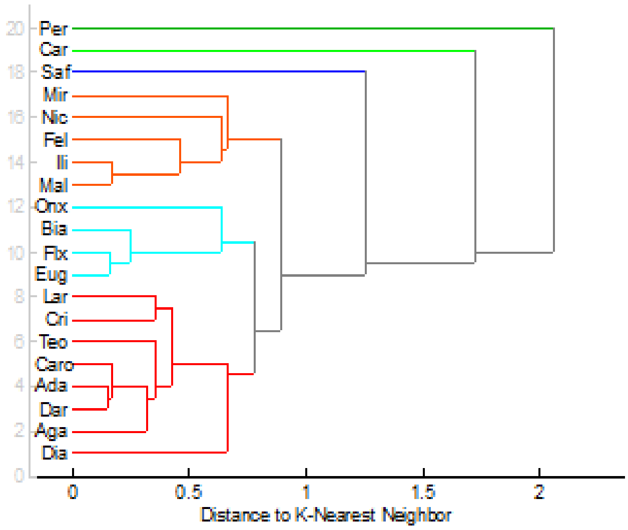

Figure 3. Dendrogram obtained for the PCA model (K-Nearest Neighbour method)

Table 1: Experimental data obtained for soybean genotypes from RDSA Turda

Genotype	Dry matter [%]	Lipids [g/100 g DW]	Proteins [g/ 100 g DW]	Total carotenoids [mg/kg DW]	Ash [g/100 g DW]	Total carbohydrates [g/100 g DW]	Yield [kg/ha]	TKW [g]	Height [cm]	Insertion [cm]
Diamant	93.83	14.48	41.55	10.81	5.69	32.11	2214	161	-	-
Perla	**94.29**	**18.38**	40.37	10.04	5.73	29.81	**3467**	144	95	13
Agat	94.24	16.50	39.12	8.62	5.15	33.48	2560	152	-	-
Safir	93.97	15.31	**42.76**	13.28	4.70	31.21	1905	142	-	-
Eugen	94.20	16.04	40.82	13.65	4.63	32.72	2679	157	121	14
Onix	94.01	18.28	41.08	13.51	4.56	30.09	2565	129	137	16

Felix	93.66	17.35	40.61	11.77	5.07	30.63	2492	158	125	16
Darina TD	93.67	17.62	39.66	12.82	**5.80**	30.59	2508	150	**157**	19
Cristina TD	93.89	15.22	40.44	13.02	5.63	32.60	2285	152	126	15
Malina TD	94.17	17.07	37.53	15.20	5.45	34.12	1874	125	118	15
Carla TD	93.84	12.07	40.10	12.06	5.47	36.20	1838	**164**	124	16
Larisa	93.98	15.99	39.20	12.44	5.19	33.60	2355	148	131	16
Caro TD	93.88	17.02	39.65	14.10	5.30	31.91	2500	133	137	16
Ilinca TD	93.76	16.72	37.80	14.08	5.62	33.62	2278	151	153	21
Bia TD	93.73	16.82	41.27	12.91	5.15	30.49	2419	138	135	17
Ada TD	93.81	16.11	39.92	13.02	5.52	32.26	2831	150	149	20
Teo TD	93.86	18.06	38.84	13.36	5.47	31.50	2577	145	148	**23**
Miruna TD	93.77	16.23	37.20	15.08	5.47	34.87	1941	**164**	126	17
Nicola TD	94.04	16.28	35.77	**16.19**	5.48	**36.51**	2497	160	128	16
Felicia TD	93.87	15.56	36.49	11.12	5.60	36.23	2545	149	152	21

CONCLUSIONS

The obtained results for soybean cultivars created at ARDS Turda have identified valuable genotypes that can be used successfully in the crossing program for creating new varieties that align with the European soybean objectives.

Turda soybean varieties came forward with a high insertion of the basal pods, Teo TD being representative for this character (23 cm). The highest genotype in 2018 was Darina TD (157 cm), the biggest yield was obtained by Perla variety (3467 kg/ha) and TKW reached the value of 164 g (Carla TD and Miruna TD).

Nine out of twenty varieties had the protein content higher than 40 g/ 100 g DW, Safir variety obtaining the highest value for this quality parameter (42.76 g/100 g DW). For improving oil content, varieties with fat content higher than 18 g/100 g DW can be used: Perla, Onix and Teo TD. The sweetest varieties proved to be Nicola TD, Felicia TD and Carla TD, all with the total carbohydrate content higher than 36 g/100 g DW.

ACKNOWLEDGEMENT

This work was supported a CCCDI-UEFISCDI grant of the Romanian Ministry of Research and Innovation, project number PN-III-P1-1.2-PCCDI-2017-0301, contract number 28PCCDI/2018 – "Integrated management system of the agroecosystem resistance against pests in order to promote sustainable agriculture under the conditions of climate change", within PNCDI III."

REFERENCES

Britton G., Liaaen-Jensen S., Pfander H. (ed.), (1995b). *Carotenoids. Vol.1B: Spectroscopy*. Birkhauser Verlag, Basel, 131–9.62.

Clemente, T.E., Cahoon, E.B. (2009). Soybean oil: genetic approaches for modification of functionality and total content. *Plant physiology*, *151*(3), 1030–1040.

Dima, D.C. (2018), The yield performance of various soybean genotypes in five experimental fields in Romania and Bulgaria in 2015 and 2016. *Scientific Papers. Series A. Agronomy, 59*, 81–84.

Gandhi, A.P. (2009). Quality of soybean and its food products. *International Food Research Journal*, *16*. 11–1.

Goldflus, F., Ceccantini, M., Santos, W. (2006). Amino acid content of soybean samples collected in different Brazilian states: harvest 2003/2004. *Brazilian Journal of Poultry Science*, *8*(2), 105–111.

Idrisa, Y.L., Ogunbameru, B.O., Amaza, P.S. (2010), Influence of farmers socio-economic and technology characteristics on soybean seeds technology in Southern Borno State Nigeria, *African Journal of Agricultural Research*, *5*(12), 1394–1398.

Karr-Lilienthal, L.K., Grieshop, C.M., Merchen, N.R., Mahan, D.C., Fahey, G.C. (2004). Chemical composition and protein quality comparisons of soybeans and soybean meals from five leading soybean-producing countries. *Journal of Agricultural and Food Chemistry*, *52*(20), 6193–6199.

Lagos, L.V., Stein, H.H. (2017). Chemical composition and amino acid digestibility of soybean meal produced in the United States, China, Argentina,

Brazil, or India. *Journal of Animal Science, 95*(4), 1626–1636.
Liu, K. (1997). *Soybeans: chemistry, technology and utilization*. Aspen Publishers, New York.
Lombardi, A.R., Epure, L.I., Roman, Gh.V. (2013). Studies on soybean growing in Tulcea County. *Scientific Papers, Series A, Agronomy*, *56*, 308–313.
Jiang, G.L., Chen, P., Zhang, J., Florez-Palacios, L., Zeng, A., Wang, X., Bowen A.R., Miller A., Berry, H. (2018). Genetic analysis of sugar composition and its relationship with protein, oil, and fiber in soybean. *Crop Science*, *58*(6), 2413–2421.
Latimer, G.W. (2012). *Official methods of analysis of AOAC International*. AOAC international.
Mello Filho, O.L.D., Sediyama, C.S., Moreira, M.A., Reis, M.S., Massoni, G.A., Piovesan, N.D. (2004). Grain yield and seed quality of soybean selected for high protein content. *Pesquisa Agropecuária Brasileira*, *39*(5), 445–450.
Mihalache M. 2006. *Soils genesis, properties and taxonomy*. Ceres Publishing House, Bucharest.
Muresanu, E., Marginean, R. (2011). Early varieties of soybeans for the Transylvanian plain. *Bulletin UASVM Agriculture*, *68*, 1–12.
Muresanu, E., Marginean, R. (2011a). Soiuri de soia create la SCDA Turda pentru Campia Transilvaniei. *Agricultura Transilvana, Cultura plantelor de camp*, *4*, 56–59.
Mureşanu, E., Mărginean, R., Urdă, C. (2016). Soiul de soia „Ada TD", creat la Staţiunea de Cercetare-Dezvoltare Agricolă Turda. Analele *INCDA Fundulea*, *LXXXIV*, 107–114.
Mureşanu, E., Mărginean, R., Urdă, C. (2017). *Contribuţii ale cercetării ştiinţifice la dezvoltarea agriculturii*, *VII*, 131–158.
Specht, J.E., Hume, D.J., Kumudini, S.V. (1999). Soybean yield potential—a genetic and physiological perspective. *Crop Science, 39*(6), 1560–157.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

MAIZE YIELD RECORDED IN RAINFED CONDITIONS OF SOUTHEASTERN ROMANIA AS INFLUENCED BY FERTILIZATION RATES AND HYBRID

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Abstract

Maize is a staple food worldwide, and the issues of growing it in current drought-oriented scenario of climate change rise challenges in agricultural practices, especially for the rainfed agriculture that is widely extended in Romania. The current paper presents the results of a field research conducted during 2017 and 2018 at Moara Domnească Didactic Farm, located in Southeastern Romania, aiming to highlight the influence of technological factors, as fertilization and hybrid, on maize yield recorded in rainfed conditions. Rainfalls capitalization by the maize crop shows yield increases between 25.2 kg and 37.8 kg grains per mm rainfalls, depending on hybrid and fertilization rates. The highest yield is recorded by the hybrid DKC 5086 (8512,7 kg/ha) at the fertilization level $N_{80}P_{40}$. Yield growth generated by mineral fertilization recorded an average value for the researched hybrids of 14.1 kg per kg of nitrogen applied and of 9.1 kg per kg of phosphorus.

Key words: *maize, rainfalls, nitrogen, phosphorus, yield.*

INTRODUCTION

Favorable growing conditions in Romania allowed maize crops (*Zea mays* L.) to have a positive dynamic, from ensuring 2.2% of the global maize yield in 1998 (Lăzăroiu et al., 2008) to being placed secondly in terms of yield and first in terms of surface in the European Union and in Romania (Ion et al., 2013). In 2018 Romania was first country in EU in terms of maize yield (USDA, Circular-Series, 2019).

Maize nutritional value but also its biological characteristics turn it into a valuable species of the *Poaceae* family, grown worldwide in 166 countries, thus being the second grown crop at global level after *Triticum aestivum* (Roman et al., 2011; Rouf-Shah et al., 2016; CONABIO, 2017; Ghete et al., 2018).

In countries from Latin America and Africa, maize is a vital source of food, a staple food for millions of people, thus it's growing being an issue of food security (Jones & Thornton, 2003; Ramirez-Cabral et al., 2017).

Romanian farmers currently benefit of a large assortment of maize hybrids from different precocity groups for a proper capitalization of soil and climatic conditions specific to cropping area and depending on the applied technology (Dumbravă et al., 2016; 2017).

In terms of cropping technology, fertilization is an important factor because it influences yield and yield indicators (Ion et al., 2015; Bășa et al., 2016). Nitrogen (N) is in general the most important macronutrient in crop's development, with a 50% rate of influence on the increase of global grains yield (Erisman et al., 2008) but also influencing yield inferior limits (Blumenthal et al., 2008) especially associated with water availability (Bacon 2004; Huang et al., 2006).

Maize has a superior capitalization of N fertilization, especially in a three years crop rotation compared to monoculture (Ciontu et al., 2012). For extra-early hybrids, nitrogen (along with foliar fertilization) in a maize crop irrigated with a volume of 900 m^3 water generated the best biomass yield increase for maize grown in successive crop at Moara Domnească (Mureșeanu et al., 2014). Other research conducted at Moara Domnească (Ciontu et al., 2011; Gidea et al., 2015) highlight a significant yield increase and a positive economic return proportional to the increase of the N fertilization rate obtained for the maize crop in a three years rotation system, while similar research conducted by Bășa et al. (2016) highlight the importance of phosphorus (P) along with N fertilization.

MATERIALS AND METHODS

The experimental field was located at Moara Domnească Didactic Farm (belonging to University of Agronomic Sciences and Veterinary Medicine of Bucharest), on a chromic luvisol. The experimental design was based on two factors. Experimental variants had a surface of 56 square meters (8 rows with a length of 10 m at a distance of 70 cm between rows) and where arranged as split blocks with four replications.
Studied factors consisted of five maize hybrids (*Zea mays* L.) of different FAO (350-510) and three fertilization levels:

Factor A - fertilization levels:
- A_1 - N_0P_0 **(Ct)**
- A_2 - $N_{80}P_0$
- A_3 - $N_{80}P_{40}$.

Factor B - maize hybrids:
- B_1 - DKC 4590
- B_2 - DKC 5068
- B_3 - P9911
- B_4 - P0216
- B_5 - OLT

Sowing was conducted with a SPC-6 sowing machine on April 13, 2017 and on April 18, 2018 at a density of 62,000 of plants/ha, at a distance of 70 cm between rows. Fertilizers were applied gradually during seed bed preparation with a dose of 50 kg/ha active substance N:P (20: 20: 0), and with a dose of 30 kg/ha nitrogen active substance as ammonium nitrate during vegetative development.
Weed control was conducted chemically in preemergence with Dual Gold 960 EC (*s-metholachlor* 960 g/l) at a dose of 1.3 l/ha, and in postemergence with DicopurTop 464 SL (*2.4 D 344* g/l + *dicamba* 120 g/l) with a dose of 1.1 l/ha. Mechanical weed control consisted of two harrows applied during vegetative development. Climatic conditions were analyzed during plants vegetative development and compared to the multiannual values specific for the area. Thus, during crops vegetative development (April-August) average monthly temperature of the two years (2017-2018) was 21.4°C by 1.4°C over the multiannual value, while rainfalls amount recorded in the same period recorded for the two years an average monthly value of 243.8 mm, cu 71.9 mm lower that the multiannual value. July was highlighted because the difference recorded compared to the multiannual value was 43.3 mm (Table 1).

Table 1. Climatic data during maize vegetative development, average 2017-2018, Moara Domnească

Month	Temperature (°C)		Rainfall (mm)		
	Avg. monthly 2017-2018	Normal	Avg. monthly 2017-2018	Normal	Diff. compared to Normal
April	16.5	16.4	45.0	48.1	**- 3.1**
May	19.2	19.7	23.7	67.7	**- 44.0**
June	22.5	22.5	50.0	86.3	**- 36.3**
July	24.7	23.0	106.4	63.1	**+ 43.3**
August	24.2	20.4	18.7	50.5	**- 31.8**
Avg. or Sum	**21.4**	**20.4**	**243.8**	**315.7**	**- 71.9**

RESULTS AND DISCUSSIONS

Yield growth as influenced by rainfall
Average maize yield growth determined by 1 mm rainfall was 28.9 kg/mm for the fertilization N_0P_0, 34.1 kg/mm when $N_{80}P_0$ was applied and 35.8 kg/mm for $N_{80}P_{40}$ fertilization (Table 2). The lowest yield increase recorded under the influence of rainfall was obtained by Olt hybrid for the unfertilized variant (28.7 kg/mm). The highest yield increase generated by rainfall was recorded at the fertilization level $N_{80}P_{40}$ (37.8 kg/mm) by DKC 5068 hybrid, a mid-late hybrid that capitalized through its vegetative development higher rainfalls recorded especially in June and July.
An interdependence was observed when analyzing maize yield related to rainfall amount during the vegetative development of the crop, highlighted by a correlation coefficient r = 0.7669 (Figure 1). Considering regression analysis results around 59% (R^2 = 0.5881) of the maize yield increase can be associated to the amount of rainfall fallen during maize crop vegetative development.

Table 2. Maize yield increase (kg) per mm of rainfall, (Moara Domnească, 2017-2018)

Hybrid	Fertilization level	Yield increase (kg/mm rainfall)
DKC 4590	$N_0 P_0$	28.9
	$N_{80} P_0$	34.1
	$N_{80}P_{40}$	35.8
DKC 5068	$N_0 P_0$	30.1
	$N_{80} P_0$	36.1
	$N_{80}P_{40}$	37.8
P9911	$N_0 P_0$	27.3
	$N_{80} P_0$	32.0
	$N_{80}P_{40}$	33.7
P0216	$N_0 P_0$	28.7
	$N_{80} P_0$	33.6
	$N_{80}P_{40}$	35.2
OLT	$N_0 P_0$	25.2
	$N_{80} P_0$	29.4
	$N_{80}P_{40}$	30.8
Average hybrids	$N_0 P_0$	28.9
	$N_{80} P_0$	34.1
	$N_{80}P_{40}$	35.8

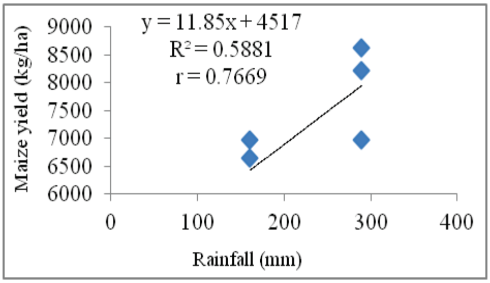
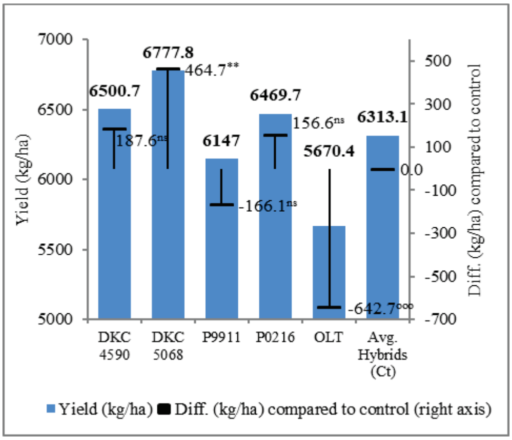


Figure 1. Rainfall influence (mm) on grain yield

Yield recorded in rainfed conditions as influenced by hybrid.

For the unfertilized variant (Figure 2) the five maize hybrids recorded an average yield (control) of 6313.1 kg/ha. Compared to control the highest yield increase, distinctly significant in statistical terms, was recorded by DKC 5068 (464.7 kg/ha), while the highest decrease was obtained by Olt hybrid, with a very significant negative difference of -642.7 kg/ha.



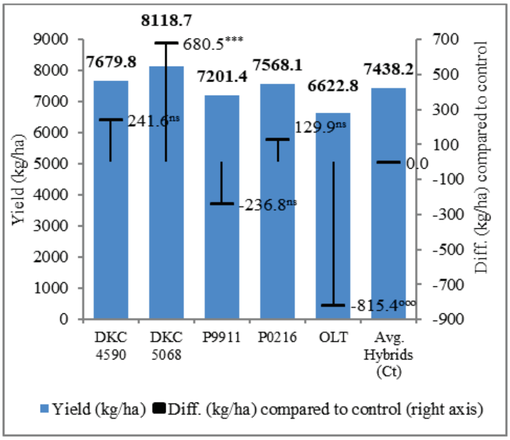
LSD 5% = 325.0 kg/ha; LSD 1% = 455.6 kg/ha; LSD 0.1% =644.0 kg/ha

Figure 2. Hybrid influence on grain yield for the fertilization level $\mathbf{N_0P_0}$

Other researched hybrids obtained yield differences compared to control with values between -166.1 kg/ha and 187.6 kg/ha, but non-significant in statistical terms (Figure 2).

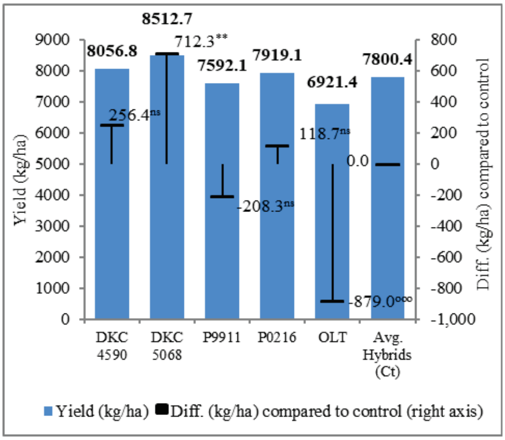
DCK 5068 hybrid obtained the highest yield (8118.7 kg/ha) for the fertilization level $\mathbf{N_{80}P_0}$, with a very significant increase of 680.5 kg/ha compared to the average yield of the hybrids (control) (Figure 3). The smallest yield was recorded by Olt (6622.8 kg/ha), with a very significant difference of -815.4 kg/ha from control (Figure 3). Hybrids DKC 4590 and P0216 recorded yield increases from control, but non-significant in statistical terms, while hybrid P9911 had a negative difference from control but not statistically assured.

For the variant fertilized $\mathbf{N_{80}P_{40}}$ (Figure 4) a distinctly significant yield growth of 712.3 kg/ha compared to control was recorded by the hybrid DCK 5068, thus obtaining also the highest grain yield with a value of 8512.7 kg/ha. Hybrids DCK 4590 and P0216 also had higher yield than the average value but the differences were not statistically assured. Smaller yields than the average were recorded by P9911 and Olt, only the latest having a very significant negative difference of -879.0 kg/ha.



LSD 5% = 403.2 kg/ha; LSD 1% = 565.2 kg/ha; LSD 0.1% =798.9 kg/ha

Figure 3. Hybrid influence on grain yield for the fertilization level $\mathbf{N_{80}P_0}$



LSD 5% = 422.8 kg/ha; LSD 1% = 592.8 kg/ha; LSD 0.1% =837.9 kg/ha

Figure 4. Hybrid influence on grain yield for the fertilization level $\mathbf{N_{80}P_{40}}$

Compared to DCK 4590 hybrid (b_1), DKC 5068 (b_2) was the only one that recorded higher yields with statistically assured differences for the fertilized variants (Tabel 3). Hybrids P9911 (b_3), P0216 (b_4) and Olt (b_5) had lower yields, with differences statistically assured only for P9911 and Olt. These hybrids also had lower yields than DKC 5068 (with distinctly significant (b_4-b_2, except the unfertilized variant) and very significant differences (b_3-b_2, b_5-b_2) for all fertilization levels. Compared to the hybrid P9911, P0216 recorded higher yields with a significant difference only for the fertilized variant N_{80} P_0, while Olt had lower yields with statistically significant differences. Olt also recorded lower yields compared to P0126, the negative differences being very significant in statistical terms.

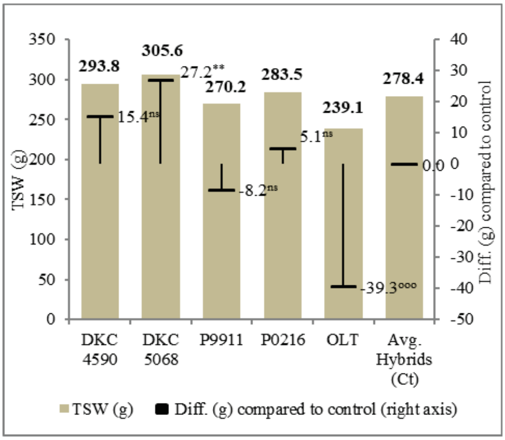
Thousand seeds weight (TSW) recorded in rainfed conditions as influenced by hybrid

Hybrid influence on maize thousand seeds weight obtained for the unfertilized variant (Figure 5) reflects on TSW increase for the hybrid DKC 5068 with a distinctly significant difference (27.2 g) compared to the average value of the hybrids (control). Positive influence of the hybrid on TSW was also recorded for DKC 4590 and P0216 hybrids, but increases compared to control were not statistically assured. P9911 had a lower yield than control with a insignificantly, with an non-significant negative difference (-8.2 g), while Olt recorded the smallest TSW with a very significant difference compared to control (-39.3 g).

Table 3. Yield differences among maize hybrids (Moara Domnească, 2017-2018)

Fertilization level	b_2-b_1	b_3-b_1	b_4-b_1	b_5-b_1	b_3-b_2	b_4-b_2	b_5-b_2	b_4-b_3	b_5-b_3	b_5-b_4
N_0P_0	277.1^{ns}	-353.7^{o}	-31.0^{ns}	-830.3^{ooo}	-630.8^{ooo}	-308.1^{ns}	-1107.4^{ooo}	322.7^{ns}	-476.6^{oo}	-799.3^{ooo}
$N_{80}P_0$	438.9*	-478.4^{oo}	-111.7^{ns}	-1057.0^{ooo}	-917.3^{ooo}	-550.6^{oo}	-1495.9^{ooo}	366.7*	-578.6^{oo}	-945.3^{ooo}
$N_{80}P_{40}$	455.9**	-464.7^{oo}	-137.7^{ns}	-1135.4^{ooo}	-920.6^{ooo}	-593.6^{oo}	-1591.3^{ooo}	327.0^{ns}	-670.7^{ooo}	-997.7^{ooo}

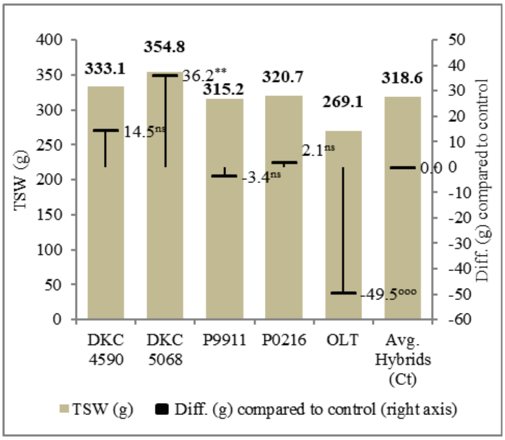
LSD 5% = 332.4 kg/ha; LSD 1% = 453.4 kg/ha; LSD 0.1% =613.9 kg/ha



LSD 5% = 15.6 g; LSD 1% = 21.8 g; LSD 0.1% = 30.9 g

Figure 5. Hybrid influence on TSW for the fertilization level $\mathbf{N_0P_0}$

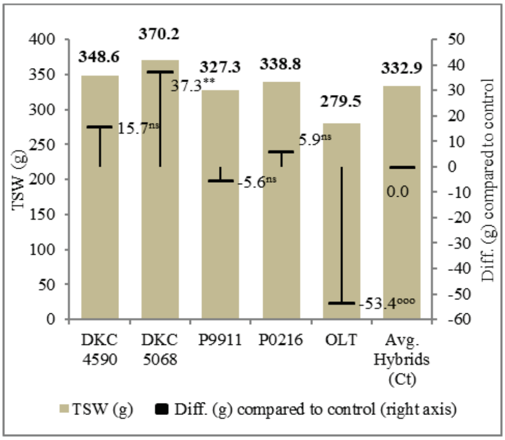
For the fertilization level $N_{80}P_0$ hybrid DKC 5068 recorded the highest and also distinctly significant TSW increase compared to control (average TSW of the hybrids). The smallest TSW with the greatest negative difference (-49.5), very significant in statistical terms was recorded by Olt hybrid (Figure 6). Hybrids DKC 4590, P9911 and P0216 varied compared to control in statistical terms but the recorded differences either negative or positive where not statistically assured.



LSD 5% = 18.6 g; LSD 1% = 26.1 g; LSD 0.1% = 36.9 g

Figure 6. Hybrid influence on TSW for the fertilization level $\mathbf{N_{80}P_0}$

On the fertilization level $\mathbf{N_{80}P_{40}}$ (Figure 7) a distinctly significant compared to control growth of 37.3 g was recorded by the hybrid DCK 5068, this recording the highest TSW. Hybrids DCK 4590 and P0216 also recorded higher differences but not statistically assured. Smaller TSW compared to the average were recorded by P9911 and Olt, only the latest having a very significant negative difference.



LSD 5% = 19.4 g; LSD 1% = 27.2 g; LSD 0.1% = 38.5 g

Figure 7. Hybrid influence on TSW for the fertilization level $\mathbf{N_{80}P_{40}}$

Grain yield and TSW variation under different fertilization rates

Fertilization positively influenced both maize yield and a thousand seeds weight compared to control, unfertilized variant (Table 4). For the average yield of the five hybrids nitrogen fertilization at a rate of 80 kg N a.s./ha generated a very significant yield increase of 1125.0 kg/ha, while complex mineral fertilization with N_{80} P_{40} determined also a very significant yield increase of 1487.3 kg/ha. The highest yield increase compared to the unfertilized variant was recorded by the DKC 5068 fertilized N_{80} P_{40} (1734.9 kg/ha) while the smallest growth was recorded by Olt fertilized with $N_{80}P_0$ (952.4 kg/ha). Both differences were very significant positive. Due to fertilization a thousand seeds weight recorded very significant differences compared to control (unfertilized variant) for all the researched hybrids. The average TSW increased by 40.1 g for the fertilization level $N_{80}P_0$ and by 54.4 g for the fertilization level $N_{80}P_{40}$ (Table 4).

Table 4. Fertilization influence on maize yield and TSW (Moara Domnească, 2017-2018)

Hybrid	Fertilization level	Yield (kg/ha)	%	Diff. (t/ha)	Signf.	TSW (g)	%	Diff. (g)	Signf.
DKC 4590	**N_0 P_0**	6500.7	**100.0**	**Ct**	-	293.8	**100.0**	**Ct**	-
	N_{80} P_0	7679.8	118.1	1179.10	***	333.1	113.4	39.30	***
	$N_{80}P_{40}$	8056.8	123.9	1556.10	***	348.6	118.7	54.80	***
DKC 5068	**N_0 P_0**	6777.8	**100.0**	**Ct**	-	305.6	**100.0**	**Ct**	-
	N_{80} P_0	8118.7	119.8	1340.9	***	354.8	116.1	49.2	***
	$N_{80}P_{40}$	8512.7	125.6	1734.9	***	370.2	121.1	64.6	***
P9911	**N_0 P_0**	6147.0	**100.0**	**Ct**	-	270.2	**100.0**	**Ct**	-
	N_{80} P_0	7201.4	117.2	1054.4	***	315.2	116.7	45.0	***
	$N_{80}P_{40}$	7592.1	123.5	1445.1	***	327.3	121.1	57.1	***
P0216	**N_0 P_0**	6469.7	**100.0**	**Ct**	-	283.5	**100.0**	**Ct**	-
	N_{80} P_0	7568.1	117.0	1098.4	***	320.7	113.1	37.2	***
	$N_{80}P_{40}$	7919.1	122.4	1449.4	***	338.8	119.5	55.3	***
OLT	**N_0 P_0**	5670.4	**100.0**	**Ct**	-	239.1	**100.0**	**Ct**	-
	N_{80} P_0	6622.8	116.8	952.4	***	269.1	112.5	30.0	***
	$N_{80}P_{40}$	6921.4	122.1	1251.0	***	279.5	116.9	40.4	***
Average hybrids	**N_0 P_0**	6313.1	**100.0**	**Ct**	-	278.4	**100.0**	**Ct**	-
	N_{80} P_0	7438.2	117.8	1125.0	***	318.6	114.4	40.1	***
	$N_{80}P_{40}$	7800.4	123.6	1487.3	***	332.9	119.6	54.4	***

LSD 5% = 330.9 kg/ha; LSD 1% = 442.8 kg/ha; LSD 0.1% =581.5 kg/ha

LSD 5% = 14.8 g; LSD 1% = 19.8 g; LSD 0.1% = 26.0 g

Correlation coefficient analysis confirms the strong dependency relation (r = 0.9951), for the field conditions of the current research, between maize yield and the amount of fertilizers active substance applied (Figure 8).

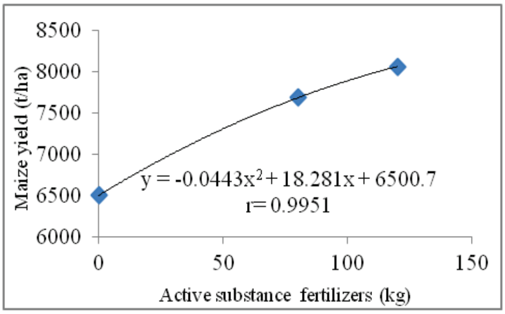


Figure 8. Fertilizer active substance influence on maize yield

Yield increase (Table 5) generated by nitrogen fertilization varied between 11.9 kg per kg of nitrogen active substance (Olt $N_{80}P_0$) and 16,8 kg per kg of nitrogen active substance (DKC 5068 $N_{80}P_0$). Yield growth generated by phosphorus fertilization recorded values from 7.5 kg/kg a.s. P and 9.9 kg/kg a.s. P. On average a kg of phosphorus generated a yield increase of 9.1 kg, while nitrogen generate an average yield growth of 14.1 kg.

Table 5. Yield increase (kg) per kg of fertilizer active substance (Moara Domnească, 2017-2018)

Hybrid	Fertilization level	Yield increase (kg/kg a.s. N)	Yield increase (kg/kg a.s. P)
DKC 4590	N_{80} P_0	14.7	-
	$N_{80}P_{40}$	-	9.4
DCK 5068	N_{80} P_0	16.8	-
	$N_{80}P_{40}$	-	9.9
P9911	N_{80} P_0	13.2	-
	$N_{80}P_{40}$	-	9.8
P0216	N_{80} P_0	13.7	-
	$N_{80}P_{40}$	-	8.8
Olt	N_{80} P_0	11.9	-
	$N_{80}P_{40}$	-	7.5
Average hybrids	N_{80} P_0	14.1	-
	$N_{80}P_{40}$	-	9.1

CONCLUSIONS

Considering this research results we can conclude that maize yield was significantly influenced both by fertilization and hybrid, while its increase was also conditioned by the amount of rainfall during the vegetative period. The use of nitrogen based mineral fertilizers generated an average yield increase of 117.8% compared to the unfertilized variant, and a TSW increase of 114.4%, while complex fertilization $N_{80}P_{40}$ generated an average yield growth of 123.6% and a average TSW increase of 119.6%.

Hybrid influence was highlighted by distinctly significant yield growths recorded by DCK 5068 compared to the average yield of the hybrids for all fertilization levels and very significant negative differences recorded by Olt hybrid.

Yield analysis under the combined influence of fertilization and rainfall highlights the following:

- a kg per ha of nitrogen generates an average yield increase of 14.1 kg;
- a kg per ha of phosphorus determines an average yield growth of 9.1 kg;
- a mm of rainfall generates yield increases between 25.2 kg and 37.8 kg.

ACKNOWLEDGEMENTS

This research was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Bacon, M.A. (2004). *Water Use Efficiency in Plant Biology*. Blackwell Publishing, (CRC Press) Oxford, UK.

Bășa, G.A., Ion, V., Dumbravă, M., Temocico, G., Epure, L.I., Ștefan, D. (2016). Grain yield and yield components at maize under different preceding crops and nitrogen fertilization conditions. 5th International Conference-Agriculture for Life, Life for Agriculture. *Agricultural and Agricultural Science Procedia*, *10*, 104–111.

Blumenthal, J.M., Baltensperger, D.D., Cassman, K.G., Mason, S.C., Pavlista, A.D. (2008). Importance and effect of nitrogen on crop quality and health. In: *Nitrogen in the Environment: Sources, Problems and Management. Eds. - Follett, R.F. and Hatfield, J.L.,* 45–63.

Ciontu, C., Săndoiu, D.I., Penescu, A., Gîdea, M., Obrișcă, M. 2011. Research concerning influence of crop rotation on maize grown on the reddish preluvosoil from Moara Domnească. *Scientific Papers. Series A. Agronomy, LIV*, 217–222.

Ciontu, C., Săndoiu, D.I., Penescu, A., Gîdea, M., Șchiopu, T., Obrișcă, M., Dincă, L. (2012). The essential role of crop rotation and nitrogen fertilization in wheat and maize in the sustainable agriculture system of reddish preluvosoil. *AgroLife Scientific Journal*, *1*(1), 68–77.

CONABIO, (2017). Ecosystem and agro-biodiversity across small large-scale production systems. TEEB Agriculture& Food, UNEP, Geneva, 2–8.

Dumbravă, M., Ion, V., Bășa, A.G., Dobrin, I., Niculae, I., Dincă, N. (2017). Yield components at different maize hybrids under the specific conditions from south Romania. *Scientific Papers. Series A. Agronomy*, *LX*, 246–251.

Dumbravă, M., Ion, V., Bășa, A.Gh., Dușa, E.M. (2016). The behaviour of some maize hybrids under the specific conditions from South Romanian Plain. *16th GeoConference International Multidisciplinary Scientific (SGEM), Energy and Clean Technologies Conference Proceedings, Book 4 - Renewable Energy Sources and Clean Technologies, I*, 691–696.

Erisman, J.W., Sutton, M.A., Galloway, J., Klimont, Z., Winiwarter, W. (2008) How a century of ammonia synthesis changed the world. *Nature Geoscience*, *1*(10), 636–639.

Ghete, A.B., Duda, M.M., Varban, D.I., Varban, R., Moldovan, C., Muntean, S. (2018). Maize (*Zea Mays*), a prospective medicinal plant in Romania. *Hop and Medicinal Plants*, *XXVI*(1-2), 44–51.

Gîdea, M., Ciontu, C., Sandoiu, D.I., Penescu, A., Schiopu, T., Nichita, M. (2015). The role of rotation and nitrogen fertilization level upon the economic indicators at wheat and corn crops in condition of a long term experience. 4th International Conference - Agriculture for Life, Life for Agriculture. *Agricultural and Agricultural Science Procedia*, *6*, 24–29.

Huang, R., Birch, C.J., George, D.L. (2006). Water use efficiency in Maize production-The challenge and improvement strategies. *6thTriennial Conference. Maize Association of Australia*.

Ion, V., Dicu, G., State, D., Fîntîneru, G., Epure, L.I., Bășa, A.G., Toader, M., (2013). Yield components of different hybrids of maize (*Zea mays* L.) cultivated in South Romania under drought conditions. *Scientific Papers. Series A. Agonomy*, *LVI*, 276–283.

Ion, V., Bășa, A. G., Dumbravă, M., Epure, L. I., Dincă, N., Toader, M. (2015). Grain yield and yield components at maize under different preceding crops and soil tillage conditions. *AgroLife Scientific Journal*, *4*(2), 27–32.

Jones, P.G., Thornton, P.K. (2003) The potential impacts of climate change on maize production in Africa and Latin America in 2055. *Global Environmental Change*, *13*(1), 51–59.

Lăzăroiu, A., Levițchi, A.C., Ciocăzan, I., Oșelea, S., Săbăilă, I., Șarpe, N., Petersen, L., Cojocaru, I., Pașol, P., Gheorghieș, C., Lăzăroiu, M., Tianu, A., Alexe, G., Urban, T. (2008). Porumbul - să valorificăm cât mai bine potențialul de producție ridicat al hibrizilor Pioneer. Editura Speteanu, București, 1–47.

Mureseanu, S., Marin, D.I., Bolohan, C. (2014). Fresh biomass production of double crop maize (*Zea mays* L.) hybrids cultivated in Moara Domneasca, Ilfov. *Scientific Papers. Series A. Agronomy*, *LVII*, 276–280.

Ramirez-Cabral, N.Y.Z., Kumar, L., Shabani, F. (2017). Global alterations in areas of suitability for maize production from climate change and using a mechanistic species distribution model (CLIMEX). *Scientific Reports*, *7*(1), 1–13.

Roman, G.V., Tabără, V., Robu, T., Pîrșan, P., Ștefan, M., Axinte, M., Morar, G., Cernea, S. (2011). *Fitotehnie - Cereale și leguminoase pentru boabe*. Editura Universitară, București, Vol. 1., (pp. 189–234).

Rouf-Shah, T.., Prasad, K., Kumar, P. (2016). Maize - a potential source of human nutrition and health: review. *Cogent Food & Agriculture*, *2*(1), 1–9.

USDA, United States Department of Agriculture (2019). World Agricultural Productions, Circular Series WAP-2 February, Foreign Agricultural Service/Office of Global Analysis, 2.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

MACRONUTRIENTS CONCENTRATION IN DURUM WHEAT VARIETIES GROWN AT DIFFERENT NITROGEN RATES

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Abstract

The concentration of nitrogen, phosphorus and potassium in the grain and straw of seven Bulgarian durum wheat cultivars (Progress, Vazhod, Victoria, Predel, Deana, Zvezdica and Elbrus) in dependence on N rates 0, 60, 120 and 180 kg.ha^{-1} was studied in a field experiment with cotton-durum wheat crops rotation during the period 2010-2014 under non-irrigation conditions. It was established that rates N120 and N180 significantly increased N concentration of the wheat grain. Strong genotypic response of durum wheat to the grain N concentration was obtained in non-fertilized plants with variation from 1.96% N (Vazhod) to 2.40% N (Zvezdica). Predel and Zvezdica varieties showed higher grain N content. Newer varieties (Victoria, Predel, Deana, Zvezdica and Elbrus) were characterized with higher N concentration of the straw. N rates 0-180 kg N.ha^{-1} slightly changed the phosphorus concentration in the wheat grain (0.58-0.61% P_2O_5) and in the straw (0.10-0.19% P_2O_5). Deana and Vazhod varieties had higher grain phosphorus concentration when they were grown without fertilization, whereas Zvezdica variety was exceeded other varieties at rates N120 and N180. Predel and Zvezdica varieties showed higher potassium concentration of the grain.

Key words: *concentration of macronutrients, nitrogen, durum wheat.*

INTRODUCTION

The agrochemical evaluation of cultivars and hybrids was established as a compulsory part of modern breeding for high productivity of cereals in the last years (Sylvester-Bradley & Kindred, 2009). The newer genotypes of durum wheat have higher productive potential but they realize it after greater nutrient input (Johnson, 2004). Grain quality is the most important criterion in the breeding of durum wheat to produce high quality pasta (Ammes et al., 2003; Uppal et al., 2002). The new genotypes combine high productivity with good quality (Panayotova & Valkova, 2010; Rharrabti et al., 2003). Many studies have been conducted to examine the effects of nitrogen fertilizers and preceding crops on cereal grain yield. Some authors (Bauer et al., 1987; Carcea, 2003; Kostadinova, 2000) reported that the increasing nitrogen rate and rich soil fertility enhanced the content of grain protein and nitrogen in the straw. The responsiveness of different cultivars to nitrogen accumulated in the vegetative plant parts was established (May et al., 2008; Panayotova, 2010). Panayotova (2001) appoints a genotype specific in relation with grain yield depending on the nutrition level. Fertilizing effectiveness of varieties with different genetic traits under different soil fertility was established (Hawkesford, 2012; López-Bellido & López-Bellido, 2001; Mohammadi & Amri, 2009; Panayotova, 1999). Durum wheat requires optimum fertilizer rates, consistent with the conditions of the area and the specifics of the field to increase the grain quality (Panayotova & Gorbanov, 1999). Nitrogen has a strong influence on growth, but its impact on yield, quality and dry matter formation depends on the growing conditions. In meteorological terms, the favorable years strongly manifest the effect of higher nitrogen rate (Panayotova & Yanev, 2001; Rharrabti et al., 2003). The main requirement for good quality of the grain is for the plant to obtain the optimal nitrogen amount during vegetation (Fixen, 2009). The interaction between environmental conditions and nitrogen rates has a significant impact on grain quality (Sanjeev et al., 2000). Most research on wheat fertilization refers mainly to common wheat and only a limited number of them are on durum wheat, though not to the present-day most widespread intensive varieties. The aim of this study was to investigate the effect of nitrogen fertilization rates on the concentration of macronutrients nitrogen, phosphorus and potassium in the grain and straw of new Bulgarian durum wheat varieties.

MATERIALS AND METHODS

The concentration of nitrogen, phosphorus and potassium in the grain and straw of seven Bulgarian durum wheat cultivars (Progress, Vazhod, Victoria, Predel, Deana, Zvezdica and Elbrus) in dependence on nitrogen rates 0, 60, 120 and 180 kg $N.ha^{-1}$ was studied in a field experiment with cotton – durum wheat crop rotation during the period 2010-2014, at the Field Crops Institute - Chirpan, Bulgaria, under non-irrigation conditions. The experimental design consisted of a randomized block design with four replications. The harvested size of the plots was 10 m^2. Nitrogen fertilization in the form of NH_4NO_3 was applied before sowing (1/3 of the rate) and at early spring (2/3 of the rate). The phosphorus fertilization (P_{80}) was done before sowing in the form of triple superphosphate. The precursor crop was cotton fertilized by N_{80}.

The soil type of the experimental field was *Pellic vertisols* (FAO), generally referred to as the so called Mediterranean chernozems. The soil type is one of the most generous and widely spread and significant in Bulgaria. It is suitable for growing most of the field crops and has a potential for high yield. The main parent materials were pliozen clay deposits. It has a high-powered humus horizon (70–80 cm), with a compact zone of the profile (united horizon). By humus content it belongs to the mean humus soils. It characterizes with high humidity capacity, caused by the high percentage of clay minerals, with clay soil texture, small water-permeability, bulk density of the arable soil layer 1.2-1.3 $g.cm^{-3}$, with specific gravity 2.4-2.6 and low total porosity, neutral soil reaction and high cation exchange capacity (CEC) 35-46 meq per 100 g soil, with a high degree of bases saturation (93.4-100.0%), with total N in the arable layer ranging within 0.095-0.14% and low content of total phosphorus (0.05-0.11%), poor to medium supplied with hydrolyzed nitrogen, poorly supplied with available phosphorus and well-supplied with available potassium.

Regarding the temperature during durum wheat vegetation, one of the experimental years was characterized as very warm, two as hot and two with values close to the multi-annual rate. In terms of precipitation, two years were dry and in the three harvest years the rainfall values ware close to the norm.

The concentration of nitrogen, phosphorus and potassium were analyzed in the grain and straw of wheat cultivars. The samples of 0.5 g ground and dry plant material were mineralized using a wet digestion by H_2SO_4 and H_2O_2 as a catalyst (Mineev, 2001). The concentration of nitrogen and phosphorus in plant samples were determined by colorimetric methods and potassium concentration was analyzed by flame photometer model PFP-7 (Tomov et al., 2009).

The data were statistically analyzed with the ANOVA procedure within the SPSS statistical program and Duncan's multiple range test (P = 0.05) to find significant differences among means.

RESULTS AND DISCUSSIONS

Nitrogen concentration in durum wheat grain is the main quality indicator for durum wheat (Uppal et al., 2002). According to Klimashevskiy (1990) genotypic differences are well-expressed in the zone of deficient and moderate mineral nutrition. Other researchers found that at high nitrogen levels, the genotypic differences in the percentage of nitrogen in the wheat grain were large and associated with differences in distribution of grain nitrogen, whereas at low nitrogen levels the variation was very low and dependent on the presence of nitrogen in the soil to the anthesis (Ortiz-Monasterio et al., 1997).

The average nitrogen content in the grain of wheat varieties grown at four levels of nitrogen fertilization ranged from 2.18% N to 2.72% N (Table 1). Nitrogen rates N_{120} and N_{180} increased the nitrogen concentration of the grain, whereas the effect of applied low rate N_{60} was not significant compared to N_0 control. Genotypic response was more pronounced in non-fertilized plants where the nitrogen concentration significantly changed from 1.96% N (Vazhod) to 2.40% N (Zvezdica).

Predel and Zvezdica varieties were characterized by higher N content in the grain without nitrogen fertilization and at low rate N_{60}, as well as an average of the studied N levels.
The highest concentration of grain nitrogen was obtained in Vazhod variety (2.89% N) fertilized with N_{180}. The N concentration of the straw of studied varieties ranged from 0.36% N (Progress at N_0) to 0.97% N (Predel & Zvezdica at N_{180}) (Table 1). Nitrogen fertilization increased the average nitrogen concentration of the straw - from 0.56% N in the non-fertilized varieties to 0.73-0.78% N in the fertilized plants. The results showed no proven differences in the average nitrogen content of the straw at rates N_{60}, N_{120} and N_{180}. Older Progress and Vazhod varieties demonstrated lower straw nitrogen concentration (below 0.60% N on average) compared to newer Victoria, Predel, Deana, Zvezdica and Elbrus varieties.
The average concentration of phosphorus in the grain of the studied wheat varieties was changed within a narrow range of 0.58-0.61% P_2O_5 depending on nitrogen fertilization in the rates of 0-180 kg $N.ha^{-1}$ (Table 2). The genotypic specificity of the concentration of phosphorus in the grain of wheat varieties was established. On average, for the studied nitrogen rates, Zvezdica variety showed the highest grain phosphorus content of 0.65% P_2O_5, while Progress and Elbrus varieties showed lower values of 0.54-0.56% P_2O_5. The Deana and Vazhod varieties were characterized with higher grain phosphorus concentration when were grown without fertilization, whereas the Zvezdica variety was exceeded other varieties at rates N_{120} and N_{180}. The studied durum wheat varieties demonstrated a low phosphorus concentration in the straw, which changed within a narrow range of 0.10-0.19% P_2O_5. Nitrogen fertilization with 120 and 180 kg $P_2O_5.ha^{-1}$ increased the average phosphorus content of the wheat straw. The average phosphorus concentration in the straw of durum wheat slightly depended on genotype.
Potassium concentration of the grain of the durum wheat changed from 0.16% K_2O (Vazhod at N_{180}) to 0.32% K_2O (Predel at N_{60}) (Table 3). The higher nitrogen rate N_{180} decreased the average potassium content of wheat grain compared to that of the plants grown without nitrogen and the plants fertilized with lower rate N_{60}. The genotypic reaction of durum wheat to the grain potassium concentration was proven.

Table 1. Nitrogen concentration of durum wheat varieties (N, %)

Variety \ Nitrogen rates	N_0	N_{60}	N_{120}	N_{180}	Average
Grain					
Progress	2.29 b	2.32 ab	2.50 c	2.55 d	**2.42 b**
Vazhod	1.96 f	1.95 e	2.26 d	2.89 a	**2.26 d**
Victoria	2.19 c	2.28 bc	2.65 b	2.62 cd	**2.43 b**
Predel	2.32 b	2.39 a	2.68 b	2.81 ab	**2.55 a**
Deana	2.08 d	2.07 d	2.52 c	2.64 c	**2.33 c**
Zvezdica	2.40 a	2.43 a	2.65 b	2.75 b	**2.56 a**
Elbrus	2.03 e	2.21 c	2.76 a	2.77 b	**2.44 b**
Average	**2.18 b**	**2.24 b**	**2.58 a**	**2.72 a**	
Straw					
Progress	0.36 d	0.66 cd	0.64 ef	0.68 bc	**0.58 c**
Vazhod	0.43 c	0.61 d	0.60 f	0.72 b	**0.59 c**
Victoria	0.63 b	0.69 c	0.68 de	0.65 c	**0.66 bc**
Predel	0.62 b	0.82 a	0.94 a	0.97 a	**0.83 ab**
Deana	0.44 c	0.69 c	0.73 cd	0.74 b	**0.65 c**
Zvezdica	0.82 a	0.87 a	0.84 b	0.97 a	**0.87 a**
Elbrus	0.64 b	0.75 b	0.74 c	0.75 b	**0.72 abc**
Average	**0.56 b**	**0.73 a**	**0.74 a**	**0.78 a**	

**Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.*

Table 2. Phosphorus concentration of durum wheat varieties (P_2O_5, %)

Variety \ Nitrogen rates	N_0	N_{60}	N_{120}	N_{180}	**Average**
Grain					
Progress	0.60 bc	0.54 c	0.45 d	0.57 c	**0.54 e**
Vazhod	0.63 ab	0.49 cd	0.66 ab	0.53 d	**0.58 cd**
Victoria	0.47 e	0.74 a	0.60 c	0.58 c	**0.60 bc**
Predel	0.60 c	0.60 b	0.64 abc	0.62 b	**0.61 b**
Deana	0.63 a	0.47 d	0.62 bc	0.64 b	**0.59 bc**
Zvezdica	0.60 bc	0.64 b	0.67 a	0.70 a	**0.65 a**
Elbrus	0.52 d	0.42 e	0.65 ab	0.63 b	**0.56 de**
Average	**0.58 ns**	**0.56**	**0.61**	**0.61**	
Straw					
Progress	0.10 c	0.10 b	0.14 bc	0.17 a	**0.13 ns**
Vazhod	0.10 c	0.11 b	0.14 bc	0.15 bc	**0.12**
Victoria	0.10 c	0.15 a	0.19 a	0.13 c	**0.14**
Predel	0.13 b	0.10 b	0.12 c	0.14 cd	**0.12**
Deana	0.10 c	0.11 b	0.18a	0.16 ab	**0.14**
Zvezdica	0.15 ab	0.15 a	0.15 b	0.14 cd	**0.15**
Elbrus	0.16 a	0.15 a	0.15 b	0.16 ab	**0.16**
Average	**0.12 b**	**0.12 b**	**0.15 a**	**0.15 a**	

**Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.*

Table 3. Potassium concentration of durum wheat varieties (K_2O, %)

Variety \ Nitrogen rates	N_0	N_{60}	N_{120}	N_{180}	Average
Grain					
Progress	0.27 b	0.23 d	0.22 b	0.19 d	**0.23 c**
Vazhod	0.29 a	0.30 b	0.25 a	0.16 e	**0.25 b**
Victoria	0.23 c	0.28 c	0.22 b	0.22 c	**0.24 b**
Predel	0.30 a	0.32 a	0.22 b	0.27 a	**0.28 a**
Deana	0.22 cd	0.31 ab	0.22 b	0.23 bc	**0.24 b**
Zvezdica	0.29 a	0.28 c	0.26 a	0.23 b	**0.27 a**
Elbrus	0.21 d	0.21 e	0.19 c	0.19 d	**0.20 d**
Average	**0.26 ab**	**0.28 a**	**0.23 bc**	**0.21 c**	
Straw					
Progress	1.03 f	1.16 b	1.20 d	1.39 c	**1.20 b**
Vazhod	1.24 e	1.31 ab	1.86 a	1.90 a	**1.58 a**
Victoria	1.69 a	1.21 ab	1.64 b	1.05 d	**1.40 ab**
Predel	1.37 d	1.33 a	1.68 b	1.45 c	**1.46 ab**
Deana	1.43 c	1.32 ab	1.42 c	1.71 b	**1.47 ab**
Zvezdica	1.62 b	1.34 a	1.61 b	1.73 b	**1.58 a**
Elbrus	1.23 e	1.26 ab	1.41 c	1.37 c	**1.32 ab**
Average	**1.38 ab**	**1.27 b**	**1.55 a**	**1.51 ab**	

**Values in each column followed by the same letters are not significantly different at $p<0.05$ according to Duncan's multiple range test.*

Predel and Zvezdica varieties were characterized by a higher average potassium concentration in the grain. Progress and Elbrus varieties showed lower average potassium concentration in the grain, and the other three Vazhod, Victoria and Deana varieties were occupied an intermediate position.

The percentage of potassium in the straw of durum wheat was nearly six times higher than that in the grain. It changed to a wider range of 1.03% K_2O (Progress at N_0) to 1.90% K_2O (Vazhod at N_{180}). Genotypic differences in the average potassium content of the straw were significant between Vazhod and Zvezdica varieties (0.58% K_2O) and Progress variety (1.20% K_2O).

Positive and negative correlations between examined parameters of durum wheat were recorded in the study (Table 4). Nitrogen fertilization was highly positively correlated with the concentration of nitrogen in the grain (r = 0.812**).

Also, a positive significant correlation was achieved between the nitrogen fertilization rates and concentration of nitrogen (r = 0.518**) and phosphorus (r = 0.514**) in the straw. A positive relationship was established between grain nitrogen concentration and the concentration of nitrogen, phosphorus and potassium in the straw. Nitrogen fertilization in rates from 0 to 180 kg $N.ha^{-1}$ was negatively correlated with concentration of potassium in the grain of durum wheat varieties.

Table 4. Correlation among nitrogen fertilization rates, concentration of nitrogen, phosphorus and potassium in grain and straw of durum wheat

Parameters	% N grain	% P_2O_5 grain	% K_2O grain	% N straw	% P_2O_5 straw	% K_2O straw
N fertilization	0.812**	0.220	-0.510**	0.518**	0.514**	0.341
% N grain		0.356	-0.498**	0.620**	0.519**	0.407*
% P_2O_5 grain			0.109	0.225	0.201	0.232
% K_2O grain				-0.044	-0.459*	-0.249
% N straw					0.379*	0.403*
% P_2O_5 straw						0.284

***Correlation is significant at the 0.01 level*
**Correlation is significant at the 0.05 level*

CONCLUSIONS

Nitrogen rates N_{120} and N_{180} significantly increased average nitrogen concentration of the wheat grain, whereas the effect of lower rate N_{60} was not significant compared to N_0. Strong genotypic response of durum wheat to the grain nitrogen concentration was obtained in non-fertilized plants with variation from 1.96 % N (Vazhod) to 2.40% N (Zvezdica). Predel and Zvezdica varieties showed higher grain nitrogen content. Newer varieties (Victoria, Predel, Deana, Zvezdica and Elbrus) were characterized with higher N concentration of the straw. N rates 0-180 kg $N.ha^{-1}$ slightly changed the concentration of phosphorus in the wheat grain (0.58-0.61% P_2O_5) and in the straw (0.10-0.19% P_2O_5). Deana and Vazhod varieties had higher grain phosphorus concentration when they were grown without fertilization, whereas Zvezdica variety was exceeded other varieties at rates N_{120} and N_{180}. The average phosphorus concentration in the straw of durum wheat slightly depended on genotype. The higher nitrogen rate N_{180} decreased the average potassium content of wheat grain compared to that of plants grown without nitrogen and plants fertilized with lower rate N_{60}. Predel and Zvezdica varieties showed higher average potassium grain concentration. The potassium percentage in the straw of durum wheat was nearly six times higher than that in the grain and it was changed to a wider range of 1.03% K_2O (Progress at N_0) to 1.90% K_2O (Vazhod at N_{180}). Nitrogen fertilization was highly positively correlated with the grain nitrogen concentration (r = 0.812**) and it was negatively correlated with the concentration of potassium in the grain.

REFERENCES

Ammes, N. P., Clark, J. M., Dexter, J. E., Woods, J., Schols, F., Marchylo, B. (2003). Effects of nitrogen fertilizer on protein quantity and gluten streng parameters in durum wheat (*Triticum turgidum* L. var. *durum*) cultivars of variable gluten strength. *Cereal Chemistry, 80*, 203–211.

Bauer, A., Frank, A., Black, A. (1987). Aerial parts of hard red spring wheat. II. Nitrogen and phosphorus concentration and content by plant development stage. *Agron. J.*, *79*, 852–858.

Carcea, M. (2003). The quality of durum wheat in Italy. *Sementi Elette*., *49*. 14–15.

Fixen, P. (2009). Nutrient Use Efficiency in the Context of Sustainable Agriculture, In J. Espinosa and F.

García (Eds.), Nutrient Use Efficiency, *International Plant Nutrition Institute* (IPNI), USA, 1–10.

Gastal, F., Lemaire, G. (2002). Nitrogen uptake and distribution in crops: an agronomical and ecophysiological perspective. *Journal of Experimental Botany*, *53*(370), 789–799.

Hawkesford, M. (2012). The Diversity of Nitrogen Use Efficiency for Wheat Varieties and the Potential for Crop Improvement. *Better Crops*, *96*(3), 10–15.

Johnson, A. (2004). Agricultural nutrients and climate change, Crop nutrients and the environment. *Progress Knowledge*, Canada, 202–211.

Klimashevskiy, E.P. (1991). *Genetic aspects of mineral nutrition of plants*. Agropromizdad, Moskva, 25–29.

Kostadinova, S. (2000). Response of winter common wheat (*Triticum aestivum* L.) to the level of nitrogen nutrition, *Ph.D. Thesis*, Sofia.

López-Bellido, R., López-Bellido, L. (2001). Efficiency of nitrogen in wheat under Mediterranean conditions: effect of tillage, crop rotation and N fertilization. *Field Crops Research*, *71*, 31–46.

May, W., Fernandez, M., Holzapfel, C., Lafond, G. (2008). Influence of Phosphorus, Nitrogen, and Potassium Chloride. Placement and Rate on Durum Wheat Yield and Quality. *Agronomy Journal*, *100*(4), 1179.

Mineev, V.G. (2001). *Manual of Agrochemistry*. Academic publisher of Moscow State University, Russia.

Mohammadi, R., and Amri, A. (2009). Analysis of Genotype x Environment Interactions for Grain Yield in Durum Wheat, *Crop Science*, *49*(4), 1177–1186.

Ortiz-Monasterio, J.I., Manske, G.B., Van Ginkel, M. (2001). Nitrogen and phosphorus use efficiency. In: Reynolds, M.P., Ortiz-Monasterio, J.I., McNab, A. (Eds.), *Application of Physiology in Wheat Breeding*. CIMMYT, Mexico, 200–207.

Panayotova, G. (1999). Nutrition of durum wheat (*Tr. durum* Desf.) cultivated with cotton in crop rotation, *Ph.D. Thesis*, Sofia.

Panayotova, G. (2001). Response of Durum Wheat Genotypes to Nitrogenous Fertilizers. *Plant Science, 5-6*, 203–207.

Panayotova, G. (2010). Effect of Soil Fertility and Direct Nitrogen Fertilization on the Durum Wheat Varieties in the Conditions of Central Southern Bulgaria. *Proceedings 12th International Symposium "Materials, Methods & Technologies"*, *4*(1), 281–293. June 11-15, Sunny Beach, Bulgaria.

Panayotova, G., and Yanev, Sh. (2001). Response of Durum Wheat Genotypes to Nitrogen Fertilization. *Animal Science*, *6*, 109–111.

Panayotova, G., and Gorbanov, S. (1999). Influence of the fertilization on the properties of durum wheat grain and pasta products. *Bulgarian J. of Agric. Science*, *5*, 425–430.

Panayotova, G., and Valkova, N. (2010). Effect of previous and direct nitrogen fertilization on some grain properties of durum wheat varieties. *Proceedings 14th International Eco-Conference® SAFE FOOD*, 22nd-25th September 2010, Novi Sad, Serbia, 131–138.

Rharrabti, Y., Villegasb, D., Royo, C., Martos-Núñeza, V., García del Moral, L. F. (2003). Durum wheat quality in Mediterranean environments: II. Influence of climatic variables and relationships between quality parameters, *Field Crops Research*, *80*(2), 133–140.

Sanjeev, K., Rajender, K., Harber, S. (2000). Influence of time sowing and NP fertilization on grain quality of macaroni wheat (Triticum durum). *Haryana Agricultural University Journal of Research, 32*(1), 31–33.

Sylvester-Bradley, R., and Kindred, D.R. (2009). Analysing nitrogen responses of cereals to prioritize routes to the improvement of nitrogen use efficiency. *Journal of Experimental Botany*, *60*, 1939–1951.

Tomov, T., Rachovski, G., Kostadinova, S., Manolov, I. (2009). *Handbook of Agrochemistry.* Academic publisher of Agricultural University of Plovdiv, Bulgaria.

Uppal, R., Singh, R., Jagrup, S. (2002). Effect of nitrogen levels and time of application on quality of durum wheat. *Crop Improvement*, *VL*(29), 58–64.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

BARLEY VARIETIES CREATED AND REGISTERED IN ROMANIA DURING 1921-2018 PERIOD

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Abstract

Nowadays barley (Hordeum vulgare L.) has become the fifth crop as importance worldwide. Although barley crop has a high adaptability in different geographic regions, according to FAO data the harvested areas have registered a decreasing trend during last decades at international level, from 83.69 million ha in 1979 to 47.00 million ha in 2017. In Romania, the barley harvested area in the period 1961-2017 registered an increase from 195,500 ha in 1964 to a maximum of 1,017,736 ha in 1991, so that to decrease after that to 455,460 ha in 2017, with a minimum harvested area in the period after 1991 of 317,235 ha in the year 2003.
The objective of the present paper was to analyse the evolution of barley varieties (winter and spring) created and registered in Romania during 1921-2018. In this respect, different informational sources were collected, analysed and a barley varieties database was created since 1960 till 2018.
In Romania, the barley history begins in 1675, when the first mention about barley was made by Nicolae Milescu, but only in 1880 appears some published data about cultivated area. During 1921-2018 period, in Romania has been registered a total number of 149 barley varieties.
Among the winter six-row barley varieties cultivated in Romania, the Miraj variety registered the highest lifespan, being cultivated for 29 years and being cultivated on 1.01 million hectares in 1991. The record lifespan for winter two-row barley was registered by Andreea variety with 23 years, while for spring two-row barley the record was registered by Dvoran variety with 26 years.

***Key words**: barley, varieties, traits, Romania.*

INTRODUCTION

Barley (*Hordeum vulgare* L.) ranks the fifth place as world harvested area, after wheat, maize, rice, and soybeans. It is used as raw material in the malt and beer industry, as feed for livestock, but also as food for human alimentation, either pure or combined with other cereals.
The cereal production (including rice) in the EU was 295.5 million tons in 2018 (about 11.1% of global production). This represented a decrease of 4.6% or 14.4 million tons compared to 2017, due especially to abiotic stress, respectively high-level temperature during the main developmental plant stages registered in many of Central and Southern European regions, while the cultivated areas remained almost the same, being between 55.5 and 55.6 million hectares. To put this in some context, the EU's production of cereals in 2018 was some 37 million tons lower than the record harvest of 332.6 million tons registered in 2014 (Eurostat database).
In the general context described above regarding the cereal situation in EU, the EU production of barley in 2018 was of 56.7 million tonnes, with a decrease of 2 million tons compared to 2017 (58.7 million tons), the lower harvested production reflecting the drought conditions for growing barley.
Barley is one of the most genetically diverse cereals which is categorized as spring or winter types, two-row or six-row, hulled or hulless by the presence or absence of hull tightly adhering to the grain, and malting or feed by end-use type; therefore, breeding programs depend on high level of genetic diversity which provides a significant opportunity for achieving progress (Gozukirmizi & Karlik, 2017). All barley breeding programs have as main objective to develop new genotypes that have some

improvement over existing variety traits like yield potential, resistance to biotic and abiotic stress factors as well as proper end-use quality. Also, one of the main barley breeding objectives is creation of barley varieties suitable for malt and beer production (Matthies et al., 2014) because the key to success in the seed and seed market for malt and beer industry depends to quality parameters of barley variety (Križanova et al., 2010).

The obtained results until present, concerning the latest Romanian winter barley varieties registered in Romanian Official Catalogue of the Varieties and Plant Species, had demonstrated a significant progress regarding the stability of yield potential and also stability of grain quality parameters (Vasilescu et al., 2014). Practically, the Romanian barley breeding program has been developed many barley varieties with increased yielding potential and significant improvements regarding the resistance to biotic and abiotic stress factors.

Usually, after minimum of 8 years from crossing genotypes, breeders choose the best lines depending on their traits (high yield potential, improved quality, presence of different genes for resistance to biotic and abiotic stress factors), and introduce them into the official testing procedures for official registration of that lines in the countries they are testing them. After that, lines have to pass minimum two years DUS (D-distinctiveness, U-uniformity, S-stability) and three years VCU (Value for Cultivation and Use) tests to be allowed for official registration.

The Romanian Official Catalogue of the Varieties and Plant Species represents a real support for farmers, due to a professional and constantly evaluation system for new barley varieties registration, developed to ensure the cultivation of only performant varieties.

Regarding the research methods used to obtain the barley varieties, these are different from a barley breeding program to another. In the present time, there are used two methods: conventional breeding and *bulbosum* method. Bulbosum method consists in the interspecific hybridization of cultivated barley (*Hordeum vulgare* L.) with *Hordeum bulbosum* L. (Kasha & Kao, 1970). Scientists and breeders rapidly adopted the bulbosum method because DH (doubled haploids) provide several advantages over standard selection and breeding methods (Devaux, 2003).

Since 1992, a program for the induction and production of haploids lines was initiated in winter six-row and two-row barley Romanian breeding program by using the *bulbosum* method. The principle was adapted and developed at National Agricultural Research and Development Institute of Fundulea (NARDI Fundulea), Romania, in order to ensure the achievement of absolute genetic uniformity (which corresponds with requested criteria DUS and VCU). This represented an approach to obtain a significant improvement of seed quality parameters.

Compared to the conventional breeding method (based on individual selection), whose cycles amount to 10-12 years, the new method leads to a significant shortening of the duration for obtaining new winter barley varieties, up to 5 years (Bude and Vasilescu, 2007).

In Romania, the high barley level yield and the yield quality are highly dependent on the grown variety and climatic conditions during vegetation period.

The objective of the present paper was to analyse the evolution of barley varieties (winter and spring) created and registered in Romania during 1921-2018.

MATERIALS AND METHODS

The analyse of the barley varieties (winter and spring) created and registered in Romania during 1921-2018 was made by studding the collection of the Romanian Official Catalogue of the Varieties of Plant Species and different documentation sources. The Romanian Official Catalogue is annually updated, and therefore, in order to be registered, all barley genotype needs to achieve preliminary tests (DUS and VCU) to the State Institute for Variety Testing and Registration from Romania.

To accomplish the paper objective, a database was created with all winter barley varieties (two-row and six-row barley varieties), facultative (six-row barley varieties) and spring barley varieties (two-row barley varieties) registered and included into the Romania Official Catalogue of the Varieties of Plant Species (from 1960 up to 2018). The database was helpful to calculate the number of barley varieties (winter, facultative and spring) registered during the entire analysed period, the lifespan of them (number of years that the barley varieties were officially allowed to be cultivated since 1960 up to 2018). Also this gave us the possibility to discover the history of this cereal crop in Romania (Drăghici et al., 1975).

Three main analysed periods were the following:

- 1921-1944 period, which represents the beginnings of agricultural research in Romania due to foundation of Institute of Agronomic Research of Romania (I.C.A.R.) in May 1927. This was the beginning of an unprecedented development of agricultural research in Romania.
- 1945-1980 period, when starting with the year 1962, the activity of creation of winter barley varieties was taken over by the Research Institute for Cereal and Industrial Crops from Fundulea - ICCPT Fundulea (the present National Agricultural Research and Development Institute from Fundulea - NARDI Fundulea), and in 1968 the first winter barley variety was obtained (winter six-row barley variety namely Intensiv 1).
- 1981-2018 period, when in barley breeding program there was used a new barley germplasm from different continents, and the registered barley varieties as number and origin drastically has changed. In 2007, Romania joined the European Union and all the new winter and spring barley registered varieties had to fulfill distinctiveness, uniformity and stability criteria.

RESULTS AND DISCUSSIONS

Period 1921-1944

At the beginning of the 10th century, there was a significant demographic increase that has led to the development of agriculture in the southeastern areas of the Carpathians. However, the first mentions referring to the barley crop appear in the notes of Nicolae Milescu from 1675-1678.

Another mention was made in 1868 by Ion Ionescu de la Brad, stating that although the soil provids good growing conditions for millet, barley and oat crops, the areas cultivated with them did not provide the necessary productios (Drăghici et al., 1975).

Petre Sebeşanu Aurelian mentioned in 1875 that barley ranks the 3rd place as cultivated area in Romania, after wheat and maize, at that time being cultivated on 250,000 hectares.

The beginnings of agricultural research in Romania, from the second half of the 19th century and especially at the beginning of the 20th century, were due to the great agronomists such as Gheorghe Ionescu-Şişeşti, Teodor Seidel, Traian Săvulescu, Wilhelm Karl W. Knechtel and many others who have integrated Romanian agricultural sciences into the modernization current that was manifested at that time throughout the world. On this basis, the Institute of Agronomic Research of Romania (I.C.A.R.) was established by law no. 1205, voted on 4 May 1927.

In 1927, Nowacki mentioned that during 1922-1926 period, barley was cultivated on an area of over 1.7 million hectares, ranking third place after maize and wheat. Another important mention was that barley areas were occupied 94.5% with spring varieties and only 5.5% were sown with winter varieties (Nowacki, 1927). These data are confirmed by Munteanu (1929), who also mantioned that the most cultivated varieties at that time were Chevalier, Hanna and Imperial, these varieties being the best acclimatized to the Romanian conditions.

In the period preceding the establishment of ICAR (1921-1927), Walter Mader and Friedrich Dotzler created at Cenad farm, from Timiş county (located into West of Romania), the first barley Romanian varieties, respectively the spring two-row varieties named Sămînţa nr. 3 and Cenad 375 and the winter six-row varieties named Sămînţa nr. 112, Cenad 395 and Extensiv 1. These varieties were created by individual selection from the Banat local populations (Bude & Vasilescu, 2007)

In the decade 1930-1940, maize and some technical plants expanded extensively in Romania. As a consequence, the areas cultivated with barley fell in order to reach 839,200 ha in the period 1934-1938, preserving approximately the same proportion between the spring and winter cultivated barley. At that time, an average yield of 720 kg/ha was achieved, with an annual production of about 600,000 tonnes (Drăghici et al., 1975).

Both in the years of World War II and thereafter, from a quantitative point of view, a little quantity of seeds of the cultivated varieties was produced, and therefore the barley cultivated areas decreased drastically.
In 1940, at the Experimental Stations Cluj and Câmpia Turzii, there was created the spring two-row barley variety named Cluj 123. Two years later, in the Iași county, there was created the spring two-row barley variety Tg. Frumos 240. In the same year, at Cenad Experimental Station, a facultative six-row barley variety was created by Ecaterina Constantinescu, namely Cenad 396, which represented a cross between Cenad 395 and Extensiv 1 varieties.

Period 1945-1980
In 1949, at the Breeding Plant Section from the Research Agricultural Institute of Bucharest, there was created the spring barley variety with four rows, namely I.C.A.R. 143.
In 1959, at Cenad Experimental Station, Şiclovan obtained the six-row barley variety named Cenad 345 (facultative barley variety). This variety was characterised by increased resistance to frost, which led to the expansion of the winter barley in Romania. Also, in 1960 there was registered a new spring two-row barley variety, named Perfecta.
The success of the Fundulea Maize Research Institute's initiatives, with direct and immediate economic effects in the Romanian agriculture, has created a tendency to gradually expand the concerns of this Institute.
In 1961, on May 22, the Fundulea Maize Research Institute was unified with the Institute of Agronomic Research of Romania, forming the Institute of Agricultural Research (ICA Bucharest-Fundulea), which becomes in May 1962 (by law no. 1/1962, art. 26) Research Institute for Cereals and Industrial Plants, located in Fundulea, Călărași county (ICCPT Fundulea). The Institute was subordinated to the Central Institute for Agricultural Research (ICCA), as the head of all agricultural research in Romania, a function subsequently taken over by the Academy of Agricultural and Forestry Sciences (ASAS, 1969).
Starting with the year 1962, the activity of creation of varieties of winter barley was taken over by ICCPT Fundulea, and in 1968 the first winter six-row barley, named Intensiv 1, was obtained. This barley variety was the most widespread in Romania and occupied over 80% of the cultivated area with winter barley. Two years earlier (1966), there were registered in the Romania Official Catalogue of the Varieties of Plant Species another two spring varieties, Karlsberg and Proctor.
During 1971-1974 period, in the Romanian Official Catalogue of the Varieties of Plant Species, there were registered seven winter six-row barley varieties (Caracal 6, Cluj 230, Ager, Intensiv 2, Miraj, Nr. 1050, and Pamina), five winter two-row varieties (Beta with two rows, Gloria, Sofia 3, Sofia 4, and Azuga) and five spring two-row variety (Dvoran, Alsa, Elgina, MK 42, and Taplani).
In 1973, the first six-row facultative barley variety was created at ICCPT Fundulea, this being named Intensiv 2. In comparison to the Intensiv 1 barley variety, this was imposed into practice because it could be sown in the spring as well, this variety occupying at that time about 5% from the total cultivated barley area in Romania.
Azuga is the first local winter two-row variety created by ICCPT Fundulea by crossing a winter two-row barley variety with a spring two-row barley variety. In the same year (1974), there was obtained the winter six-row barley variety named Miraj, both varieties being introduced into production, respectively they were started to be cultivated in 1974.
Azuga variety was characterized by superior technological quality, but the level of resistance to frost was lower, this being recommended for cultivation only in the plain areas of the western part of Romania, as well as in Muntenia and Oltenia regions. On the other hand, Miraj variety, only 5 years after its approval, has replaced all the other barley varieties usualy sown in the autumn.
In 1980, Miraj variety was cultivated on over 900,000 hectares, this representing the largest cultivated area by a winter six-row barley variety until present days. The generalization of the Miraj variety over the entire Romanian area cultivated with barley in 5 years since its approval has been a remarkable contribution to the agricultural production increase. At the same time, the Miraj variety was grown on large areas in countries such as Bulgaria and Turkey.

In the 1975-1980 period, only two winter six-row barley variety were registered (Robur and Valja), one winter two-row barley variety in 1977, named Victoria (which has achieved an increased yield level above the Azuga variety), eight spring two-row barley varieties (Ametist, Triumpf, Peast, Belfor, Pauline, Favorit, Rapid, and Spartan) and 4 winter two-row varieties (Kelibia, Novosadski 290, Kristal, and Ladoga).

In 1961, the barley cultivated area in Romania was of 284,000 ha and since 1965 up to 1980 it has been registered an increase from 232,771 ha to 809,457 ha (Figure 1).

An historical barley cultivated area in Romania was registered in 1981 and 1982 with 916,975 ha and 943,061 ha, respectively, followed then by a slight decrease.

The average barley yield (Figure 2) also increased from 1,648 kg/ha to a maximum of 3,193 kg/ha in 1978 (Eurostat database).

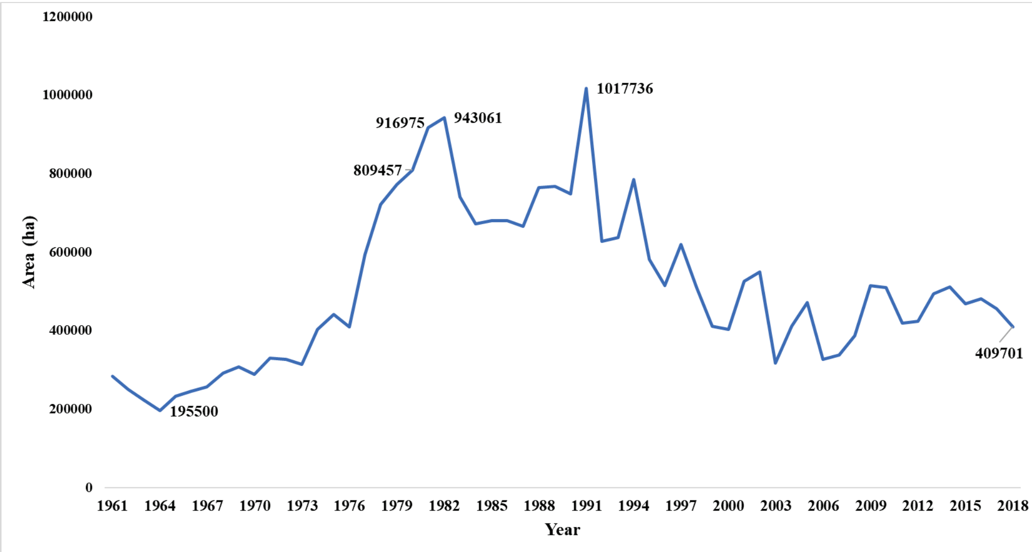

Figure 1. Evolution of barley cultivated area in Romania, during the 1961-2018 period

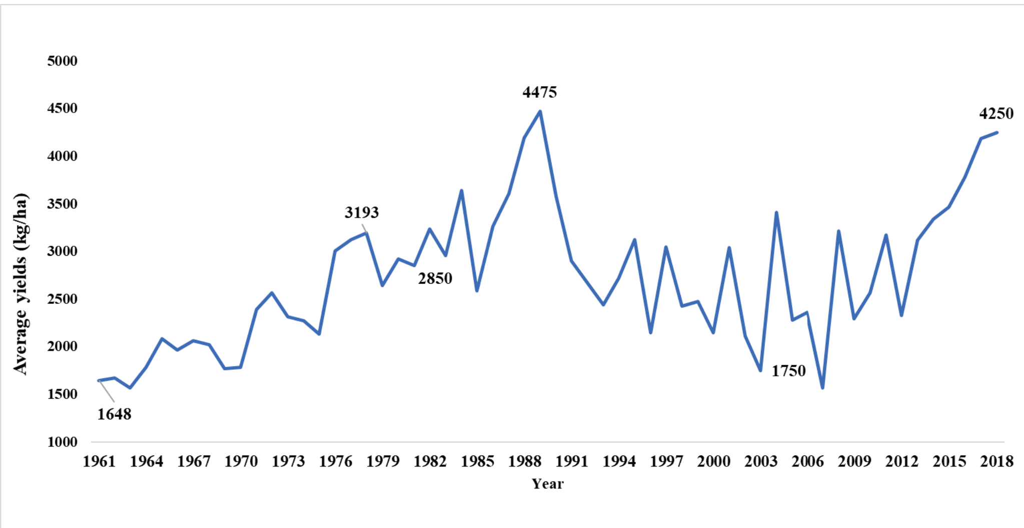

Figure 2. Evolution of barley average yield in Romania, during the 1961-2018 period

Regarding the number of registered barley varieties in the Romanian Official Catalogue of the Varieties of Plant Species, during the analysed period, there were registered a number of 15 spring two-row varieties, 7 winter two-row varieties and 12 winter six-row varieties.

Period 1981-2018

During this period, there were registered more spring varieties comparing with winter barley six and two-row varieties.

In 1981, five barley varieties were registered, respectively: the winter six-row barley varieties Productiv and Novisad 27; the winter two-row barley variety Grivița; the spring two-row barley varieties Grit and Koral. The winter six-row barley variety Productiv, with an increased yield potential by more than 150 kg/ha, represented the first step in valuing barley North American germplasm. The achievements in terms of the yield potential of the existing varieties have also generated negative effects in the malt and beer processing industry due to the unevenness of the grains, an essential condition for the malting process, so the improvement targets are suddenly directed towards improving two quality parameters, the size and uniformity of the grains. Also, in the same year, the breeding works brought satisfactory results in the winter two-row barley, combining the precocity with the size of the grains in the winter two-row variety Grivița.

In 1986, there was registered the winter six-row variety Precoce, which despite the fact that it did not record so high yield it was highlighted by superior grain size to the other barley varieties homologated until that time.

In 1992, by individual selection of the Grivița variety and by making an improvement of the resistance to frost and drought, Laura variety was registered, this achieving a yield increase by 7%.

In 1993, winter six-row barley variety Dana was registered, this showing a yield increase by 8% over Miraj variety. Since then up to date, due to the anthocyanin colour of the awn but also to a high yielding capacity, Dana variety was used as check in most of the breeding works. The winter six-row barley variety Amical (previous Adi) was registered in the same year, and due to its resistance to lodging, it achieved 8-10% higher yield than Miraj variety.

In 1994, the winter two-row barley variety Andreea was registered, this possessing both yield increases due to a higher tilling capacity and quality parameters for raw industry (malt and beer). Also in 1994, by hybridization between the Miraj variety and a Mexican line, followed by individual selection, there was obtain and registered the winter six-row barley variety Mădălin, which is characterized by harvest stability and resistance to the main foliar barley disease. In 18 crop growing conditions, Mădălin variety obtained a higher yield by 8-9% than the check.

In 1996, the Sistem (previous Orizont) winter six-row barley variety was obtained by the same method as Mădălin variety. Sistem variety is characterised by shorter straw that gives to it good resistance to lodging.

In 1998, the Compact winter six-row barley variety was registered, this having dense spikes and being the only local variety belonging to the *paralellum* variety. Also in 1998, the Productiv variety and a German line whose genes with resistance to thermic and hydric stress was combined harmoniously in the winter six-row barley variety Andrei, which in the 14 experimental stations, located in all areas of barley cultivation all over Romania registered a yield increase of nearly 500 kg/ha compared to Dana variety.

Registered in 2000 and tested in 14 agricultural research stations, the winter six-row barley variety Mareșal (previous Regal) confirms the high yield potential and a good resistance to foliar diseases.

Uniformity of grains, yield stability, resistance to main foliar diseases, reduction of growing period, all these attributes are found in facultative six-row barley variety Cardinal FD, registered in 2003 and obtained from a local line combined with a German line. Cardinal FD variety confirmed a yield potential increase by 9-10% over the Dana variety check. One year later (2004), the winter six-row barley variety Univers was registered, this being tested in 42 conditions in 14 different locations, and showing an increase yield by 4% to its successor Dana variety.

During 2012-2017 period, three six-row barley varieties (Ametist, Smarald, and Simbol) were registered in the Romanian Official Catalogue of the Varieties of Plant Species, these having improved agronomic traits. The winter six-row barley variety Ametist (registered in 2012) represents a genetic progress with an increase of grain weight by 2 units compared to other barley genotypes with six rows. Ametist variety combines the size of grains with a superior level of average starch content, with high yield performance (7.5% increase in yield compared to winter two-row barley variety Andreea). The winter two-row barley variety Artemis (registered in 2012) represents a progress regarding the tolerance to pathogenic agent *Pyrenophora teres* f. *teres* and resistance to lodging.

The facultative six-row barley variety Smarald (registered in 2013) is the first barley variety created by biotechnological method *bulbosum*, which has a remarkable adaptability but also improved resistance to lodging and hydric stress. It is the first variety with a superior stability of quality parameters (average starch content 62.7% and average protein content 11.0%), being suitable as raw material in beer industry.

Winter six-row barley variety Symbol (registered in 2015) is tolerant to *Pyrenophora teres* f. *teres* pathogen and is distinguished by the particular capacity to achieve high yields at lower plant densities. It has a special flexibility regarding the sowing season, being well adapted to later sowing (in first decade of November), producing higher yields than other Romanian barley varieties under such conditions.

In 2017, two winter barley varieties were registered, respectively the winter six-row barley variety Onix and the winter two-row barley variety Gabriela (the second variety obtained by *bulbosum* method). A year later (2018), another winter six-row barley variety was registered, named Lucian.

Besides the mentioned varieties, according to Romanian Official Catalogue of the Varieties of Plant Species there were registered 14 winter six-row barley varieties, 25 winter two-row barley varieties and 45 spring two-row barley varieties.

In the period 1971-2018 (Figure 3), the number of winter six-row barley varieties registered annually in Romania, varied between 2 (1982, 1983, and 2007 year) and 16 (2008 year).

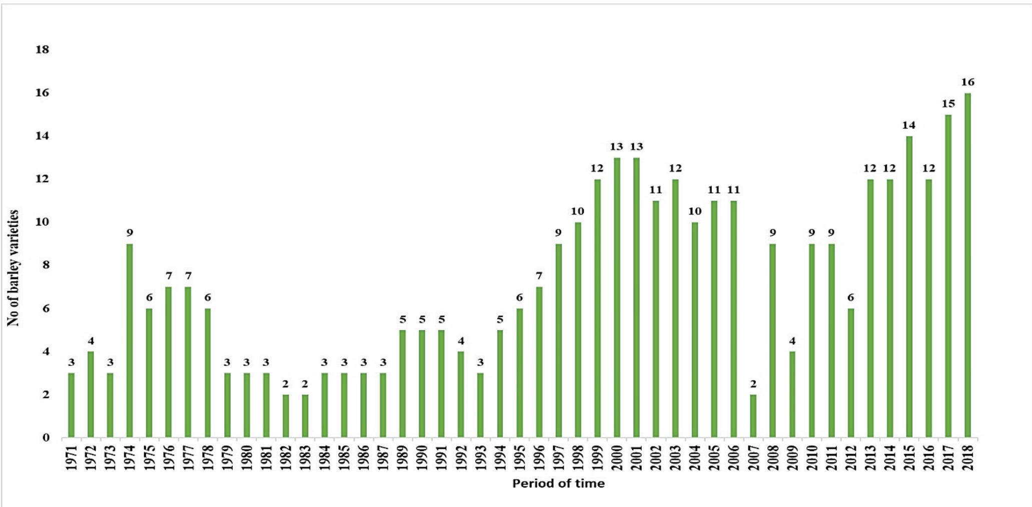

Figure 3. Number of the winter six-row barley varieties registered to be cultivated annually in Romania (included in the Official Catalogues of the Varieties of Plant Species)

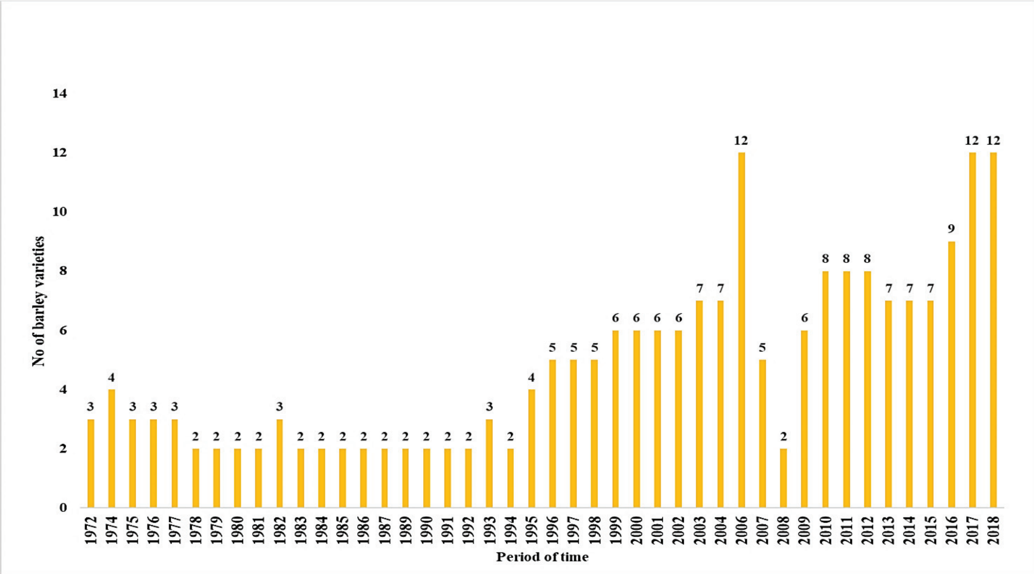

Figure 4. Number of the winter two-row barley varieties registered to be cultivated annually in Romania (included in the Official Catalogues of the Varieties of Plant Species)

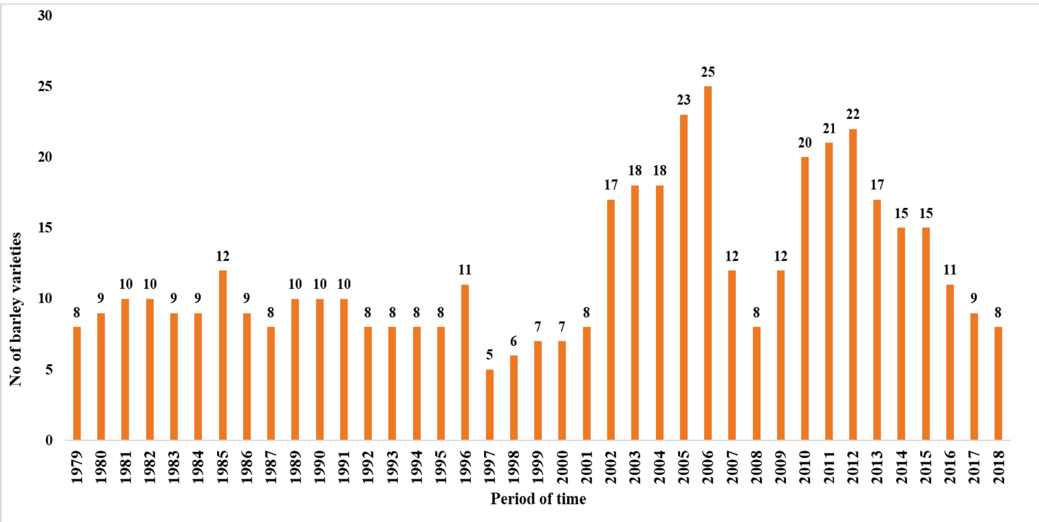

Figure 5. Number of the spring two-row barley varieties registered to be cultivated annually in Romania (included in the Official Catalogues of the Varieties of Plant Species)

During 1972-2018 period, the number of winter two-row barley varieties registered annually in Romania was between 2 and 12 (Figure 4), meanwhile the number of spring two-row barley varieties ranged between 5 and 25 (Figure 5). A decrease of number of barley varieties occurred in 2007 year when Romania joined the European Union.

The highest barley cultivated area was registered in 1991, with 1,017,736 ha (Figure 1), when the Romanian winter six-row barley variety Miraj was sown on about 900,000 ha. The highest historical average yield at barley in Romania was registered in 1989, with 4,475 kg/ha (Figure 2). Since then, the average yield varied from year to year. Thus, in 2003, due to lower temperatures and lack of snow cover during winter season, the obtained average yield was only of 1,750 kg/ha. But, the lowest historical average yield at barley in Romania of 1,568 kg/ha was registered in 2007, this year being characterised as one of the most drought year in the cereal vegetation period. In 2018, Romania's production of barley was of 1.8 million tons with a constant increase of total production, the average for the last 10 years being of 1.3 million tons, this representing 3% of total EU barley production (Agriculture, Forestry and Fishery Statistical Book, 2018). In 2018, the cultivated area with barley in Romania was of 409,701 ha (Figure 1), which represents 40.3% from the maximum cultivated area with barley in 1991 (Eurostat database), while the average yield has increased to 4,250 kg/ha (Figure 2). Comparing the obtained results in 1945-1980 analysed period, the number of barley varieties registered was 46, with an average lifespan between 5.3 and 9.6 years. Regarding 1981-2018 period, the number of barley varieties increased to 96 and also the average lifespan for winter six-row and two-row barley varieties, meanwhile for spring two-row barley varieties decreased.

For winter six-row barley varieties, the lifespan was in average of 9.1 years, with a variation from 1 to 29 years (Figure 6). The variety with the highest lifespan was Miraj (29 years).

For winter two-row barley varieties, the lifespan was in average of 5.3 years, with a variation from 2 to 23 years (Figure 7). The variety with the highest lifespan was Andreea (23 years).

For spring two-row barley varieties, the lifespan was in average of 9.6 years, with a variation from 2 to 26 years (Figure 8). The variety with the highest lifespan was Dvoran (26 years).

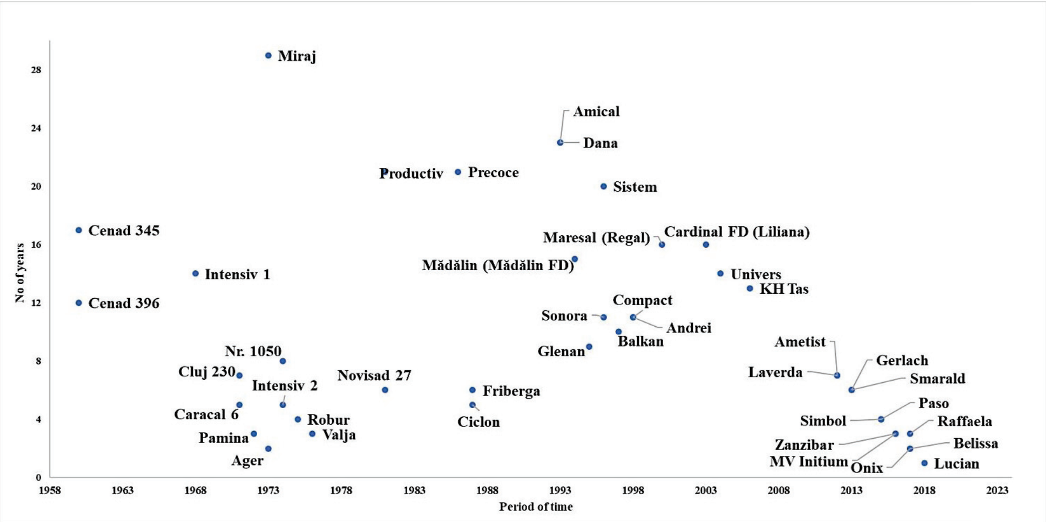

Figure 6. Lifespan of winter six-row barley varieties registered in the Romanian Official Catalogues of the Varieties of Plant Species

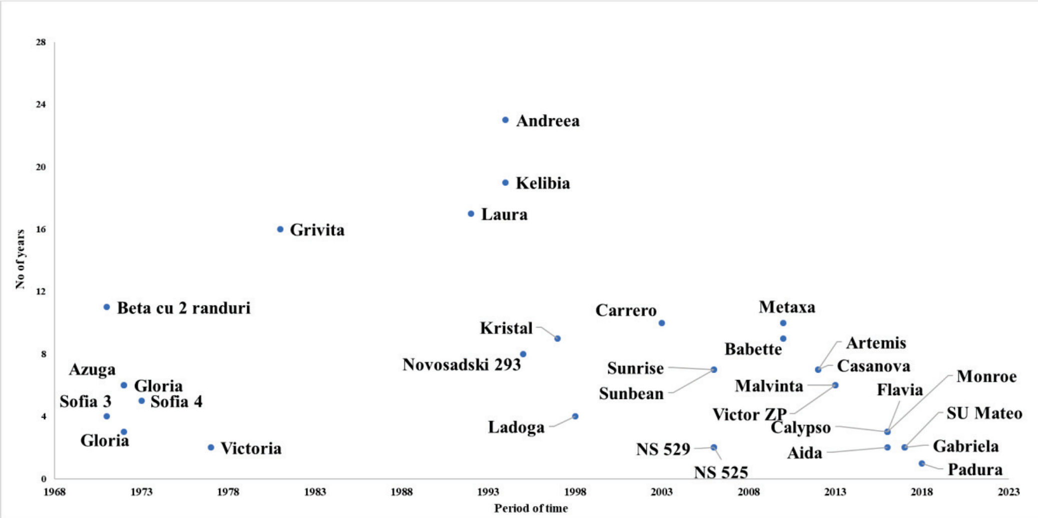

Figure 7. Lifespan of winter two-row barley varieties registered in the Romanian Official Catalogues of the Varieties of Plant Species

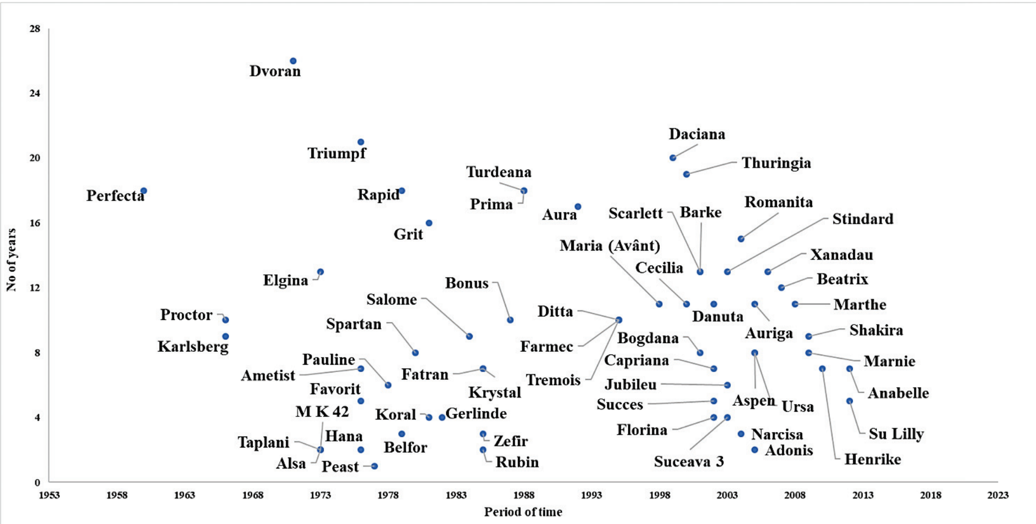

Figure 8. Lifespan of spring two-row barley varieties registerd in the Romanian Official Catalogues of the Varieties of Plant Species

CONCLUSIONS

In Romania, the barley history begins in 1675, when the first mention about barley was made by Nicolae Milescu, but only in 1880 appears some published data about the cultivated area. The registration of 8 barley varieties (4 spring varieties, 3 winter varieties, and 1 facultative variety) during 1921-1944 period, marked an important step in Romanian barley breeding activity. After obtaining these barley varieties, since 1945 until 1980 the evolution of the barley cultivated area, the number of registered varieties and their type have oscillated.

During 1921-2018 period, in Romania has been registered a total number of 149 barley varieties, out of which: 42 (28.2%) winter six-row barley varieties, 4 (2.7%) facultative six-row barley varieties, 32 (21.5%) winter two-row barley varieties, and 71 (47.6%) spring two-row barley varieties.

In 1921-1944 period, there were registered 8 barley autochthonous varieties (winter and facultative six-row barley, spring two-row barley), then in the 1945-1980 period, there were registered 46 varieties (9 winter six-row varieties, 3 facultative six-row varieties, 14 winter two-row varieties, and 20 spring two-row varieties).

In 1981-2018 period, the total number of barley varieties registered was of 95. On the first place, there was situated the spring two-row barley with 47 registered varieties, while winter six-row barley ranked the second place with 30 registered varieties, followed by winter two-row barley with 18 registered varieties.

Among the winter six-row barley cultivated varieties in Romania, the Miraj variety registered the highest lifespan, being cultivated for 29 years, this being followed by Dana, Amical, Productiv, Precoce, and Cardinal FD varieties. Also, Miraj variety registered the highest cultivated area in Romania, respectively 1.01 million hectares in 1991.

The record lifespan for winter two-row barley belongs to Andreea variety with 23 years, followed by varieties Laura with 17 years and Grivița with 16 years.

The lifespan of spring two-row barley varieties was closely to above mentioned data, Dvoran variety being cultivated for 26 years, Triumpf for 21 years and Daciana for 20 years.

The average lifespan of the winter six and two-row barley varieties, increased during the last 38 years, with 1 and 1.1 years respectively, meanwhile the average lifespan for spring two-row barley decreased with 1.2 years.

REFERENCES

Agriculture, Forestry and Fishery Statistical Book, 2018. Retrieved from *ttps://ec.europa.eu/eurostat*

Bude, Al., Vasilescu, L. (2007). Rezultate obținute în ameliorarea orzului de toamnă la Fundulea. *Analele I.N.C.D.A. Fundulea, LXXV*, 83–93.

Devaux, P. (2003). The *Hordeum bulbosum* (L.) method. In M. Maluszynski et al. (Ed.), Doubled Haploid Production in Crop Plants (15–19). IAEA.

Drăghici, L., Bude, Al., Sipoș, Gh., Tușa, C. (1975). *Orzul*. Editura Academiei Republicii Socialiste România.

Eurostat database, https://ec.europa.eu.

FAO database, http://www.fao.org/faostat/en

Gozukirmizi, N., Karlik, E. (2017). Barley (*Hordeum vulgare* L.) Improvement Past, Present and Future. In Makoto Kanauchi (Ed.), *Brewing Technology* (49–78), IntechOpen.

INCDA Fundulea, 2017. 60 de ani de cercetare ştiinţifică în sprijinul agriculturii, I.C.C.P.-I.N.C.D.A. Fundulea, 1957-2017. Editura Total Publishing, pp. 28–30.

Kasha, K. J., Kao, K. N. (1970). High Frequency Haploid Production in Barley (*Hordeum vulgare* L.). *Nature, 225*, 874–876.

Križanova, K., Psota, V., Sleziak, Ľ., Žofajova, A., Gubiš, J. (2010). Spring Barley Breeding for Malting Quality. *Potravinarstvo, 4*(2), 39–44.

Matthies, I.E., Malosetti, M. Roder, M.S., van Eeuwijk, F. (2014). Genome Wide Association Mapping for Kernel and Malting Quality Traits Using Historical European Barley Records. *PLoS ONE 9*(11), e110046.

Munteanu, A. (1929). Production végétale. En: *La Roumanie agricole* (pp. 88-93). XIVme Congrès International d'Agriculture, Bucarest, 7, 8 et 10 Juin.

Nowacki, A. (1927). *Cultivarea cerealelor*. Cartea Românească, Cluj.

Petre Sebeşanu, A. (1875). *Terra nostra. Schiţe economice asupra României*. Typographia Laboratorilor Romani, București.

Vasilescu, L., Alionte, E., Bude, Al. (2014). Comportarea unor soiuri și linii de orz de toamnă la INCDA Fundulea în perioada 2008-2013, sub aspectul stabilității producției și calității. *Analele I.N.C.D.A. Fundulea, LXXXII*, 69–82.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

IDENTIFICATION OF VARIABILITY IN VEGETATIVE GROWTH OF SOME WINTER WHEAT VARIETIES UNDER ECOLOGICAL AGRICULTURE WITH NDVI

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Abstract

Early recognizing of plant growth variability can aid in identifying yield-limiting factors such as genotype, soils, nutrient availability and/or environmental limitations.
Normalized Difference Vegetation Index (NDVI) is considered as a potential screening tool for estimation of grain yield in wheat. NDVI has been associated with percent ground cover, leaf area index (LAI), biomass accumulation, and nitrogen use efficiency. In this study we use NDVI to established variability in growth of several winter wheat genotypes and relationships with grain yield under ecological agriculture system. For recording the value of NDVI, Green seeker (Trimble) was used. NDVI values were measured at different stage of vegetations through the growing seasons of 2017-2018 in winter wheat (Triticum aestivum L.) grown at southeast part of Romania (Fundulea) on cambic chernozem soil and 1-2-3-4 years alfalfa stands. Results showed differences in growth of different winter wheat genotypes could be identified with NDVI index. At the stage of tillering, stem elongation and anthesis the relationship between LAI and NDVI is positive and linear while during at grain filling and maturity stage were not correlations. The relationship between NDVI and grain yield was also established. There was the simple correlation between grain yield and NDVI scores at the time of tillering, stem elongation and anthesis, in both seasons, and insignificant association between grain yield and NDVI was also found at grain filling and maturity stage. This demonstrated the opportunity to use this index in characterizing production potential of different winter wheat cultivars under ecological agriculture system.

Key words*: vegetative growth, winter wheat, ecological agriculture, normalized difference vegetative index.*

INTRODUCTION

NDVI is a satellite product that measures the vigour and greenness of vegetation on the earth's surface. It is calculated as the ratio of visible spectral wave bands to near-infrared spectral wave bands. Healthy, green vegetation has a high presence of chlorophyll pigment, which causes low reflectance in visible wave bands and high reflectance in near-infrared wave bands. The reverse is true in vegetation under stress. NDVI is a unitless index, with values ranging from −1 to 1. Healthy vegetation has the highest positive values, while bare soil, water, snow, ice, or clouds have NDVI values of zero or that are slightly negative (Mkhabela et al., 2005). In present there are many portable instruments which are able to measure NDVI. This could be used as a potential screening tool for estimation of grain yield in wheat and many studies were performed in this sense especially under conventional agriculture system. In this study we use NDVI index to established variability in growth of two winter wheat genotypes in culture pure and two mixtures of these and relationships with grain yield under ecological agriculture system.

MATERIALS AND METHODS

The experiments were carried out at National Agricultural Research and Development Institute Fundulea, Ecological Research Center, during 2017-2018, on cambic chernozem soil. The experiment was with randomized complete block design (RCBD) with split plot arrangement having four replications. The plot size was 15 m × 1.5 m. The experimental variants were: year, cultivar and different year alfalfa stands. We used two cultivars (Glosa and Izvor) as pure culture and two mixtures of these cultivars (50% Glosa + 50 Izvor and 25% Glosa and 75% Izvor) and as previous plants was soybean and 1-2-3-4 years old alfalfa stands.

Normalized Difference Vegetation Index (NDVI) was measured by a spectroradiometer (Green-Seeker Hand Held Crop Sensor, Trimble unit), above the canopy at 50 cm height at different growth stages (tillering, stem elongation, anthesis, grain filling, and maturity - milk).
The leaf area was record with leaf area meter (LAI 2000.).
The yield was determined by weighing the seeds after harvesting

RESULTS AND DISCUSSIONS

The years of experimentation were totally different from the viewpoint of quantity and monthly repartitions of rainfall. In 2017, the cumulated rainfall during April - June exceeded with 55 mm the normal of the zone (180.6 mm), suggesting favourable conditions for winter wheat crop.
But the cumulated rainfall during June exceeded only with 22.1 mm the normal of the zone (74.3 mm), suggesting more little favourable conditions for grain filling than in 2018 (Figure 1).

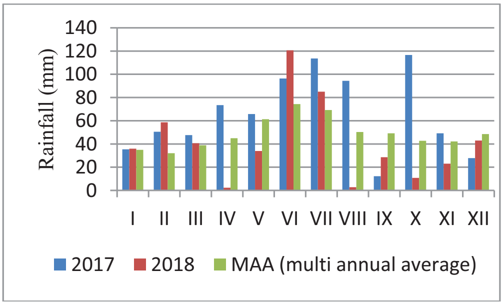

Figure 1. Rainfall during years of experimentations

Concerning temperatures, in 2018, the average of temperature during April and May exceeded the normal of the zone with 4.6°C and 2.5°C respectively, while in 2017 in the same period the temperatures were lower by 0.6 and 0.2°C respectively (Figure 2).
In 2018, the moisture deficits from April up to May created unfavourable conditions during stem elongations and reproductive organs appearance but the cumulated rainfall during June exceeded with 46.3 mm the normal of the zone (74.3 mm), suggesting favourable conditions for grain filling (Figure 1).

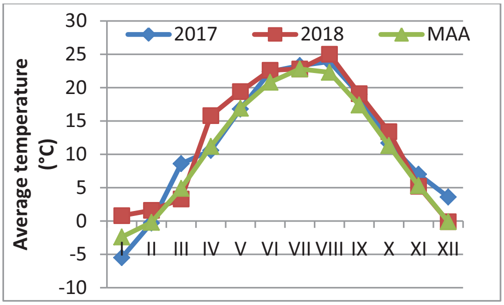

Figure 2. Monthly temperature during years of experimentations and multi annual temperatures (from 1960 to 2018)

The values of leaf area index LAI were the highest in 2018 year for all cultivars. The lowest value was recorded by all cultivar at maturity. The peak LAI was the highest for Glosa cultivar at anthesis in 2018 year conditions and lowest value was recorded by the same cultivar at maturity. During 2017 the highest values was reached by Mixture 2 at all stage of vegetations (except stem elongation) (Table 1).

Table 1. The dynamics of leaf area index for winter wheat cultivars and mixtures at different growth stages during 2017 and 2018 conditions

Winter wheat cultivars and mixtures	Tillering	Stem elongation	Anthesis	Grain Filling	Maturity
2017					
Glosa	1.35	2.75	2.71	2.92	1.31
Izvor	1.25	2.55	2.69	2.15	1.28
Mixture 1 (75% Izvor+25% Glosa)	1.33	2.5	2.75	3.12	1.54
Mixture 2 (50 % iz + 50% Glosa)	1.38	2.49	2.81	3.05	1.51
2018					
Glosa	1.85	2.85	3.21	1.42	0.45
Izvor	1.73	2.65	2.95	0.98	0.82
Mixture 1 (75% Izvor+25% Glosa)	1.8	2.73	2.98	1.51	1.13
Mixture 2 (50 % iz + 50% Glosa)	1.78	2.56	2.98	1.42	0.95

The NDVI reached a maximum during anthesis and started to decline from this date onwards (Table 2).
Many researchers concluded that increases in red reflectance were related to the decreases in chlorophyll content resulting from lower N supply, decreases in NIR reflectance mostly responded to decreases in LAI and green

biomass, as has been widely reported for wheat crops, (Jensen et al., 1990; Fiela et al., 1995).

Table 2. Effect of different wheat cultivars and mixtures on NDVI score at different growth stages

Winter wheat cultivars and mixtures	Tillering	Stem elongation	Anthesis	Grain Filling	Maturity
2017					
Glosa	0.37	0.52	0.53	0.50	0.31
Izvor	0.33	0.47	0.51	0.48	0.28
Mixture 1 (75% Izvor+25% Glosa)	0.42	0.57	0.55	0.50	0.3
Mixture 2 (50 % iz + 50% Glosa)	0.44	0.48	0.56	0.48	0.3
2018					
Glosa	0.6	0.68	0.78	0.42	0.34
Izvor	0.53	0.62	0.76	0.43	0.33
Mixture 1 (75% Izvor+25% Glosa)	0.53	0.58	0.75	0.51	0.22
Mixture 2 (50 % izvor + 50% Glosa)	0.54	0.5	0.72	0.42	0.31

We can observe that at grain filling stage NDVI decreases only with 0.08 in 2017 while during 2018 reached up to 0.42, because crop becomes under stressed and its capacity to absorb PAR is reduced. Fernandez et al. (1994) described that NDVI score reached up to 0.4 in productive environments which have high LAI thus showing the vigorous crop canopy as dark foliage.

At the stage of tillering, stem elongation and anthesis the relationship between LAI and NDVI is positive and linear (r = 0.95, 0.68, 0.92) (Figures 3, 4 and 5) while during the grain filling and maturity stage were not correlations, as Figures 6 and 7 shows.

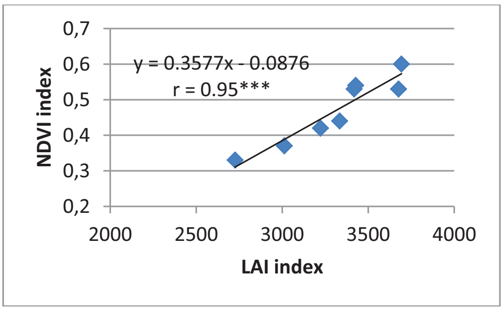

Figure 3. The relationships between leaf area and NDVI index during tillering stage in years of experimentations

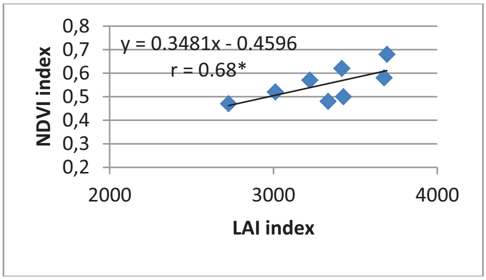

Figure 4. The relationships between leaf area and NDVI index during stem elongation stage in years of experimentations

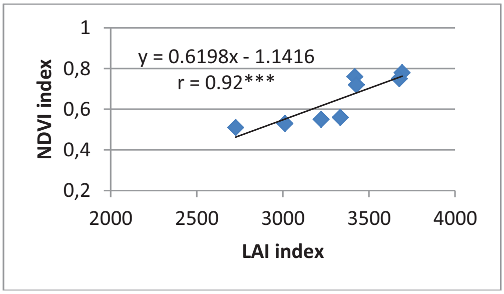

Figure 5. The relationships between leaf area and NDVI index during anthesis stage in years of experimentations

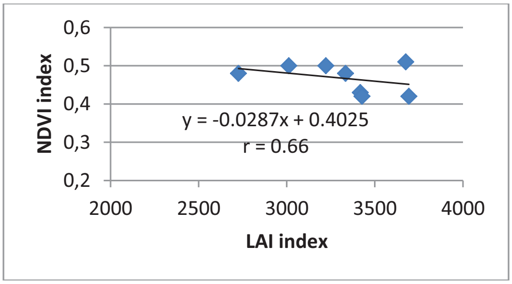

Figure 6. The relationships between leaf area and NDVI index during grain filling stage in years of experimentations

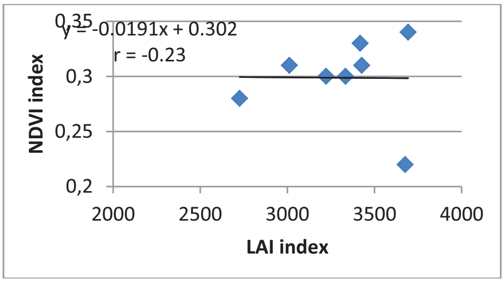

Figure 7. The relationships between leaf area and NDVI index during maturity stage in years of experimentations

Our results are in concordance with onother studies which shown that a variation in LAI implies different intercepted radiation that, according to the Radiation Use Efficiency (RUE), is directly related to the production of biomass that will determine the possible yield (Carcova et al., 2003).
Martin and Heilman (1986) found that spectral vegetation indices were highly correlated with LAI at small research plots.
Other studies at paddy rice also showed that there were statistically sign cant correlations between LAI and spectral vegetation indices derived from the NOAA Advanced Very High Resolution Radiometer (AVHRR) sensors (Lu 1997, Zhao et al. 1993; 1996)
The analysis of variance for NDVI shows the very significant influence of the years, the genotypes but also their interaction on this character. The highest variance being given by factor B (year) (Table 3).

Table 3. Analyses of variance for NDVI

Source of variance	Degree of freedom	Mean square	S2	F factor and significance
Factor A (cultivar)	3	0.01715	0.00571	8.65***
Error	9	0.0059	0.00066	
Factor B (year)	1	0.2182	0.2182	187.27***
A x B	3	0.0275	0.00919	7.89***
Error	12	0.0139	0.00116	

***significant differences for P<0.001

Concerning the yield, analysis of variance revealed the very positive effect of the cultivars in both years, while under moderate stress (2018) there was a significant difference (P<0.001) for yield between studied old alfalfa stands (Table 4).

Table 4. Analysis of variance for the yield

Source of variance	Degree of freedom	F factor and significance 2017	F factor and significance 2018
Factor A (cultivar)	3	5.98***	19.98**
Error	9		
Factor B (old alfalfa stands)	3	2.86	23.80***
A x B	9	0.69	0.12
Error	36		

Glosa in pure culture and mixture 1 and 2 showed higher grain yield in 2018 under 3 years old alfalfa stand (Figure 8).

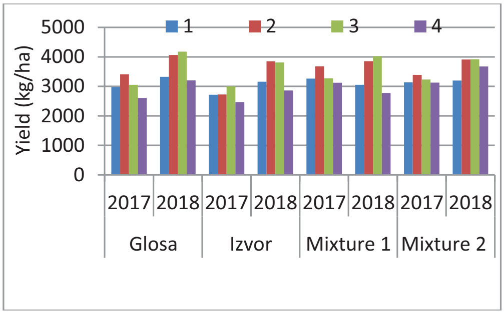

Figure 8. The yield of studied cultivars under 1-2-3-4 year old alfalfa stands. Fundulea 2017 and 2018

Table 5 shows the simple correlation between grain yield and NDVI scores. At the time of tillering, stem elongation and anthesis, these relationships were highly positively correlated in both seasons, and insignificant association between grain yield and NDVI was also found at grain filling and maturity stage.
Other study in conventional agriculture system showed a clear association between grain yield and NDVI measured but at maturity stage correlation between grain yield and NDVI was greater than NDVI values recorded at different growth stages (Syeda Refat et al., 2014).

Table 5. The relationships between NDVI scores at different stage of vegetation and the yield

Stage of vegetation	Coefficient of correlation (r) between yield x NDVI
Tillering	r = 0.93***
Stem elongations	r = 0.68**
Anthesis	r = 0.86***
Grain filling	r = 0.36
Maturity (milk stage)	r = -0.0097

***significant differences for P<0.001
**significant differences for P<0.01

The statistical analysis indicated there was a significant difference (P<0.001) for NDVI index between compared cultivars and previous crop. The variation due to the old alfalfa stands (factor B) was greater than that due to the cultivar (factor A) or the interaction of the two factors in both years of experimentation (Table 6).

Table 6. Analysis of variance for NDVI index at stem elongation

Source of variance	Degree of freedom	F factor and significance 2017	F factor and significance 2018
Factor A (cultivar)	3	20.64***	6.37**
Error	9		
Factor B (old alfalfa stands)	3	47.33***	71.90***
A x B	9	4.79***	1.38
Error	36		

***significant differences for P<0.001
**significant differences for P<0.01

The higher values of NDVI was reached by Glosa cultivar under 2 and 3 years old alfalfa stands in both years of experimentations. The higher temperatures from spring of 2017 (4.6°C over the normal of the zone in April) accelerated the growth of the plants which explains the higher values of NDVI in this year for all studied cultivar. Also, we can have observed that the higher values of NDVI were recorded in 2 and 3 years alfalfa stands as compared with 1 and 4 years old alfalfa stands (Figure 9). This may be in concordance with some studies which suggests that alfalfa 2-3 years long stands have the potential of making significant benefits to soil N status.

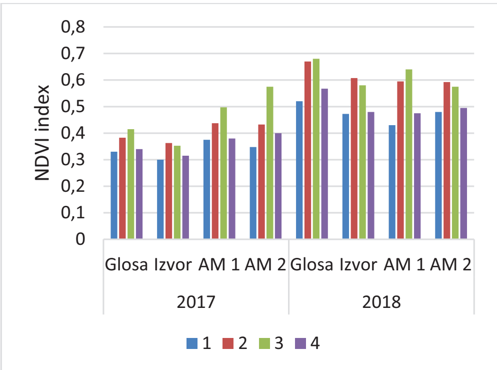

Figure 9. NDVI index of studied cultivars at stem elongation under 1-2-3-4 year old alfalfa stands. Fundulea 2017 and 2018

Also, it is obviously that under 2017 conditions mixture of winter wheat genotypes (especial mixture 2, in proportion of 50:50) have higher values of NDVI than NDVI of pure winter wheat genotypes. Matching with the studies performed by Toncea et al. (2017; 2018) shown that in ecological agriculture, the yield of the mixture of winter wheat genotypes is, often, equal or high, with about 230 kg/ha than yield of pure winter wheat genotypes.

CONCLUSIONS

The potential of NDVI to differentiate wheat cultivars for grain yield under different years and ecological agriculture was demonstrated. The NDVI was able to differentiate cultivars at different growth stages.
Under ecological agriculture system the NDVI scores at the tillering, stem elongation and anthesis stages can be used as yield predictors in wheat; the study showed a clear association between grain yield and NDVI measured at these growth stages.
The highest NDVI values have been obtained where in crop rotation alfalfa was cultivated for 2-3 years.
Under more stressful condition NDVI measurements reveals that wheat varieties mixtures are more intense green coloured.

ACKNOWLEDGEMENTS

The present work was funded through the Ministry of Agriculture and Rural Development - ROMANIA, Research Project ADER122(2015-2018).

REFERENCES

Jensen, A., Lorenzen, B., Spelling-Ostergaard, H., Kloster-Hvelplund, E. (1990). Radiometric estimation of biomass and nitrogen content of barley grown at different nitrogen levels. *International Journal of Remote Sensing, 11*(10), 1809–1820.
Filella, I., Serrano, L., Serra, J., Penuelas, J. (1995). Evaluating wheat nitrogen status with canopy reflectance indices and discriminant analysis. *Crop Science, 35*(5), 1400–1405.
Fernández, S., Vidal, D., Simón, E., Solé-Sugrañes, L. (1994). Radiometric characteristics of *Triticum aestivum* cv. Astral under water and nitrogen stress. *International Journal of Remote Sensing, 15*(9), 1867–1884.
Cárcova, L.G. Abeledo, M. López Pereira. (2003). *Análisis de la generación del rendimiento: Crecimiento, partición y components*. In: E.H. Satorre, R.L. Benech Arnold, G.A. Slafer, E.B. de la Fuente, D.J. Miralles, M.E. Otegui (Eds.), Producción de Granos: Bases funcionales para su manejo, Facultad de Agronomía, UBA, Argentina, pp. 75–98.
Le Toan, T., Ribbes, F., Wang, L., Floury, N., Ding, K., Kong, J., Fujita, M., Kurosu, T. (1997). Rice crop

mapping and monitoring using ERS-1 data based on experiment and modeling results. *IEEE Transactions on Geoscience and Remote Sensing, 1*, 41–56.
Toncea, I., Cană, L., Stan, O. (2017). Amestecul de genotipuri factor tehnologic de stabilitate cantitativă şi calitativă a producţiei de grâu cultivat în sistem ecologic. I. Amestecul de genotipuri factor de stabilitate a producţiei de grâu ecologic. I. *Analele INCDA Fundulea*, *LXXXV*. 109–123.
Martin, R.D., Jr., and Heilman, J.L. (1986). Spectral reflectance patterns of flooded rice. *Photogrammetric Engineering and Remote Sensing, 52*, 1885–1897.
Mkhabela, M.S., and Mashinini, N.N. (2005). Early maize yield forecasting in the four agro-ecological regions of Swaziland using NDVI data derived from NOAA's-AVHRR. *Agric. For. Meteor*, *129*, 1–9.
Toncea, I. (2018). Amestecul de genotipuri – factor tehnologic de stabilitate cantitativă și calitativă a producției de grâu cultivat în sistem ecologic. II. Amestecul de soiuri – factor de stabilitate a calității producției de boabe la grâul cultivat în sistem ecologic. *Analele INCDA Fundulea*, *LXXXVI*.
Syeda Refat, S., Amjed, A., Ashfaq, A., Muhammad, M., Zia-Ul-Haq, M., Shakeel, A,, Sezai, E., Hawa, Z.E. Jaafar. (2014). Normalized Difference Vegetation Index as a Tool for Wheat Yield Estimation: A Case Study from Faisalabad, Pakistan. *Scientific World Journal*. doi: 10.1155/2014/725326.
Zhao, R., Dai, J., Jiang, N., and Wang, Y. (1993). Exploration on rice yield estimation by remote sensing in Wuxi. In Experimental Research Article on W heat, Maize and Cotton Production Estimation Using Remote Sensing Technology, edited by S. Chen. Beijing: Press of Science and Technology, pp. 214–219.
Zhao, R., Wang, Y., and Dai, J. (eds) (1996). *Dynamic Monitoring and Production Estimation of Paddy Rice Agriculture in China*. Beijing: Press of Science and Technology.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESULTS REGARDING NEW SUNFLOWER GENOTYPES RESISTANT TO HERBICIDES, OBTAINED AT NARDI FUNDULEA

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Abstract

The purpose of effective weed management is the inclusion of the best measures and strategies to make sustainable sunflower production, and unfavourable for weeds. Application of pre-emergent herbicides and using herbicides tolerant sunflowers will give a good protection for this crop.
Introduction of imidazolinone and tribenuron-tolerant sunflower hybrids in practice is a revolutionary advancement in sunflower production.
Using the sources of genes for resistance to herbicides for transferring this characteristics into valuable inbred lines there have been obtained high tolerant sunflower genotypes, after many generations of selection. Resistant sunflower hybrids, with high seed yield potential, as well as with high oil content have been released. Such hybrids are very well protected of the broomrape (Orobanche cumana), due to the imidazolinone herbicides control on this parasite.
New sunflower inbred lines having the new gene, CLHA Plus have been obtained. The new hybrids which can be used in CLEARFIELD Plus system will be better protected of weeds, also of broomrape parasite.

***Key words**: sunflower, resistance, herbicides, lines, hybrids.*

INTRODUCTION

Weeds continue to pose a huge challenge for the sustainable production of sunflower despite decades of implementation of contemporary methods in order of their control.
Identification of sunflower resistance to herbicides, in the wild sunflower *H. annuus* (Al Khatib, 1998; Miller, 2000) it was like a revolution in cultivation of this crop. After launch in 2003, Clearfield production system has been well adopted in sunflower growing countries, due to a wide spectrum of weeds and broomrape (*Orobanche cumana*) control, a high level of consistency, flexibility in the timing of herbicides application, season - long weed control and a low rate of herbicide application (Malizda et al., 2000; 2003; 2012; Zollinger, 2004; Nagy et al., 2006; Phening et al., 2008; Kukorelli et al., 2011; Kaya et al., 2012; 2013). It is expected that the combination of improved imidazolinone formulation in new Clearfield Plus production system will provide a more efficient and reliable weed control in sunflower, including more freedom in crop rotation (Sala et al., 2012; Phening et al., 2012; Weston et al., 2012). Tribenuron-methyl contributes to weed control in sunflower by controlling annual broadleaf weeds and the perennial *Cirsium arvense* post-emergence, increasing the range of available herbicides in sunflower, increasing no-till/conservation

tillage process (Jocic et al., 2008; Bozic et al., 2012).
Sunflower hybrids tolerant to ALS-inhibiting herbicides are useful in controlling *Ambrosia artemisiifolia*, even some authors reported that this weed started to become resistant in some part of the world (Heap, 2016).

MATERIALS AND METHODS

For obtaining inbred lines, having good resistance to herbicides, imidazolinone or sulfonylurea types, there have been used the genes IMISUN (aditiv: *Imr1* and *Imr2*). For this type of resistance, the sources have been named, HA 442 and RHA 443. For resistance to tribenuron-methyl there have been used SURES 1 and SURES 2.
For obtaining inbred lines having resistance controlled by CLHA Plus gene, we used the sources BTI-M1 and BTI-R1.
To accelerate the process of selection for resistance it has been used the Protocol for molecular diagnosis, PCR method, for IMISUN genes and CLHA Plus gene.
We introduced in the process of genes transferring, the best inbred lines from our collection, having good agronomic traits.
It has been used a scheme for transferring the characteristics of resistance, presented in Figure 1. The number of generations of selection is higher, comparing with transferring another characteristics.

Elite inbred line (recurrent line) x Source of resistance
irir IRIR**F1 x Recurent line**
F1BC1 x Recurent line
IRir irir
F1BC2 x Recurent line
IRiririr
F1BC3 x Recurent line
IRiririr
F1BC4 x Recurent line
IRiririr
F1BC5 x Recurent line
IRiririr
F1BC6 x Recurent line
IRir irir
F1BC6
IRir
Selfpollination (F2BC6–F5BC6) – Homozygous IRIR

Figure 1. Scheme of genes transferring for resistance to herbicides

RESULTS AND DISCUSSIONS

In Table 1 there are presented the lines which have been introduced in process of transferring genes for resistance to imidazolinone herbicides (IMISUN and CLHA Plus), also genes for resistance to sulfonylurea herbicides (SURES 1 and SURES 2). There are CMS lines, also restorer lines. The same line can have three variants: resistance to IMI or to SU or CL Plus.
Having ready lines resistant to herbicides we released hybrids resistant to imidazolinone or sulfonylurea herbicides. In Figure 2 there are presented the results regarding the seed yield, obtained for some of these hybrids, in demo plots, in two locations: Fundulea and Brăila, in year 2018.

Table 1. Sunflower inbred lines introduced in the process of genes for resistance to herbicides transferring

No.	Line	Gene of resistance	Line type
1.	LC 1093	IMI; SU; CL Plus	CMS
2.	LC-1029	IMI; SU; CL Plus	CMS
3.	LC 1004	IMI; SU; CL Plus	CMS
4.	LC 1019	IMI; SU; CL Plus	CMS
5.	LC 991	IMI; SU; CL Plus	CMS
6.	LC 1050	IMI; SU	CMS
7.	AC - 1402	IMI; SU	CMS
8.	AC - 1421	IMI; SU	CMS
9.	AC - 1445	IMI; SU	CMS
10.	AC - 1532	IMI	CMS
11.	LC 1066	IMI; SU; CL Plus	Rest.
12.	LC 1085	IMI; SU; CL Plus	Rest.
13.	LC 1095	IMI; SU; CL Plus	Rest.
14.	LC 1103	IMI; SU; CL Plus	Rest.
15.	Rf - 642	IMI	Rest.
16.	Rf - 687	IMI	Rest.
17.	Rf - 693	SU	Rest.
18.	Rf - 699	SU	Rest.
19.	Rf - 714	IMI; SU; CL Plus	Rest.
20.	Rf - 734	IMI; SU; CL Plus	Rest.

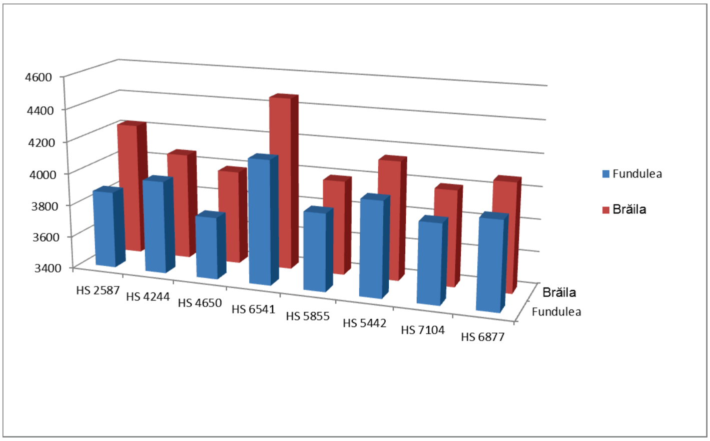

Figure 2. Seed yield (kg/ha) for sunflower hybrids

The hybrids have good seed yield in both locations, taking into consideration among the genetic potential of these, also the climatic conditions in this year. In year 2018 (Figures 3 and 4) the air temperature was good for sunflower, especially in flowering time, also the precipitation amount was favorable to this crop, in June and July.

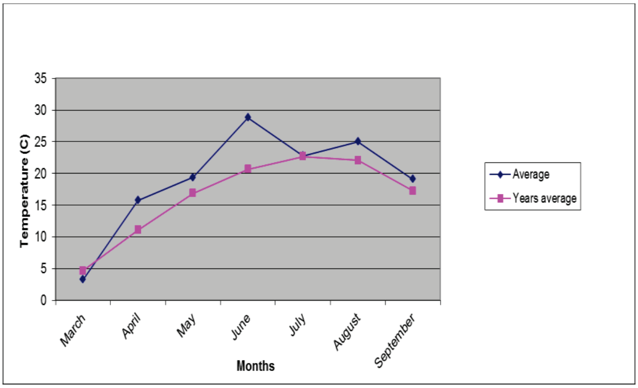

Figure 3. Temperature air in sunflower vegetation, 2018 year

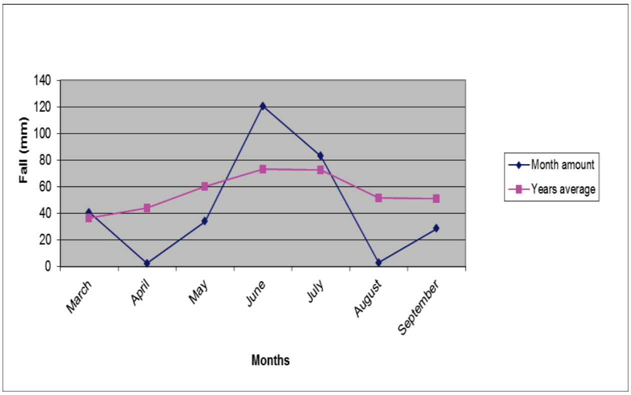

Figure 4. Fall amount in sunflower vegetation, 2018 year

In Figure 5 there are presented the results regarding the oil content of the hybrids. All of them have more than 50% oil content, in both locations.

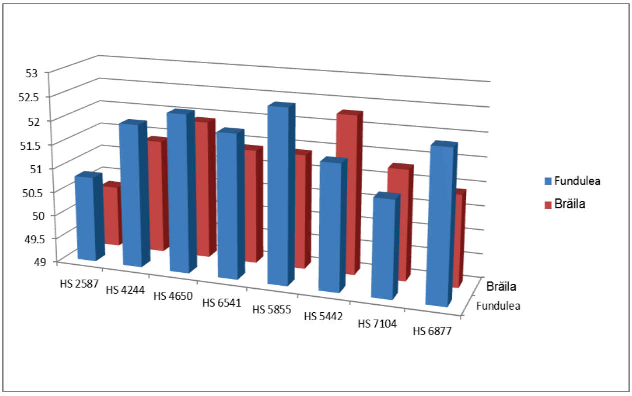

Figure 5. Oil content for sunflower hybrids

In Table 2 are presented the results regarding some important characteristics of the new obtained hybrids. All of them have a quite short vegetation period, they are a medium plant height, good head diameter, good hectolitric weight, high thousand kernels weight, and also a good tolerance to drought.

Table 2. Important characteristics of the new hybrids

Hybrid	Vegetation period (days)	Plant height (cm)	Head diameter (cm)	Hectolitric weight (kg/hl)	TKW (g)	Resistance to drought (notes)
HS 2587	118	164	25	41.4	74	3
HS 4244	125	167	23	38.2	65	2
HS 4650	124	169	24	38.3	70	2
HS 6541	119	165	24	39.1	73	3
HS 5855	125	168	25	41.2	73	2

Resistance to drought: 1 - resistant; 9 - sensitive.

CONCLUSIONS

In the breeding program for sunflower, at NARDI Fundulea, there have been obtained valuable inbred lines, having good resistance to herbicides. There have been used sources for resistance received on the base of contracts with companies which produced them.

By combination of these lines, CMS with Restorer we received good hybrids, which have been tested in demonstration plots.

The hybrids released very good seed yield, also they have very good oil content. These hybrids have good value of important characteristics.

All new hybrids will be given for testing in the network trials of State Institute for Variety testing and Registration, in order to be registered in the Official List.

REFERENCES

Al-Khatib, K., Peterson, D.E., Currie, R.S. (1998). Imazethapyr resistance in common sunflower (*Helianthus annuus* L). *Weed Science*, *46*, 403–407.

Bozic, D., Saric, M., Malidza, G., Ritz, C., Vrbnicanin, S. (2012). Resistance of sunflower hybrids to imazamox and tribenuron-methyl. *Crop Protection*, *39*, 1–10.

Heap, I. (2016). The International Survey of Herbicide Resistant Weeds. Online. Internet. www.weedscience.org.

Jocic, S., Miklic, V., Malidza, G., Hladni, N., Gvozdenovic, S. (2008). New sunflower hybrids tolerant of tribenuron-methyl. In: Proc. *17th Int. Sunfl. Conf.*, Cordoba, Spain. 505–508.

Kaya, Y., Evci, G., Pekcan, V., Yilmaz, I. M. (2013). Clearfield Technology in Sunflower and Developing Herbicide Resistance Sunflower Hybrids. *Soil-Water Journal*. *2*(2), 1713–1720.

Kaya, Y., Jocic, S., Miladinovic, D. (2012). Sunflower In S.K. *Gupta (Ed) Technological Innovations in Major World Oil Crops*, *1*, 85–130.

Kukorelli, G., Reisinger, P., Torma, M., Ádámszki, T. (2011). Experiments with the control of common ragweed in imidazolinone-resistant and tribenuron-methyl-resistant sunflower. *Herbologia*, *12*, 15–22.

Malidza, G., Jocic, S., Skoric, D. (2000). Imidazolinone-resistant sunflower (*Helianthus annuus* L.): Inheritance of resistance and response towards selected sulfonylurea herbicides. In Proc. of 15th Int. Sunflower Conf. Toulouse, France, June 12-15, 42–47.

Malidza, G., Jocic, S., Skoric, D. (2003). Weed and broomrape *(Orobanche cernua)* control in Clearfield sunflower. *European Weed Research Society (EWRS) 7th Mediterranean Symposium*, Cukurova Univ. Adana-Turkey, 51–52.

Malidza, G., Jocic, S., Rajkovic, M., Dusanic, N. (2012). Broomrape *(Orobanche cumana)* control in tribenuron-tolerant sunflower. Proceeding of the *6th International Weed Science Congress* 17-22 June, 2012 Hangzhou, China, 103.

Miller, J.F. and Al-Khatib, K. (2000). Development of herbicide resistant germplasm in sunflower. Proc. of *15th Int. Sunflower Conf.*, Toulouse, France. June 12-15. *2*, 419–423.

Nagy, S., Reisinger, P., Pomsár, P. (2006). Experiences of introduction of imidazolinone-resistant sunflower in Hungary from herbological point of view. *J Plant Dis Prot*, Special Issue, *20*, 31–37.

Pfenning, M., Palfay, G., Guillet, T. (2008). The CLEARFIELD® technology - A new broad-spectrum post-emergence weed control system for European sunflower growers. *J. Plant Dis. Protect*. Special Issue, 649–654.

Pfenning, M., Tan, S., Perez-Brea, J. (2012). Improved Weed Control in Clearfield-Plus Sunflowers with superior herbicide solutions. Proc of *18th International Sunflower Conference*, Mar del Plata, Argentina, 535–538.

Sala, C.A., Bulos, M., Altieri, E., Ramos, M.L. (2012). Genetics and breeding of herbicide tolerance in sunflower. *Helia*, *35*, 57–70.

Weston, B., McNevin, G., Carlson, D. (2012). Clearfield® Plus Technology in Sunflowers. Proc of *18th International Sunflower Conference*, Mar del Plata, Argentina, 149–154.

Zollinger, R.K. (2004). Advances in sunflower weed control in the USA. Proc. *16th International Sunflower Conference*, Fargo, ND USA, 435–439.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RHIZOBACTERIA EFFECTS ON PHOTOSYNTHETIC PIGMENTS AND NITROGEN CONTENTS OF SOYBEAN PLANTS CULTIVATED UNDER LOW PHOSPHORUS AND WATER LIMITED CONDITIONS

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Abstract

The application of plant growth promoting rhizobacteria (PGPR) is a sustainable approach to improve crops physiological processes and to overcome abiotic stresses. The effects of rhizobacteria Bradyrhizobium japonicum applied alone or in combination with Pseudomonas putida were examined on photosynthetic pigments contents and leaf nitrogen status in soybean plants subjected concomitantly to low phosphorus (P) and moderate drought conditions. The lowest content of pigments was revealed in plants subjected to both abiotic factors. The results indicated that the application of B. japonicum and P. putida significantly improved nitrogen and photosynthetic pigments status in leaves, particularly chlorophyll a and b under temporary drought compared to uninoculated ones. The application of rhizobacteria B. japonicum alone or together with P. putida significantly increased carotenoids concentrations in plants regardless of soil moisture levels. In conclusion, the detrimental effects generated by low P and drought on photosynthetic pigments status and nitrogen nutrition of soybean were partially alleviated by the use of rhizobacteria.

***Key words**: chlorophyll, drought, Glycine max L., nitrogen, rhizobacteria.*

INTRODUCTION

The various environmental factors like low soil fertility and drought stress have detrimental impact on plant growth and productivity (Gouda et al., 2018; Cura et al., 2017). Abiotic stresses, provoked by these factors, can hamper a range of physiological processes, particularly photosynthesis and cause damage to plants. The chlorophyll status in plants is an important factor in crop production because it affects the amount of radiation intercepted and, therefore, plant growth (Ashraf & Harris, 2013). Chlorophyll loss is associated with environmental stress, and the variation in total pigments contents may be an estimative indicator of stress in plants (Ashraf & Harris, 2013; Netto et al., 2005). Lisar et al. (2012) found that water stress reduces chlorophyll synthesis, while carotenoids are less affected.

Plant growth-promoting bacteria (PGPB) are reported to influence the growth, yield, photosynthetic activity and nutrient uptake by several mechanisms (Delshadi et al., 2017; Egamberdieva et al., 2017a; 2017b). Some bacterial strains directly regulate plant physiology by mimicking synthesis of plant hormones, whereas others increase mineral and nitrogen assimilation and in turn the photosynthetic activity is enhanced. One of the possible mechanisms to increase chlorophylls contents in plants could be associated with improvement of nitrogen status in crops. However, little is known about PGPR impact on nitrogen (N) nutrition of crops when the low P availability is coupled with water deficit in the soil. Reduced N supply is known to reduce the translocation of nitrates, phosphates, calcium, magnesium and amino acids in plants. These physiological disturbances significantly affect the photosynthetic pigment contents in plants. Soybean (*Glycine max*. L.) is a major legume crop and an important source of protein, vegetable oil in many countries; however, its production is restricted by low P availability and drought (Graham & Vance, 2003). To date, little is known regarding the effect of PGPR on the changes in foliar pigments of soybean in relation to P supply and soil moisture levels. Even though *Bradyrhizobium japonicum* and *Pseudomonas putida* have been studied as single isolate affecting leaf chlorophyll status in legumes (Israr et al., 2016; Kang et al., 2014;

Egamberdieva et al., 2017c), little has been done on their possible interactions under water and P limited environments. The aim of this investigation was to evaluate the effect of rhizobacteria *Bradyrhizobium japonicum* applied alone or in combination with *Pseudomonas putida* on the chlorophyll (*a* and *b*), carotenoids and nitrogen content in leaves of *Glycine max* L. under low P fertility and moderate drought conditions.

MATERIALS AND METHODS

The soil used for the experiment was chernoziom carbonated with low P availability (18 mg P/kg soil). In a controlled pot culture experiment, a set of soybean (cv. Horboveanca) plants were inoculated with rhizobacteria *B. japonicum* (Rh*)* alone and another set of plants were inoculated with *B. japonicum* in combination with soil applied *P. putida* (PP). In addition, it was a variant with P fertilization at the rate 100 mg P/kg soil. Uninoculated plants served as control treatment. Pots were arranged in a randomized complete block design with four replications. All plants were well-watered till flowering stage. After that, a set of plants were watered at 70% WHC (water holding capacity) as normal soil moisture and the other set of plants was subjected to low water conditions (35% WHC) for 12 days. After that period leaves samples were collected to determine photosynthetic pigments contents. To determine the chlorophyll content, leaf samples were extracted with acetone (80%). The absorbance of the supernatant was recorded at 645 and 663 nm against the solvent (acetone) (Arnon, 1949). The carotenoids concentration was measured at 452 nm. The total nitrogen content was determined by Kjeldahl method. The obtained data were given as the mean with standard error values by using Statistic 7 program, and the differences in the means were determined by the least significant difference (LSD) (P=0.05) test.

RESULTS AND DISCUSSIONS

Morphological, nutritional and physiological changes induced in plants colonized with PGPR contribute to their enhanced growth and resistance to abiotic stresses**.** Variations in chlorophyll patterns in leaves are indicators of senescence, stress or damage to the photosynthetic apparatus and affect the normal growth of plants. In this study, we investigated whether the application of beneficial bacteria *B. japonicum* alone or in combination with *P. putida* have potential to improve chlorophylls status in soybean cultivated under limited P supply and temporary drought. The effects of these bacterial strains on the chlorophylls contents in plants are shown in Figure 1.

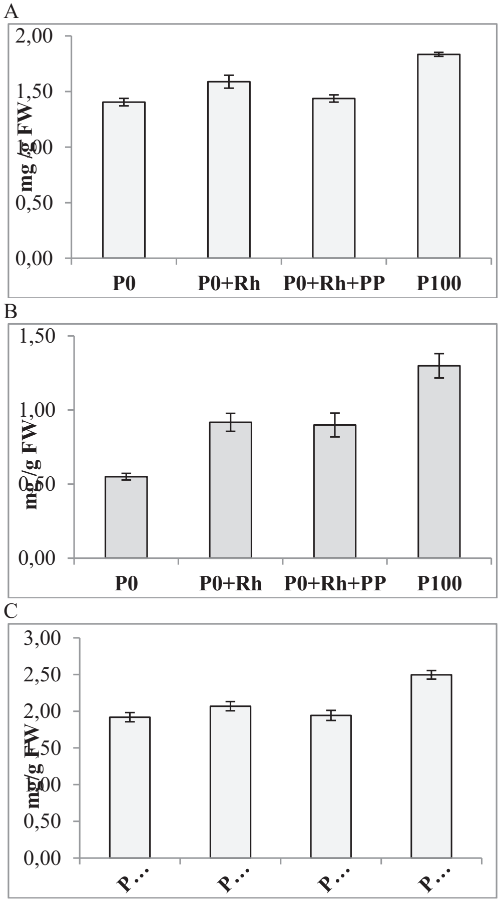

Figure 1. The effect of *Bradyrhizobium japonicum* (Rh) applied alone or in combination with *Pseudomonas putida* (PP) on (A) chlorophyll *a*, (B) chlorophyll *b* and (C) total chlorophylls content in leaves under low P and normal soil moisture. Values exhibit means ± SE

Experimental results revealed the lowest content of pigments in plants cultivated under low P availability and subjected to moderate drought. The decrease in chlorophyll (*a* and *b*) contents in drought-affected soybean plants might be attributed to the possible oxidation of chlorophyll and other chloroplast pigments coupled with instability of the pigment protein complex under abiotic stress. Similar trends have been reported by Curá et al. (2017) who revealed that drought stress significantly decreased the total chlorophyll content in leaves of maize plants.

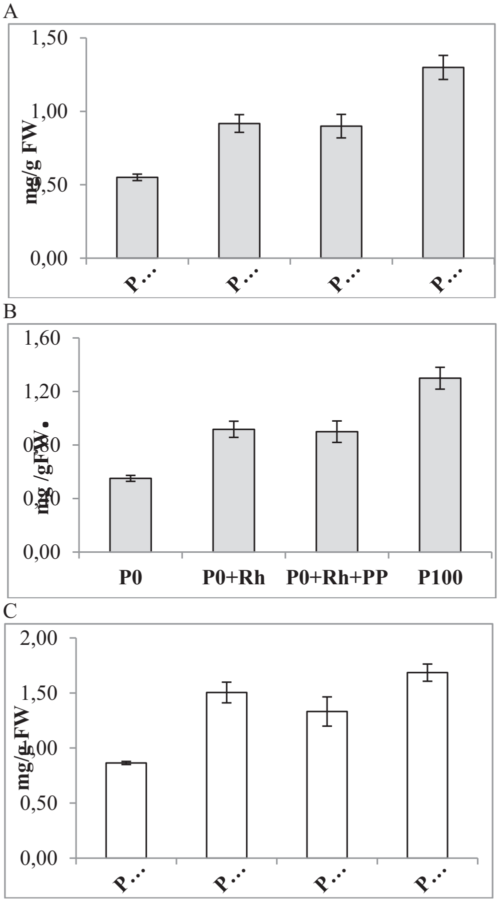

Figure 2. The effect of *Bradyrhizobium japonicum* (Rh) applied alone or in combination with *Pseudomonas putida* (PP) on (A) chlorophyll *a*, (B) chlorophyll *b* and (C) total chlorophylls content in leaves under low P and drought. Columns are means ± SE

The experimental data of this study have shown that the application of rhizobacteria *B. japonicum* improved the photosynthetic pigments contents in soybean plants under normal soil moisture as well as under water stress (Figures 1 and 2).

The content of chlorophyll *a* and chlorophyll *b* was increased by 13.6% and 76.9% due to *B. japonicum* inoculation in plants not subjected to water deficit. Plants inoculated with the combination of *B. japonicum* and *P. putida* did not increase photosynthetic pigments further compared to the inoculated plants with *B. japonicum* alone. The highest concentration of pigments was registered in treatment with P fertilization irrespective of soil moisture levels. Sufficient supply of P contributes to uptake and assimilation of nutrients especially of Mg as was reported by Vafadar et al. (2014). In addition, higher chlorophyll content in response to P supplementation might account for synthesis of more carbohydrates in fertilized plants. Investigation data revealed that the application of rhizobacteria also improved the chlorophyll *a* in soybean plants under water stress. Likewise, the chlorophyll *b* content was increased significantly in all the PGPR strain treatments. Indeed, the application of *B. japonicum* alone increased chlorophyll *a* and *b* by 67.2% and 90.3% under temporary drought compared to uninoculated plants. An increase of chlorophyll content in plants contributes to higher photosynthetic activity and assimilates production, which could be supported the plant growth improvement under unfavorable P supply and drought environment. Similar results have been reported by Mia et al. (2010), who treated banana plants with chemical fertilizers and PGPR and observed positive effects on chlorophyll contents and Ca, Mg, K uptake by plants. Kang et al. (2014) also demonstrated a stimulatory effect of *P. putida* strain on chlorophylls accumulation in soybean under salinity stress. In this study, the stimulatory effect of rhizobacteria on total pigments contents was registered in plants grown under water deficit environment. However, under insufficient supply of water the major increases were observed in inoculated plants compared to uninoculated ones. In general, plants subjected to drought irrespective of treatments had low concentration of chlorophylls compared to well-watered ones. Manivannan et al. (2008) showed that under drought stress the amount of chlorophyll and carotenoids was reduced in *Helianthus annuus*. It seems that the reason for this decrease is due to the increased destruction and/or production of pigments as consequences of oxidative stress generated by stress of abiotic factors. Likewise, the reduced chlorophyll contents of the plant under unfavorable environments might be somewhat due to limited N supply in plant tissues and changes in enzymes activity such as nitrate reductase (Delshadi et al., 2017). However, the application of tested PGPR strains attenuates adverse effects of drought on chlorophyll. Esitken et al. (2010) reported that the use of growth promoting bacteria of *Pseudomonas* increased the amount of Fe and Mg in strawberry, hence, created favorable physiological conditions for pigment synthesis. We also can suppose that tested bacterial strains alleviated iron and magnesium nutrition of soybean plants. Our results of enhanced chlorophyll contents due to PGPR application and subsequent allaying of drought induced negative effects support the results for fenugreek (Sharghi et al., 2018). Thus, the enhanced chlorophylls status in plants was because rhizobacteria treated plants maintain higher contents of magnesium and N resulting in greater physiological activity of chloroplasts (Egamberdieva et al., 2017a). Carotenoids play an important role in the process of reactive oxygen species scavenging, in the stability of photosynthetic machinery activity; participate in energy dissipation hence contributing to the reduction of adverse effects generated by unfavorable environmental conditions (Ashraf & Harris, 2013). In our experiment, the carotenoids contents in plants were detrimentally affected by drought and P deficiency. The decrease in carotenoids status due to drought and low soil fertility of P could be related to limited nutrients availability in leaves tissues. In addition, it is known that under scarce conditions of P and water deficit the uptake of Mg is restricted. The application of *B. japonicum* alone increased this physiological parameter in plants (Figure 3). This elicitor influence of rhizobacteria was observed irrespective of soil moisture levels. It is necessary to note that the leaves biomass in treatment with both strains was significantly higher than in treatment with *B. japonicum* alone (data not shown). The total carotinoids content increased by 63.2% in plants not subjected to water limited conditions and by 54.4% in soybean plants under drought due to inoculation only with N-fixer rhizobacteria (Figure 3A). However, the combined application of two bacteria strains did not increase this parameter under drought conditions compared to treatment of *B. japonicum* alone (Figure 3B). Generally, the carotenoids accumulation decreased in plants without rhizobacteria treatment regardless of soil moisture level. One of the mechanisms induced by plant growth promoting bacteria is improving plant nutrition, especially with nitrogen.

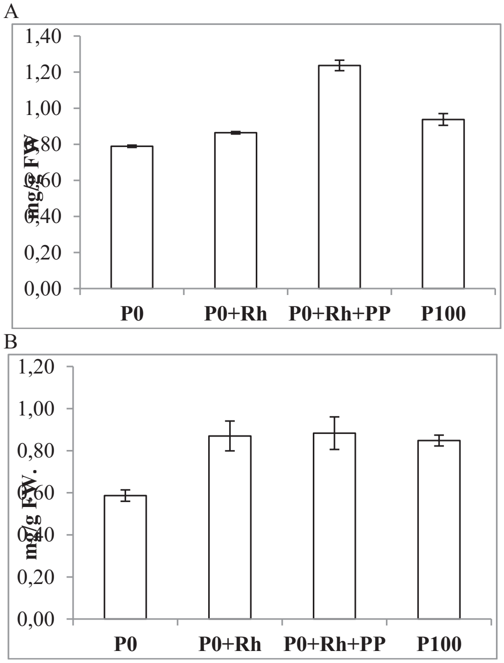

Figure 3. The effect of *Bradyrhizobium japonicum* (Rh) applied alone or in combination with *Pseudomonas putida* (PP) on carotinoids content in leaves under (A) normal soil moisture and (B) drought. Columns are means ± SE

According to literature data there is a strong correlation between photosynthetic pigments synthesis and nitrogen status of plants. For example, Dawwam et al. (2013) reported that the stimulation of pigment synthesis in potato plants by PGPR might be due to the changes in N, P and K uptake. The study's results found out that the *B. japonicum* inoculation treatment in combination with *P. putida* was the most effective treatment for increasing the foliar N concentration in plants under temporary drought. We'd like to mention that the co-inoculation effect of *Bradyrhizobium* and *P. putida* enhanced nodulation and root growth compared to the control plants (data not shown), which in turn have beneficial impact on nitrogen assimilation by soybean. Exposure of soybean to water stress reduced accumulation of essential mineral elements like nitrogen and phosphorous resulting in decrease of growth. Altogether, increased N content in inoculated plants has positive influence on the synthesis of chlorophyll pigments resulting in enhanced synthesis of photoassimilates and hence leads to improvement of resistance to abiotic factors.

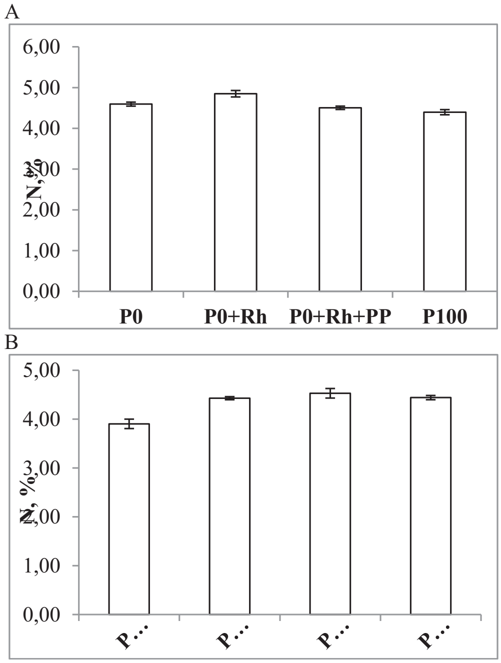

Figure 4. The effect of *Bradyrhizobium japonicum* (Rh) and *Pseudomonas putida* (PP) isolates on N content in leaves under normal soil moisture (A) and moderate drought (B). Columns are means ± SE.

Under normal water soil conditions rhizobacteria did not significantly change the nitrogen concentration in leaves tissues of soybean plants (Figure 4A). The administration of bacterial strains increased the nitrogen status in leaves of plants grown under unfavorable soil moisture conditions. The differences in nutrient contents between uninoculated soybeans and those inoculated together with *B. japonicum* and *P. putida* were significant, the nitrogen content being improved by 25% (Figure 4B). Perhaps, these rhizobacteria stimulated root growth and its physiological activity. Experimental results also suggested an increase in nodule number in soybean upon bacterial inoculation under drought (data not shown). Our experimental results agrees with the experiment by Egamberdieva et al. (2017c), where lupine inoculated with the *Bradyrhizobium* strain and with the HTC-BR inoculants contained 11% and 33% higher nitrogen contents under irrigation, and even 21% and 39% under drought relative to uninoculated plants, respectively. In uninoculated plants, the contents of total nitrogen were similar for plant tissues under drought as well as for those of non-stressed ones. According to experimental results regarding N status we can make a conclusion that combined applications of both rhizobacteria species had better impact on nitrogen status in plants compared with the inoculation with *B. japonicum* alone under temporary drought. Egamberdieva et al. (2017b) demonstrated that both inoculation treatments, either *B. japonicum* USDA 110 alone or combined with *P. putida* TSAU1, enhanced nitrogen contents in salt-stressed soybean tissues. Likewise, Israr et al. (2016) suggested that the application of *P. putida* strain not only enhances P uptake, but also improves the N and K uptake by chickpea plants grown on high pH soils, which can thus reduce the application of chemical fertilizers. In this study, the phosphorus supplementation provides the same status of nitrogen as rhizobacteria application.

CONCLUSIONS

Inoculation of soybean with *Bradyrhizobium japonicum* improved photosynthetic pigments status in plants under normal soil moisture as well as under water deficit and low P fertility of soil. Combined application of *B. japonicum* and *P. putida* had no synergic effect on chlorophylls status of plants regardless of soil moisture level. The combined application increased significantly the nitrogen contents under scarce moisture conditions. Thus, experimental results suggested that PGPR enabled the leaf to maintain high levels of photosynthetic pigments and nitrogen and reduced the adverse effects of water deficit and low P on soybean plants. These findings were obtained in greenhouse conditions; therefore, it would be necessary to carry out further experiments under field conditions which would be quite important research for sustainable crop production under unfavorable environments.

ACKNOWLEDGEMENTS

This research work was carried out with the support of Council Supreme of Sciences and Technology Development of Moldavian Academy of Sciences.

REFERENCES

Arnon, D.I. (1949). Copper enzyme is isolated chloroplast polyphenol oxidase in *Beta vulgaris*. *Plant Physioligy*, *24*, 1–15.

Asheraf, M., & Harris P.J.C. (2013). Photosynthesis under stressful environments: an overview. *Photosynthetica*, *51*, 163–190.

Curá, J.A., Franz, D.R., Filosofía, J.E., Balestrasse, K.B., Burgueño, L.E. (2017). Inoculation with *Azospirillum* sp. and *Herbaspirillum* sp. bacteria increases the tolerance of maize to drought stress. *Microorganisms*, *5*, 41–48.

Egamberdieva, D., Wirth, S., Jabborova, D., Räsänend, L., Lliao, H. (2017a). Coordination between *Bradyrhizobium* and *Pseudomonas* alleviates salt stress in soybean through altering root system architecture. *Journal of Plant Interactions*, *12*, 100–107.

Egamberdieva, D., Wirth, S., Shurigin, V., Hashem, A., Abd_Allah, E.F. (2017b). Entophytic bacteria improve plant growth, symbiotic performance of chickpea (*Cicer arietinum* L.) and induce suppression of root rot caused by *Fusarium solani* under salt stress. *Frontiers in Microbiology*, *8*, 75–82.

Egamberdieva, D., Reckling, M., Wirth, S. (2017c). Biochar-based *Bradyrhizobium* inoculum improves growth of lupin (*Lupinus angustifolius* L.) under drought stress. *European Journal of Soil Biology*, *78*, 38–42.

Esitken, A., Yildiz, E.H., Ercisli, S., Donmez, M.F., Turan, M., Gunes, A. (2010). Effects of Plant Growth Promoting Bacteria (PGPB) on yield, growth and nutrient contents of organically grown strawberry. *Scientia Horticulturae*, *124*, 62–66.

Dawwam, G.E., Elbeltagy, A., Emara, H.M., Abbas, I.H., Hassan, M.M. (2013). Beneficial effect of plant growth promoting bacteria isolated from the roots of potato plant. *Annals of Agricultural Science*, *58*, 195–201.

Delshadi, S., Ebrahimi, M., Shirmohammadi, E. (2017). Influence of plant-growth-promoting bacteria on germination, growth and nutrients' uptake of *Onobrychis sativ*a L. under drought stress. *Journal of Plant Interactions*, *12*, 200–208.

Gouda, S., Kerry, R.G., Das, G., Paramithiotis, S., Shin, H.S., Kumar, J.P. (2018). Revitalization of plant growth promoting rhizobacteria for sustainable development in agriculture. *Microbiological Research*, *206*, 131–140.

Graham, P.H., & Vance, C.P. (2003). Legume Importance and Constraints to Greater Use. *Plant Physiology, 131*, 872–877.

Israr, D., Ghulam, M., Khan, K.S., Shahzad M., Ahmad, N., Masood, S. (2016). Interactive effects of phosphorus and *Pseudomonas putida* on chickpea (*Cicer arietinum* L.) growth, nutrient uptake, antioxidant enzymes and organic acids exudation. *Plant Physiology and Biochemistry*, *108*, 304–312.

Kang, S.M. et al., 2014. Plant growth promoting rhizobacteria reduce adverse effects of salinity and osmotic stress by regulating phytohormones and antioxidants in *Cucumis sativus*. *Journal of Plant Interactions*, *9*, 673–682.

Lisar, S.Y.S., Motafakkerazad, R., Hossain, M.M., Rahman, I.M.M. (2012). Water Stress in Plants: Causes, effects and responses. *InTech*., 1–14.

Manivannan, P., Jaleel, C.A., Somasundaram, R. Panneerselvam, R. (2008). Osmoregulation and antioxidant metabolism in drought stressed *Helianthus annus* under triadimefon drenching. *Comptes Rendus Biologies*, *331*, 418–425.

Mia, M.A.B., Shamsuddin, Z.H., Wahab, Z., Marziah, M. (2010). Effect of plant growth promoting rhizobacterial (PGPR) inoculation on growth and nitrogen incorporation of tissue-cultured Musa plantlets under nitrogen-free hydroponics condition. *Australian Journal of Crop Sciences*, *4*, 85–90.

Netto, A.T., Campostrini, E., De Oliveira, J.G., Bressan-Smith, R.E. (2005). Photosynthetic pigments, nitrogen, chlorophyll a fluorescence and SPAD-502 readings in coffee leaves. *Scientia Horticulturae*, *104*, 199–209.

Sharghi, A., Badi, N.H., Bolandnazar, S., Mehrafarin, A., Sarikhani, M.R. (2018). Morphophysiological and phytochemical responses of fenugreek to plant growth promoting rhizobacteria under different soil water levels. *Folia Horticulture*, *30*, 215–228.

Vafadar, F., Amooaghaie, R., Otroshy, M. (2014). Effects of plant-growth-promoting rhizobacteria and arbuscular mycorrhizal fungus on plant growth, stevioside, NPK, and chlorophyll content of *Stevia rebaudiana. Journal of Plant Interactions*, *9*, 128–136.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

NEW SOURCES FOR GENETIC VARIABILITY WITH RESISTANCE AT DROUGHT OBTAINED BY INTERSPECIFIC HIBRIDIZATION BETWEEN CULTIVATED SUNFLOWER AND THE ANNUAL WILD SPECIES *Helianthus argophyllus*

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Abstract

Taking into account the major climatic changes recorded over the last 10 years, one of the main objectives of sunflower improvement is adaptability to environmental conditions, high productivity in extreme drought and burning conditions. Considering the global warming predicted by experts, the first step was to identify genotypes created at NARDI Fundulea for drought resistance. The new genotypes were obtained by interspecific hybridization of the Helianthus argophyllus crop rows. The wild species is recognized as having high drought resistance due to the highly developed root system and foliar pubescence that reduces evapotranspiration. Genotypes identified with increased drought resistance gave a high yield of seeds, a mass of 1000 seeds and high oil content (> 45%) were introduced as new genetic resources interspecific for the improvement of sunflower. The oil content was read using a nuclear resonator. Embryo rescue technique was used to speed up the breeding process. Thus, immature hybrid embryos were inoculated on germination media and the plants were transferred into pots with earth/sand (50/50) then in 10 kg buckets and were grown in the greenhouse, 2 generations/year were obtained.

***Key words**: Helianthus argophyllus, embryo rescue, interspecific hybridization, sunflower genotypes, gene introgression of drought resistance.*

INTRODUCTION

In Romania, Vrânceanu (2000) was able to obtain interspecific progenies between *Helianthus annuus x Helianthus argophyllus*. Interspecific hybridization is an additional technique to create new sources of genetic variability for the improvement of sunflower Christov, 2013; Prohens et al., 2018).

According to the information provided by Hussain et al (2017), the evaluation of the descendants obtained by hybridizing the sunflower cultivated *Helianthus annuus* with annual wild species *Helianthus argophyllus* showed improvements in water behavior (water consumption, stomatic behavior), but the characters of productivity, precocity have been diminished.

At the same time, the offspring of the first generation of hybrids were strongly branched, small caps and a long vegetation period (Merrien et al., 1996).

Improving drought resistance is quite complex due to the polygenic control of this character. Many research has been carried out to identify a single character that would serve as a basis for drought resistance selection, but this approach has proved to be unrealistic (Vrânceanu, 2000).

Characters identified as correlated with resistance to water stress are: the average weight of achenes (Andrich et al., 1996), high productivity achieved through a large amount of biomass at maturity, rapid phenological development combined with a long reproduction period and thick stem strains to provide sufficient amounts of carbonated hydrates in the critical seed filling period (Barron, 1991).

Baldini et al. (1993) reported that the annual wild species *Helianthuus argophyllus* can be used in drought resistance programs because it absorbs water better during long periods of

drought due to the highly developed root system.
Following the testing and selection under natural climatic and soil conditions on agricultural areas, Saucă (2010a; 2010b), Saucă and Lazăr (2016) reported the existence of a large genetic variability within the newly created interspecific germplasm both for drought resistance and for resistance to various pathogens.

MATERIALS AND METHODS

In this paper will be presented the results obtained in experimental years 2016-2017 at Fundulea (Calarasi County) and Stupina (Constanta County) where observations were made for: drought resistance, seed production and oil content in new genotypes created.
In the collection from NARDI Fundulea, there are many sunflower genotypes (inbred lines based on cytoplasmic androsterility - A lines, inbred lines maintaining fertility - B lines and fertility restoration lines - C lines, synthetic populations, hybrids) with different degrees of drought resistance.
In order to improve this character without diminishing the seed production and the percentage of seed oil, and in order to support the farmers practicing the classic system, we make interspecific hybridization between 6 fertility maintaining lines (1B-M2, 2B-M4, 3B-M6, 4B-M7, 6B-M11 and M12), and one synthetic population (5B-M10) with the annual wild species *Helianthuus argophyllus* in year 2014 followed by generations of backcross and selection in the years 2016 and 2017, obtaining new genetic material (widening the genetic basis of sunflower genotypes) to be used in the process of breeding for various stressors, including drought and heat (Table 1).

Table 1. Name/code of sunflower genotypes

Code of sunflower genotype	Name of sunflower genotype
1B-M2	Polet11B
Bio1	Polet11B x *H. argophyllus*
2B-M4	O-7493B
Bio3	O-7493B x *H. argophyllus*
3B-M6	RPC-46B
Bio5	RPC-46B x *H. argophyllus*
4B-M7	Tard85-2B
Bio7	Tard 85-2B x *H. argophyllus*
5B-M10	Pop. 79-16B
Bio9	Pop79-16B x *H. argophyllus*
6B-M11	Tard85-1B
Bio11	Tard85-1B x *H. argophyllus*
M12	LC-1093B
Bio13	LC-1093B x *H. argophyllus*

We sowing micro plots with four rows per sunflower genotype (7 m/row), in 3 replications (Figure 1).

Figure 1. Aspects from field with sunflower genotypes tested for resistance to drought, Fundulea 2016

We harvest total number of plants/plot and obtain seed yield per plot and after that we determinated seed yield per plant.

$$\text{Seed yield per plant} = \frac{\text{Seed yield per plot}}{\text{Total number of plants per plot}}$$

We determining the oil content with a **Analyzer Spin Track with** Nuclear Magnetic Resonance (Figure 2).

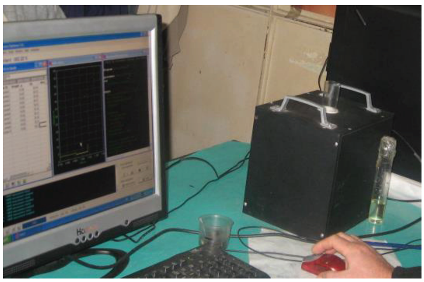
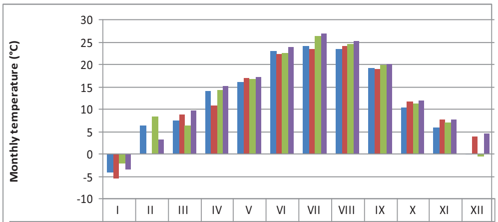


Figure 2. Analyzer Spin Track with Nuclear Magnetic Resonance

RESULTS AND DISCUSSIONS

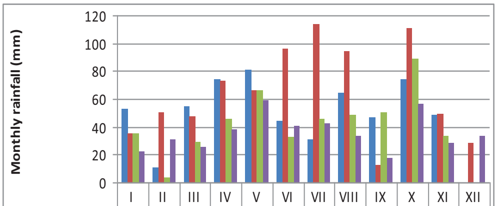
During the sunflower growing season (20-25 April - sowing, 20-25 September - harvesting) average temperatures (Figure 3) are in April, at sunflower sowing, the temperatures recorded at both weather stations from Stupina and Fundulea, were higher in 2016 at about 3ºC compared to 2017; in July and August, in the phenophase of filling and maturity of the achenes, at Stupina were recorded higher temperatures with 2ºC in both experimental years, compared to the station in Fundulea. From Figure 3 it can be noticed that during the experimental period the air temperature at both Fundulea and Stupina in January is below 0°C.



	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
Fundulea 2016	-4.3	6.2	7.3	13.9	15.9	22.9	24.1	23.4	19.1	10.3	5.7	-0.3
Fundulea 2017	-5.5	0	8.6	10.6	16.8	22.2	23.3	23.9	19	11.6	7.5	3.7
Stupina 2016	-2.3	8.2	6.2	14.3	16.6	22.5	26.2	24.5	19.8	11.1	6.9	-0.7
Stupina 2017	-3.5	3.2	9.5	15.1	17.1	23.7	26.8	25.1	20	11.8	7.5	4.5

Figure 3. Monthly average temperatures recorded in Fundulea and Stupina in the years 2016-2017

Regarding the amount of rainfall during the calendar years under study, we can conclude that, without exception, the rainfall was significantly higher at Fundulea compared to those recorded at Stupina (Figure 4).



	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
Fundulea 2016	53.3	10.3	54.9	73.7	81.2	43.7	31.3	64.6	46.6	74.4	48.8	0
Fundulea 2017	35.4	50.5	47.6	73.6	65.8	96.4	114	94.4	12.2	112	49.2	27.8
Stupina 2016	35.4	3.6	28.9	45.9	66.6	32.8	45.7	48.3	50.2	89.3	33.2	0.35
Stupina 2017	22.5	30.9	25.6	38.2	58.9	40.3	42.5	33.9	17.8	56.8	28.3	33.6

Figure 4. Monthly average rainfall recorded in Fundulea and Stupina in the years 2016-2017

Noteworthy is that under high temperature conditions in April-September, rainfall was significantly low at Stupina in 2017 in the same months. Newly created sunflower genotypes have undergone simultaneous thermal and hydric stress.

From Figure 5 it can be seen that both the fertility maintaining lines and the interspecific hybrids derived from the hybrids produced a higher amount of seeds in the weather conditions at Fundulea. It is noted the Bio 7 genotype that was obtained from interspecific hybridization with the 4B-M7 fertility maintaining line that consistently behaved in both experimental years achieving a higher seed yield/sunflower plant than the 4B-M7 fertility maintaining line both in weather conditions from Fundulea and from Stupina. Also noteworthy are the Bio5 and Bio 11 genotypes that have exceeded seed production over the 3B-M6 and 6B-M11 fertility maintaining lines in both locations in 2016 and 2017.

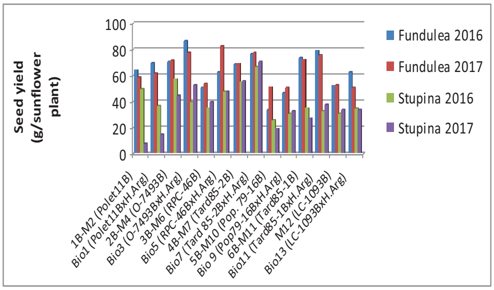


Figure 5. Seed yield (g/sunflower plant), fertility maintaining lines (lines B), synthetic populations and descendants derived from interspecific hybridization between B lines, synthetic populations and the annual wild specie *Helianthus argophyllus* in the years 2016 and 2017 in Fundulea and Stupina

Regarding the oil content, it can be seen from Figure 6 that the Bio 7 and Bio 9 genotypes have an oil content of nearly 50% under the conditions of Fundulea, exceeding the 4B-M7 maintaining fertility line and the synthetic population 5B-M10. And for this character, the Bio 7 genotype exhibits stability and superiority to the 4B-M7 maintaining fertility line over the two years, and the two locations (50%). It is followed by Bio 9 genotypes with an oil content of over 45% and Bio 11 with 44% obtained in the Stupina in 2017.

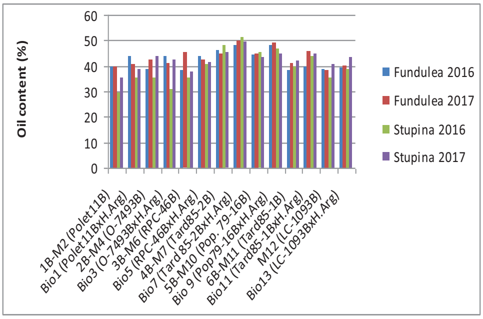


Figure 6. Oil content (%) of newly created genotypes compared to fertile maintenance and synthetic population in the years 2016 and 2017 in Fundulea and Stupina

1000 weight seed (TWS) is a parameter strongly influenced by the amount of precipitation that fell during from the sowing period (favoring the development of the root system) until at the filling of the seed.

As can be seen in Figure 4, in 2017 in Fundulea, it was a year with many rainfall. This explains the fact that MMB was higher in all genotypes in Fundulea in 2017 and in all cases higher in interspecific descendants compared to the fertile maintenance line and the synthetic population in both locations and experimental years (Figure 7).

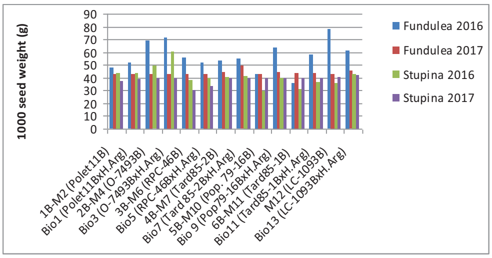


Figure 7. One thousand seed weight of sunflower genotypes, obtained in the years 2016 and 2017 in Fundulea and Stupina

Seed yields (kg/ha) with only a few exceptions were higher in 2017 at Fundulea, a year with rainfall during the whole sunflower growing season and in sufficient quantities in June-July-August.

In the extreme conditions of drought and heat from Stupina we notice the genotype Bio 7, followed by Bio 5 and Bio 13 (Figure 8).

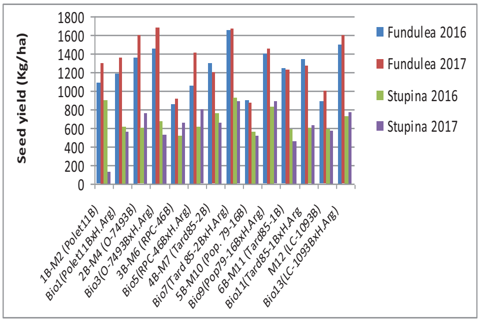


Figure 8. Seed yield (kg/ha) of sunflower genotypes studied in the years 2016 and 2017, in Fundulea and Stupina

CONCLUSIONS

Three newly created sunflower genotypes, Bio 7, Bio5 and Bio 11 (Figure 9), were identified as a result of interspecific hybridization with annual wild species *Helianthus argophyllus*, which had a higher seed yield (g/sunflower plant) compared to the genotypes 4B- M7, 3B-M6 and 6B-M11.

Figure 9. Sunflower genotype, Bio 11, in Fundulea, 2017

The genotypes Bio 7, Bio 9 (Figure 10) and Bio 11 have a good oil content and sunflower genotypes Bio7, Bio 9 and Bio 13 have a better 1000 grain weight in both locations, Stupina si

Fundulea and experimental years 2016 and 2017.

Figure 10. Sunflower genotype, Bio 9, in Fundulea, 2017

For seed yield (kg/ha), genotypes Bio7, Bio 9 and Bio 13 (Figure 11) recorded high yields in both locations, Fundulea and Stupina.

Figure 11. Sunflower genotype, Bio 13, in Stupina, 2017

For all the parameters studied, the newly created genotype, Bio 7 (Figure 12), is superior both compared to the fertility maintenance line 4B-M7 and compared to the other genotypes for all studied parameters.

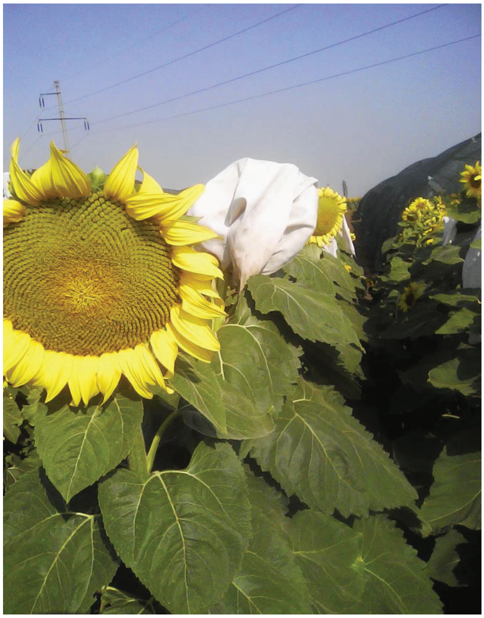


Figure 12. Sunflower genotype, Bio 7, in Fundulea, 2017

REFERENCES

Andrich, G., Balyini, S., Yinnai, A., Silvestri, S., Gallopini, C. (1996). The effect of drought stress on some characteristics of sunflower seeds. *Agricoltura Meditteranea*, *126*(3), 285–291.

Baldini, M., Cecconi, F., Vanozzi, G.P. (1993). Influence of water deficit on gas exchange and dry matter accumulation in sunflower cultivars and a wild species (*Helianthuus argophyllus T&G*). *Helia*, *19*, 1–10.

Barron, J.E. (1991). A factor analysis of plant variables related to yield in sunflower under water stress conditions. *Helia*, *14*(15), 55–64.

Christov, M., 2013. Contribution of interspecific and intergeneric hybridization to sunflower breeding. *Helia*, *36*(58), 1–19. ISSN 2197-0483 online.

Hussain, M.M., Rauf, S., Riaz, M.A., Al-Khayri, J.M., Monneveux, P. (2017). Determination of drought tolerance related traits in *Helianthus argophyllus*, *Helianthus annuus*, and their hybrids. *Breed Sci.*, *67*(3), 257–267. doi: 10.1270/jsbbs.16095.

Merrien, A., Champolivier, L. (1996). Drought Tolerance in Sunflower, Symposium II of the 14th *Procedings of International Sunflower Conference,* Beijing, China, 72–75.

Prohens, J., Gramazio, P., Plazas, M., Dempewolf, H., Kilian, B., Diez, T.J., Knapp, S., Vilanova, S. (2017). Introgressiomics: a new approach for using crop wild relatives in breeding for adaptation to climate change. *Euphytica, 213*(7), 158.

Vrânceanu, A.V. (2000). *Floarea-soarelui hibridă*. Editura Ceres, 205–245.

Saucă, F. (2010 a). *Agricultură ecologică şi biodiversitate (Ecological agriculture and biodiversity)*. Editura EX PONTO.

Saucă, F. (2010 b). Introgression of drought-resistant gene(s) from *Helianthuss argophyllus* to *Helianthus annuus* specie, using embryo rescue techniques. *Romanian Agricultural Research*, *27*, 48–51.

Saucă, F., Lazăr, C. (2016). *Organic farming a promising way of food production*. Capt. 5 - Preliminary results regardining the use of interspecific hybridization of sunflower with *Helianthus argophyllus* for obtained new hybrids with drought tolerance, adapted to organic farming, Editura INTECH, 83–110.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

PRODUCTIVITY AND QUALITY PARAMETERS ON PERSPECTIVE ALFALFA VARIETIES IN SANDY SOIL CONDITIONS

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Abstract

The investigation was performed in the 2016-2018 period, near Plovdiv, Bulgaria by the block method in 4 replications in 10 m^2 lots in a sandy soil conditions. The aim of the research is to study productivity and quality parameters of native (Prista 3) and Dorine -yellow Jacket Rhizobium coated alfalfa varieties (new technology of adding high levels of effective Rhizobia, embedded in a protective polymer matrix, together with a nutrient booster containing all essential minerals and trace elements). The two varieties were treated with growth regulators (Tekamin Max, Fertigrain Foliar and Tekamin Brix) to help plant development under the sandy soil conditions. The results show better protein value, higher carbohydrate and cellulose content and higher green mass yield in all treated variants. Dorine significantly exceeds the values obtained at Prista 3.

Key words*: alfalfa, growth regulators, quality, yield.*

INTRODUCTION

Alfalfa yield formation is complicated process due to the interaction of plants with agro-climatic and soil conditions. For different alfalfa varieties it also depends to a large extent on the values of the productive components - plant height, number of stems per m^2, stem weight, leaf weight, stalk/leaf ratio, etc. which is found in a number of studies (Kertikova & Yancheva, 2000; Yancheva et al., 2001; Johnson et al., 2007; Petkova et al., 2007; Popovic et al., 2007)

The green mass and hay yield also depends on the yield components as plant height, stem thickness, number of shoots per unit area, weight of shoots from one plant, etc., which also change their values under the influence of various factors (Ventorini et al., 2010).

Alfalfa green mass yield is in positive correlation with the leaf surface of a plant. This is because the leaves represent 30-60% of the total fodder yield (Foutz et al., 1976; Hart et al., 1978; Sheaffer et al., 1980; Volonec et al., 1987). Different types of fertilization also have a serious influence on crop yield. The timely application and the appropriate combinations of elements can significantly enhance yield and also the quality of the production (Aguilar et al., 2012; Al-Juhaimi et al., 2014). A positive influence on crop yield can be also influenced by using various organic fertilizers and manure (Aguilar et al., 2012; Al-Juhaimi et al., 2014).

With great importance for symbiotic nitrogen fixation are Rhizobium tuberculous bacteria living in symbiosis with bean plants. On average, these bacteria fix from 0 to 50 kg N/da for a year (Kimmenov, 1994; Russelle et al., 2008). In poor (sandy) soils, nitrogen fixation is poor, and the aplication of these bacteria into the seeds is of particular importance for the regular development of the plants, allowing to achieve results tipical of rich soils.

Using coated seeds, together with the seeds in the soil can be applied variety of elements as pine, molybdenum and cobalt, which have a significant influence on the nutrition of plants (Zehirov & Georgiev, 2001; Zehirov & Georgiev, 2002). Molybdenum positively affects the synthesis of protein substances. (Kaiser et al., 2005).

Application of growth regulators in leguminous crops or leaf fertilizers (Osman et al., 2010) leads to positive changes in productivity, chemical composition, leaf area and pure productiveness of photosynthesis, increasing the symbiotic activity of tuberous bacteria, which directly affects the quality and yield of the crop.

Some authors (Hall et al., 2002) found that the application of certain leaf fertilizers and growth regulators to stimulate branching and increase

yield and quality in lucerne did not increase in same way in specific soil and climatic conditions.
Sammaura and Yaday (2008) studies show a positive effect of foliar fertilizers and regulators on the alfalfa growth, development and yield.
In recent years, many preparations with different physiological effects, connected to the efficiency of the biological nitrogen fixation, have been tested, aiming to regulate productive capacities and the growth of alfalfa in order to increase the yield and quality of the plant production.
Our studies are targeted also to a group products with a regulatory effects on productivity and confirm the above results.

MATERIALS AND METHODS

Scientific research was carried out during the 2016-2018 period at the experimental field of the Crop science Department at the Agricultural University of Plovdiv. To achieve the stated goals, a field experience has been set for establishing the influence of some leaf preparations with a regulatory effect on the productivity and quality of alfalfa. Experience is based on the fractional plot method, in 4 replicates and plot size of 10 m^2. Four preparations and their combinations (Tekamin Max, Fertigrain Foliar, Tekamin Max + Fertigrain foliar and Tekamin Brix) over two alfalfa varieties (Prista 3 and Dorine) have been tested.
The seeding was created and grown by the conventional technology for growing alfalfa for fodder (Yankov et al., 1994).
The obtained data are mathematically processed by the dispersion analysis method through the SPSS program.
The soil on which the experience was condacted is slightly salted, sandy, poorly stocked with nitrogen, moderately loaded with phosphorus, well-stocked with potassium and well-stocked with calcium and magnesium. The content of essential nutrients, combined with neutral pH creates favorable conditions for the development of alfalfa and the nitrogen fixation.
Years of the experience have been characterized as appropriate to the crop development. The lack of drastic cold in winter is a prerequisite for good wintering and garnishing of the crop. All three years are characterized as warm and reasonably well-moistened. These conditions, reveal the potential for high yields at the researched alfalfa varieties.

RESULTS AND DISCUSSIONS

A number of authors point that the height of plants is a varietiy attribute but changes under the influence of various factors (Berg et al., 2007; Wang et al., 2010; Wang et al., 2010; Ventroni et al., 2010).
The analysis of the obtained results shows, that the plants are highest in the first swath, which is typical for the crop. The exception is the first year, when the spring sowing makes highest the second swath. However, average for the researched period is the first, followed by the second and third. In the second year of the experiment, we found a strong *Phytodecta fornicata* attack, as shown on Photo 1. The control of the pest was inferred through earlier mowing without the use of chemical preparations.

Photo 1. *Phytodecta fornicata* attack damages

The tested preparations did not significantly affect the plants height. In both varieties they increase the height of the plants, but no varieties difference was observed (Figure 1). Despite of fact that Dorine variety forms higher stems than Prista in all cuts during the three experimental years, the difference in yield between the two varieties can be explained by the heavier stems (Figure 2), and same results obtained from Volonec et al. (1987), Teixeira et al. (2007), according to which the weight of the stems is one of the important components with a direct effect on the yield of green mass and hay in alfalfa.

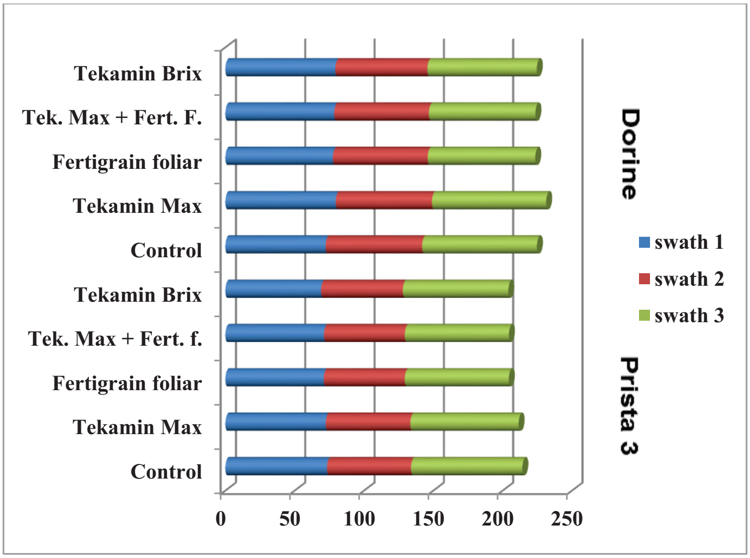

Figure 1. Plants height (average for the research period)

Table 1 shows that treated plants formed heavier stems than those in controls. In the first year spring sowing leads to heaviest stems in the second swath. In this indicator, best results were obtained when treated with Fertigrain Foliar - 24.6 and 25.2 kg ha^{-1} respectively at Prista 3 and Dorine.

Table 1. Stem weight for the period 2016-2018

Variants	Stem weight								
	2016			2017			2018		
	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3
Prista 3									
Control	9.3a	16.9a	5.8a	11b	8.6a	8.3a	10.8b	8.6a	8.9a
Tekamin Max	11.8b	18.7b	8.8c	12.4c	13.5c	11.6c	11.9c	13.2c	10.8c
Fertigrain Foliar	11.1b	24.6d	9.2c	12.3c	9.9b	8.8b	12.6c	10.1b	9.3b
Tekamin Max + Fertigrain Foliar	11.2b	20.8c	7.7bc	11.3b	14.3cd	8.9b	10.8b	13.9cd	9.4b
Tekamin Brix	11.4b	23.5d	7.1b	9.4a	9.9b	8.7b	9.2a	9.7b	9.9b
Dorine									
Control	12.6a	18.2a	6.8a	7.8a	9.5a	9.3a	7.6a	9.7a	9.9a
Tekamin Max	13.2b	21.7b	7.5b	11.5c	13c	13.8d	11.7c	13.2c	14.1d
Fertigrain Foliar	13.2b	19a	8.1bc	9.9b	10.4ab	9.9a	9.6b	10.6ab	10.8ab
Tekamin Max + Fertigrain Foliar	13.8b	25.2c	7.4b	12.5cd	14.5d	12.4c	12.4cd	14d	12.4c
Tekamin Brix	12.7a	21.2b	7.1ab	10.5b	10.2a	11.1b	10.3b	9.9a	11.1b

The difference between the control and the treated variants is strongest in the second swath and decreases in third. During the second and third years, the difference in weight is greatest in the first and second cuts, and the effect of treatment is more pronounced in each subsequent year. This shows that fertilizer activity has increased with cumulation over the years, and this means that their systemic application strengthens the treatment effect.

The differences in the stems weight due to the treatment, with few exceptions, are mathematically proven in both varieties. The only exception is the variant, treated with Tekamin Brix, where the results are contradictory and it can not be said, that treatment with the preparation influences the studied indicator for both varieties.

Many authors consider that the yield of green alfalfa is in positive correlation with the number of leaves. The leaves weigh and number are interrelated, increase the yield, and above all, the quality of the production. Our research confirms this and shows that the treatment proven increases the leaves number in all variants in Prista 3 and most in Dorine.

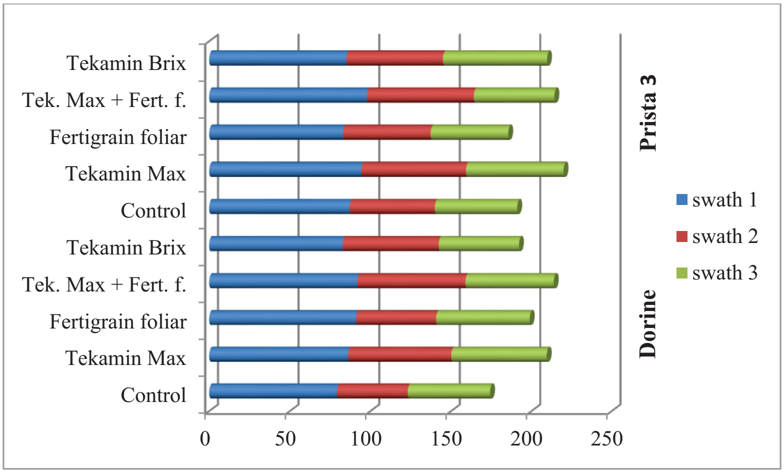

Figure 2. Number of leafs per plant (average for the research period)

Highest leaf number Prista variety forms when Tekamin Max is applied - 224, and Dorine - 216 when treated with the combination of preparations (Figure 2). On average, during the study, both varieties formed highest leaf number at first cut, and values nearly equaling the other two.

The results from Tekamin Brix treatment are quite controversial in both varieties, in some variants the values increase, while other decrease, which means that the preparation does not affect the number of the leaves.

The weight of the leaf is an important indicator for the crop yield formation, especially in well-foliated varieties. The results of Figure 3 show that the treated plants have heavier leaves than untreated, but the differences obtained in most cases are not mathematically proven, indicating that this indicator is largely influenced by climatic conditions. Values at the control are 5.4 g for the Bulgarian variety and 7.1 g for Dorine. Prista 3, realizes highest value when treated with Fertigrain Foliar - 8 g, and Dorine with combination (Tekamin Max + Fertigrain Foliar) - 11 g. The same tendency as the number of leaf counts at plants, treated with Teknamin Brix in both varieties is noted. Treatment does not lead to a one-way change which means, that this preparation does not affect the weight of the leaves.

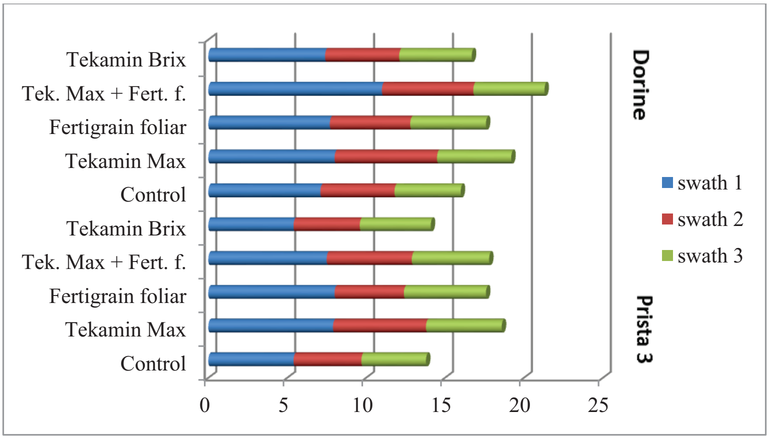

Figure 3. Leaf weight (Average for the research period)

The researched preparations increase the green mass yield in both varieties at all variants with some exceptions in 2017 - the third cut for Prista 3 and the first cut for Dorine but for the period of study in total the differences between the variants were statistically proven (Table 2).

As a result of the treatment, a variety difference is obtained. Dorin formes a higher green mass yield when treated with Tekamin Max, and Prista with the combination of Tekamin Max and Fertigrain Foliar. Highest yield Dorine realizes when treated with Tekamin Max (2016 - 4180 kg ha^{-1}; 2017 - 5550 kg ha^{-1}; 2018 – 5560 kg ha^{-1}) and Prista when treated with Tekamin Max and Fertigrain Foliar (2016 - 5385 kg ha^{-1}; 2017 - 6435 kg ha^{-1}; 2018 - 5679 kg ha^{-1}). Highest yield is obtained in the first swaths 2017 and 2018 and second in 2016.

Comparatively low yield, in the first cut in 2016 is due to strong attack by *Phytodecta fornicata* in the spring of 2016 (see Photo 1), which also required the earlier mowing of the culture. Any other attempt to solve the problem risked the complete compromise of production. In this way, on the one hand, some yield is kept, on the other - an adequate and effective plant protection is carried out without the use of chemical preparations.

Also interesting is the fact that in 2017 and 2018, the yield values of second and third swath are very similar in both study varieties. This is due to the abundant and well distributed rainfall throughout the vegetation.

Table 2. Green mass yield kg ha^{-1}

Variants	Green mass yield								
	2016			2017			2018		
	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3
Prista 3									
Control	1270a	1675a	965a	2070a	1620b	1430a	1968a	1590b	1397a
Tekamin Max	1305b	1760b	1115c	2230c	1760c	1560b	2180c	1811c	1569c
Fertigrain Foliar	1315b	1760b	1155c	2135b	1690b	1450a	2096b	1702b	1387a
Tekamin Max + Fertigrain Foliar	1380c	1880c	1020b	2205c	1665b	1560b	2331c	1688b	1490b
Tekamin Brix	1385c	1725b	1020b	2090a	1405a	1435a	2146a	1496a	1401a
Dorine									
Control	1865a	1925a	1160a	1915a	1665a	1435a	1915a	1665a	1415a
Tekamin Max	1875a	2115c	1325c	2195c	1840c	1600c	2195c	1840c	1567b
Fertigrain Foliar	1850a	2040b	1345c	2185c	1760b	1455b	2185c	1760b	1411a
Tekamin Max + Fertigrain Foliar	1910b	2265d	1210b	2165c	1965d	1580c	2165c	1965d	1549b
Tekamin Brix	1860a	2055b	1195a	2075b	1660a	1460b	2075b	1660a	1440a

The Dorine variety exceeds Prista in green mass yield on average over the study period (Dorine - 5345 kg ha^{-1}, Prista - 4280 kg ha^{-1}). With small exceptions, the same logic is also observed in the results obtained in both the treated and the untreated variants, which gives us reason to characterize Dorine, as a more productive variety (Table 3). An exception to the above with respect to the green mass yield was observed in the treatment with Tekamine Brix in both studied varieties, both in swaths and in total for the researched period. The application of the preparation does not increase the yield, which co-ordinates with the results for leaves weight. This fact and the values observed in hay yields gives us a reason not to recommend the preparation for use in lucerne production.

Table 3. Hay yield kg ha^{-1}

Variants	Hay weigth								
	2016			2017			2018		
	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3	Swath 1	Swath 2	Swath 3
Prista 3									
Control	381a	486.75a	260.6a	558.9a	453.6a	328.9a	1968a	1590b	1397a
Tekamin Max	444.3b	756.8c	379.1c	892c	585.6c	455.6c	2180c	1811c	1569c
Fertigrain Foliar	447.1b	739.2c	427.35c	683.2b	547.7b	348a	2096b	1702b	1387a
Tekamin Max + Fertigrain Foliar	496.8c	827.2d	347.4b	860c	546.1b	449c	2331c	1688b	1490b
Tekamin Brix	484.75c	621b	336.6b	568.8a	435.6a	394.2b	2146a	1496a	1401a
Dorine									
Control	578.2a	785.3a	336.4a	600.7a	500a	389.2a	588a	515a	400.2a
Tekamin Max	603.5ab	909.5b	490.25b	768.3c	607.2b	432b	734.9c	599.1c	451.3b
Fertigrain Foliar	629b	877.2b	484.2b	742.9c	651.2c	363.8a	728.1c	644.4b	389.8a
Tekamin Max + Fertigrain Foliar	691.9c	1019.3c	471.9b	779.4c	707.4d	471.2bc	769.4c	697.3d	479c
Tekamin Brix	632.4b	801.5a	361.35a	643.3b	481.4a	365a	601 a	476.2a	406.6a

The results for hay yield fully correspond to the ones obtained about the green mass. The treatment with preparations leads to an increase the values of the investigated indicator, both in swaths and total for the study period.

This once again confirms the positive influence of treatment on the productivity of the crop and gives us reason to recommend the preparations in our tried doses for use in the practice of alfalfa production. Regardless of the climatically different years, one-way results are clearly visible in both varieties. Again, the highest values were obtained at variants, treated with Tekamin Max at Prista and the combination of Tekamin Max + Fertigrain Foliar at Dorine, with the differences being mathematically proven (Table 4).

Dorine variety significantly exceeds Prista in the first year of the study. This is probably due to the fact that the introduction of Rhizobium bacteria has a strong influence on the initial development of the culture. Dorine variety has a higher crude protein content in the green biomass, compared to Prista 3 in all studied variants. The highest values for both varieties are obtained after treatment with Tekamin Max (23.73) for Dorine variety and 22.18 for Prista 3. In both varieties the treatment increases the protein content.

The treatment did not affect the cellulose content of both tested varieties, regardless of the preparations used. The Dorine variety exceeds Prista 3, but this is due to the heavier stems, which is also confirmed by stem weight data as well as by the yields of green mass.

The quality of lucerne is also determined by the common sugars content. Our studies show an increase in the indicator as a result of treatment in all combinations.

Table 4. Quality parameters of alfalfa green mass (average for the research period)

Variants	Crude protein	Common sugars	Cellulose
	% to an absolutely dry matter		
Dorine			
Control	21.68	4.66	32.28
Tekamin Max	23.73	5.31	32.33
Fertigrain Foliar	22.81	7.36	34.12
Tekamin Brix	23.00	5.63	32.88
Prista 3			
Control	19.24	4.38	32.52
Tekamin Max	22.18	6.12	33.81
Fertigrain Foliar	21.75	6.21	32.57
Tekamin Brix	19.87	6.06	31.22

Sugars increased most significantly with Fertigrain Foliar aplications in both varieties (7.36% for Dorine and 7.36% for Prista 3). The obtained data are confirmed over the hole study period, indicating that Fertigrain foliar increases the sugar content and therefore the quality of lucerne plants.

CONCLUSIONS

The application of the tested products increases the stems weight of both varieties, Dorine forms heavier stems than Prista 3.

There is proven increasing of number and weight of leaves in all treated variants by swaths in Prista 3 and in two at Dorine.

The use of the Tekamine products has been shown to increase both green and hay yield for both studied varieties on one hand and the quality of above-ground biomass on the other, so they can be used in practice for alfalfa production. Highest results for both varieties were obtained by treatment with Tekamin Max and Fertigrain Foliar.

Dorine variety exceed Prista 3 in green mass and hay yield for all 3 researched years.

Using Yellow Jacket Rhizobium coating for seeds on sandy soils increases crop yield, and improve establishment on this variety under difficult conditions.

Using new technology, as adding high levels of effective Rhizobia, essential minerals and trace elements, is easy to improve establishment and increase forage production.

REFERENCES

Aguilar, J.R., Rosales, J., Vergara, V., Flores, F., Iván Pérez, M., Galindo, R., Guadarrama, A., Colín, A. Trujillo, A. (2012). *Terra Latinoamericana*, *30*(3), 213–220.

Al-Juhaimi, F.Y., Hamad, S.H., Al-Ahaideb, I.S., Al-Otaibi, M.M, El-Garawany, M.M. (2014). Effects of fertilization with liquid extracts of biogas residues on the growth and forage yield of alfalfa (*Medicago sativa* L.) under arid zone conditions. *Pakistan Journal of Botany, 46*(2), 471–475.

Hall, M.H, Stout, R.C., Smiles, W.R. (2002). Effects of foliar fertilizers and growth regulators on alfalfa yield and quality. *Crop Management*, *1-5*, 67–72.

Berg, W.K., Cunningham, S.M.,. Brouder, S.M., Joern, B. C., Johnson, K.D,. Santini, J.B. Volenec J.J, (2007). The long-term impact of phosphorus and potassium fertilization on alfalfa yield and yield components. *Crop Science*, *47*, 2198–2209.

Kaiser, B., Kgridley, J., Brady, T., Phillips, S., Tyerman, S. (2005). The Role of Molybdenum in Agricultural Plant Production. *Annals of Botany*, *96*, 745–754.

Kertikova D., Yancheva, H., 2000. Variability of some traits in Lucerne within one breeding cycle. *Genetics and Breding*, *30*, 1–2.

Osman, M., El-Sheekh, M., El-Naggar, A., Gheda, S. (2010). Effect of two species of cyanobacteria as biofertilizers on some metabolic activities, growth, and yield of pea plant. *Biology and Fertility of Soils, 46*(8), 861–875.

Petkova, D., and Panayotova, G. (2007). Comparative study of trifoliolate and multifoliolate alfalfa (*Medicago sativa* L.) synthetic populations. Bulgarian *Journal of Agricultural Science*, *13*, 221–224.

Petkova, D., and Dukic, D. (2007). Performance and stability of some agronomical traits of trifoliate and multifoliate alfalfa germ plasms. *Zbornik radova Instituta za ratarstvo I povrtarstvo*, *44*, 35–38.

Popov, N., Dzimotudis, A., Krastev, Kalapchieva, S. (2010). The effect of RENI products on enzymatic activity of nitrogen metabolism in garden peas. *Bulgarian Journal of Agriculural Science*, *16*(5), 609–614.

Popovic, S., Cupic, T., Grljusic, S., Tucak, M. (2007). Use of variability and path analysis in determining yield and quality of alfalfa. Breeding and seed production for conventional and organic agriculture. *Proceeedings of the XXVI meeting of Eucarpia Fodder Crops and Amenity Grasses Section*, 95–99.

Russelle, M.P., Birr A.S. (2008). Large-scale assessment of symbiotic dinitrogen fixation by crops: soybean and alfalfa in the Mississippi River basin. *Agronomy Journal*, *96*, 1754–1760.

Sheaffer, C.C., Wiersma, J.V., Warnes D.D., Rabas D.L., Lueschen W.E., Ford J.H. (1986). Fall harvesting and alfalfa yield, persistence and quality. *Canadian Journal of Plant Science*, *2*. 329–338.

Ventron,i M., Volenec, J., Cangiano, C. (2010). Fall dormancy and cutting frequency impact on alfalfa yield and yield components, *Field Crop Research, 119* (2, 3), 252–259.

Volonec, J.J., Cherney, H., Jonson, K.D. (1987). Yield components, plant morphology, and forage quality of alfalfa influenced by plant population. *Crop Science*, *27*, 102–109.

Wang, W., Han, Q., Zong, Y., Jia, Z., Ding, R., Wang, J., Nie, J., Min, A., Xiao-li, M. (2010). Regression Analysis on Hay Yield and Relative Characters of Multifoliolate Alfalfa and Trifoliolate Alfalfa.

Yancheva, Ch., Dodunchev, N., Yankov, B. (2001). Structure analysis of some multifoliolate alfalfa (*Medicago sativa*). *XXXVII Croatian Symposium on Agriculture, February 19-23, Opatjia, Croatia. Pamietnik pulawski – materialy konferencji zeszyt 125*, 355–362.

Yankov B., Yancheva, Ch., Petkova, D. (1994). Eucarpia, Symposium on breeding of oil and protein crops, Bulgaria.

Zehirov, G.T., Georgiev, G.I. (2001). Effect of transient short-term boron deprivation of the growth, nodulation and N_2-fixation of soybean plants. *Bulgarian Journal of Plant Physiology*, *27*(1-2), 3–14.

Zehirov, G.T. Georgiev, G.I. (2002). Host – specificity of attachment of Bradyrhizobium japonicum 639 cells to the roots of boron deficient soybean (*Glycine max* L. Merr.) plants related to root cell wall structure, Compt. rend. *Acad. Bulg. Sci., 55*(5), 65–68.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

CHEMICAL COMPOSITION AND YIELD OF SWISS CHARD AS INFLUENCED BY METALLURGICAL SLAG AND FISH FERTILIZER ADDITION TO MARGINAL SOIL

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Abstract

The paper aimed to present the effects of Ca - containing metallurgical slag (MS) and liquid fish (LF) fertilizer appliance on yield and chemical composition of aerial parts of Swiss chard cultivated on marginal eutric cambisol type of soil. The study was performed in semi-controlled glasshouse conditions, and the effects of MS was compared to those of commercial lime materials - ground limestone and hydrated lime, in combination with and without standard mineral and LF fertilizers. The results of the paper indicate that all Ca-materials studied, including MS, along with the studied fertilizers, showed positive effects on the content of main and beneficial biogenic macroelements in chards biomass and its yield. There is a statistically significant tendency of an increase in the content of P, K and C in tested herb in the treatment with LF fertilizer in relation to other treatments. The concentration of trace metals such as Fe and Cd was within the safety limits and allowed concentrations in all the treatments in spite of significant Fe content in MS, which is a highly desirable outcome.

***Key words**: Swiss chard, Eutric Cambisol, chemical properties, metallurgical slag, mineral and organic fertilizers.*

INTRODUCTION

Certain soil properties often change with time, making soils useful relative age indicators. Cambisols combine soils with at least an incipient subsurface soil formation. Transformation of parent material is evident from structure formation and mostly brownish discoloration, increasing clay percentage, and/or carbonate removal. Decarbonation is often followed by leaching (French: *lessive*, washing), which generally presents the translocation of claysized particles in suspension from E to Bt soil horizons (Schaetzl & Anderson, 2005) and is, therefore, an important prerequisite for soil acidification.

Cambisols generally make good agricultural land and are used intensively (IUSS Working Group WRB, 2006). Intensification in their use, in the manner of mineral fertilizers overuse, causes their transformation into marginal soils.

Amelioration of the marginal soils adverse properties, such as low pH, is the first step in creating favorable soil conditions for productive plant growth. Crops vary in their ability to tolerate an acidic soil. In general, Swiss chard (*Beta vulgaris* var. *cicla*), belongs to those vegetables that grow well in a soil of pH around 6.5-6.8 and acidic conditions will stunt its growth. Thus, a necessary lime should be applied. The application of traditional alkaline liming materials such as limestone, dolomite and burnt lime to acid soils for the amelioration of acidity and consequently for improving the crop production is a common practice (Troeh & Thompson, 2005; National Slag Association, 2011; Huang et al., 2012). However, the alkaline properties of metallurgical slag and the need for its sustainable and environmentally acceptable disposal options (Lopez et al., 1995) have prompted its use as a liming material on acid agricultural soils.

Metallurgical slag mainly consists of SiO_2, CaO, Fe_2O_3, FeO, Al_2O_3, MgO, MnO, P_2O_5 and several complex minerals (Motz & Geiseler, 2001). The liming materials in metallurgical slag comprise water-soluble and less water-soluble Ca and Mg compounds. Free Ca in slag reacts rapidly with water to form $Ca(OH)_2$. The $Ca(OH)_2$ will react rapidly with soil acidity.

Anderson (1991) reported yield increases of the certain crops such as sugar cane with calcium silicate slag use. White et al. (1937) reported on field trials in Pennsylvania that crop yields of corn, wheat, oats, buckwheat and soybeans with metallurgical slag use were as good or better than an equivalent amount of limestone. In addition to the liming materials, metallurgical slag contains various concentrations of plant nutrients, such P, S, Mn, Fe, and Mo. Some slags may contain elevated concentrations of trace metals such as Fe, Cd, Cr, Cu, Pb, Mo, Ni and Zn. All of these metals occur naturally in soil, and many are essential plant nutrients. Their concentrations vary in slags from different sources. If concentrations in the slag are similar to soil concentrations, they present no problem. If they are present at substantially higher concentrations in the slag than in the soil, repeated application of the slag could significantly increase soil metal concentrations. This possibly could lead to plant toxicity, increased plant uptake and transfer of metals to animals or humans, or to other environmental problems. Nevertheless, the bioavailability of these metals in slags is very low (National Slag Association, 2001).

As a by-product of an industrial process, metallurgical slag offers considerable cost advantages over commercial limestone. Along with other lime materials (ground slag stone, saturated slag etc.) present in Serbia, metallurgical slag from steel factory (Smederevo, Serbia) can be of great importance.

As in steel industry, there is a great problem with manure waste in aquaculture industry. In recent years this industry had to develop various management strategies to reduce the environmental impacts of aquaculture manure waste. In this regard, to evaluate the potential of aquaculture solid waste for use as a fertilizer, the aquaculture industry and regulatory agencies require analytical data regarding the concentrations of various plant nutrients found in the waste (Naylor et al., 1999).

The aim of this research was to investigate the effect of Ca-containing metallurgical slag, a by-product from steel factory (Smederevo, Serbia), on yield and chemical composition of the aerial parts of chard. The effects of metallurgical slag were compared to those of other lime materials (ground limestone and hydrated lime) in combination with and without standard mineral and organic (liquid fish) fertilizers.

MATERIALS AND METHODS

The research was carried out in pot experiments under semi-controlled conditions in the glasshouse of the Institute of Soil Science (Belgrade, Serbia), during 2015. In the experiments the comparison of the effect of metallurgical slag (MS) with other lime materials (ground limestone and hydrated lime) in combination with and without mineral NPK (N: P: K = 15%: 15%: 15%) and organic (liquid fish - LF) fertilizers were studied. The ground limestone (calcium carbonate or calcite, $CaCO_3$) contains 60% of carbonate. Hydrated lime (slaked lime, $Ca(OH)_2$) reacts very rapidly and has a TNV (Total Neutralizing Value) of 135, thus 740 kg of hydrated lime is equivalent to one ton of ground limestone i.e. the TNV = 135 (Ristow et al., 2010).

The experiment was undertaken with 1.4 kg pot^{-1} of Eutric Cambisol (WRB, 2014), a type of marginal soil from Central Serbia region that has very low pH and poor physical and biological properties (Pivić et al., 2011). The following designed treatments were carried out in three replicates: T1 - control (untreated soil); T2 - NPK mineral fertilizer; T3 - $CaCO_3$; T4 - $Ca(OH)_2$; T5 - MS; T6 - LF fertilizer; T7 - NPK mineral fertilizer + $CaCO_3$; T8 - NPK mineral fertilizer + $Ca(OH)_2$; T9 - NPK mineral fertilizer + MS.

Before sewing the chard, the amount of fertilizers and slag was measured according to the experimental design and mixed with soil (calculated as for 1 ha): composite NPK fertilizer (15: 15: 15) = 500 kg ha^{-1}; LF fertilizer = 170 kg ha^{-1}; $CaCO_3$ = 4 t ha^{-1}; $Ca(OH)_2$ = 2,8 t ha^{-1}; MS = 4 t ha^{-1}. All three Ca-materials with granulation of 0.2 mm were used in the experiment.

Chemical properties and elemental composition of the plowed layer of Eutric Cambisol used in this study (Table 1) were analyzed and determined in our previous studies (Pivić et al., 2011; Stanojković et al., 2011). Accordingly, the soil is characterized by acid reaction, with pH in 1M KCl 4.98, then, potential acidity (Y) and relatively low saturation of CEC, medium to high content of nitrogen, low content of soluble phosphorus, and it is well supplied with available potassium and trace elements - iron, zinc and copper. As for toxic metal cadmium, its concentrations are below the MPV (maximum permissible value).

Table 1. Chemical properties of the studied soil (Pivić et al., 2011; Stanojković et al., 2011)

Chemical parameter	Mean	PLV/MPV* (mg kg^{-1})
pH in 1M KCl	4.98	-
The sum of bases - S (cmol kg^{-1})	21.98	-
Potential acidity - Y´	11.50	-
Cation exchange capacity - CEC (cmol kg^{-1})	29.46	-
Base saturation -V (%)	74.62	-
Total N (mg kg^{-1})	0.28	-
Available P (mg kg^{-1})	79.8	-
Available K (mg kg^{-1})	21.8	-
Available Fe (mg kg^{-1})	63	>25
Available Zn (mg kg^{-1})	2.2	>6
Available Cu (mg kg^{-1})	4.1	>2.5
Available Cd (mg kg^{-1})	0.1	0.8

*PLV - provision limit value for Fe, Zn and Cu (Ankerman & Large, 1977); MPV - maximum permissible value for Cd (OGRS, 2018).

In Serbia, eutric cambisols are mostly medium-heavy soils, with a marked texture difference through the profile. Chemical properties vary depending on the intensity of use, degree of erosion, chemical properties of the parent material, as well as the level of development (Hadžić et al., 2002).

Table 2 displays the chemical composition of metallurgical slag (MS) applied, which was in detail determined in our previous studies (Pivić et al., 2011; Stanojković et al., 2011).

Table 2. Chemical composition of MS (Pivić et al., 2011; Stanojković et al., 2011)

Parameter	Mean value
pH in H_2O	12.48
Total Ca (mg kg^{-1})	26.20
Total CaO (mg kg^{-1})	36.60
Total $CaCO_3$ (mg kg^{-1})	65.80
Available Ca (mg kg^{-1})	17.18
Total Mg (mg kg^{-1})	0.41
Available Mg (mg kg^{-1})	0.07
Total P (mg kg^{-1}, in HNO_3)	0.64
Total P (mg kg^{-1}, in 2% citric acid)	0.61
Total Fe (mg kg^{-1})	15.34
Available Fe (mg kg^{-1})	3.38
Total Mn (mg kg^{-1})	1.80
Available Mn (mg kg^{-1})	3.12
Total Zn (mg kg^{-1})	14.60
Total Cu (mg kg^{-1})	228.8

Accordingly, this material has very alkaline reaction (pH in H_2O = 12.50), with the content of calcium in oxide forms (CaO) from 33-45 mg kg^{-1}, of which about 50% is easily soluble in 1 M ammonium acetate; content of the total magnesium is about 0.40 mg kg^{-1} and it was mainly in forms of MgO (0.07 mg kg^{-1}); total phosphorous contained in the material is about 0.60 mg kg^{-1}, where nearly all the amount is in available forms for plants; content of the total iron is high (about 15 mg kg^{-1}), with noticeable lower amounts of its soluble forms; manganese is present in total amount of about 1.8 mg kg^{-1}, but with noticeable low amounts of soluble forms; zinc is contained in lower amounts (10-20 mg kg^{-1}), while the content of copper is a little higher (about 200 mg kg^{-1}).

The settleable faecal fish waste, used as liquid fertilizer in this research, was obtained from the farm growing rainbow trout (*Oncorhynchus mykiss*) in village Krupac, municipality of Pirot, Serbia. Chemical composition of the liquid fish (LF) fertilizer included the following analysis: total nitrogen (N), carbon (C) and sulphur (S) were determined on elemental CNS analyzer Vario EL III (Nelson and Sommers, 1996); available phosphorus (P) was determined by spectrophotometer and potassium (K) by flame emission photometry, using AL-method (Đurđević, 2014a), after they were heated to boiling with the mixture of concentrated sulfuric and perchloric acids.

Swiss chard seedlings were grown according to the standard growing methods from March, 31st until July, 27th in 2015, when all studied relevant parameters of the plant growth were measured/analyzed. Biomass from each experimental variant and replicates was taken, air-dried and weighed, after which it was dried for 2 hours at 105°C and weighed again. The following chemical parameters of the aerial plant parts were analyzed: contents of nitrogen (N), carbon (C) and sulphur (S) were determined on elemental CNS analyzer Vario EL III (Nelson and Sommers, 1996); phosphorus (P) and potassium (K) concentrations were determined by "wet" combustion, i.e. they were heated to boiling with the mixture of concentrated sulfuric and perchloric acids, after which, in the obtained solution, P was determined by spectrophotometer with molybdate (Đurđević, 2014b), and K - by flame emission photometry (Đurđević, 2014c); in the determination of investigated trace biogenic elements - iron (Fe), zinc (Zn) and copper (Cu), as well as cadmium (Cd) as the toxic heavy metal, plant material was converted to a solution by "dry" combustion, i.e., first by heating for several hours at 550ºC and then by treating the obtained ash with hydrochloric acid, after which these elements were determined by atomic absorption spectrometry - AAS (Wright & Stuczynski, 1996).

The evaluation of an influence of studied treatments on the analyzed chemical composition of the plant material was carried out using the analysis of variance (SPSS 20.0, Chicago, USA), followed by Duncan's Multiple Range Test (DMRT). Significant differences between means were tested by the LSD test at P = 0.05.

RESULTS AND DISCUSSIONS

By analyzing the content of the main chemical constituents of faecal fish waste in this study, it was determined its moderate to high quality for application as an organic fertilizer, containing 3.3 mg kg^{-1} of nitrogen, 36.5 mg kg^{-1} of carbon, 0.36 mg kg^{-1} of sulphur, 10.2 mg 100 g^{-1} of available phosphorus and 0.12 mg 100 g^{-1} of available potassium (Table 3).

Table 3. Chemical composition of the liquid fish (LF) fertilizer

Parameter	Average value
Total N (mg kg^{-1})	3.33
Total C (mg kg^{-1})	36.5
Total S (mg kg^{-1})	0.36
Available P_2O_5 (mg 100 g^{-1})	10.2
Available K_2O (mg 100 g^{-1})	0.12

According to the certain previous studies (Olson, 1992; Westerman et al., 1993) fish fertilizers tend to be highly variable in their chemical content, which is also the case with other manures. The various results demonstrated a wide variability in the general chemical composition of fish manure, although its macronutrients composition is of primary interest, given the end use of the manure as an agricultural fertilizer. As stated by Olson (1992), fish manure contains moderate amounts of essential plant nutrients in dry matter (2.83 mg kg^{-1} N; 2.54 mg kg^{-1} P; 0.10 mg kg^{-1} K; 6.99 mg kg^{-1} Ca; 0.53 mg kg^{-1} Mg), which is similar to the results of the current study, with an exception of phosphorus (the content was much higher in the present study - 10.2 mg kg^{-1}). Nevertheless, as suggested by Naylor et al. (1999), it is difficult to compare the values from the present study with those from others because of the differences in conditions under which the solids were produced, separated, stored and collected.

The results of the main and beneficial biogenic macroelements content in aerial parts of the tested vegetable (Table 4) show the statistically significant differences between the treatments at $P<0.05$, that are due to a higher accumulation of some elements and their mobilization from natural soil reserves primarily, as well as influenced by the additional LF fertilizer amendment and lime materials in combination with mineral ferilizer. However, these treatments had positive effect on N, P, K, C and S contents in relation to the control. There is a statistically significant tendency of an increase in the content of P, K and C in tested plant material in the treatment with LF fertilizer in relation to other treatments.

Improved organic and mineral nutrition in combination with lime materials including MS would explain the promotion of root and plant growth which led to promotion of biomass yield. The data on yield of the Swiss chard dry biomass were in accordance with chemical parameters, meaning that the yield was significantly higher in variants which included LF fertilizer and NPK mineral fertilizer in combination with MS and lime material in the form of Ca(OH) (Table 4).

As stated by Riesen and Feller (2005), the nature of applied treatments and their combinations have a great impact on trace metals accumulation, their mobility and storing capacity in plant tissues. Some trace elements may pose a toxicity threat if present at elevated levels as their availability and mobility increases under acidic conditions (Pawłowski, 1997).

The concentration of trace metals in Swiss chard aerial parts showed that there are statistically significant differences between different treatments at $P<0.05$ (Table 4). With an exception of plants from LF fertilizer treatment, it was determined elevated and critical concentrations of Fe in all other studied variants plants, although these concentrations were below the toxic value (Table 5). Nevertheless, there was not found higher accumulation of Fe in tested plants in the treatments where metallurgical slag was applied in spite of its significant content in this liming material. The content of Cu was in the range of normal and critical concentrations in chard plants from all variants studied except from control, where the content of Cu in chard was a little above the toxic value of 20 mg kg^{-1} per dry biomass (Table 4). The content of Zn and Cd in plants from all variants were in the range of normal and critical concentrations, but below the toxic (Table 5), which is a highly desirable outcome since Cd is a highly mobile element and can be easily translocated to the aerial plant parts (Sipter et al., 2008). The obtained results of Pajević et al. (2018) indicate that heavy metal accumulation significantly depends not only on soil quality, but also on plant species and in accordance, translocation of heavy metal ions from roots to aboveground plant parts. Different distribution of heavy metals within the plant was also previously reported by Al Jassir et al. (2005).

The edible parts of leafy green vegetables, such as Swiss chard, showed higher potential to accumulate heavy metals in comparison to storage organs and fruits (Sharma et al., 2008), which could explain elevated concentrations of the studied trace elements in this plant species.

Table 4. Available macroelements in Swiss chard and the yield of dry biomass depending on the treatment applied

Treatments	Macroelements (mg kg^{-1} of dry biomass)*					Yield (g pot^{-1})
	N	P	K	C	S	
T1 - control	2.17±0.04[i]	0.27±0.04[h]	3.59±0.01[f]	36.77±0.05[e]	0.75±0.05[d]	9.09±0.10[b]
T2 - NPK	3.81±0.03[b]	0.31±0.01[e]	4.03±0.12[c]	36.48±0.07[f]	1.07±0.04[a]	9.34±0.12[b]
T3 - $CaCO_3$	2.75±0.06[g]	0.37±0.02[cd]	3.83±0.06[d]	37.33±0.37[d]	0.96±0.01[b]	9.08±0.09[b]
T4 - $Ca(OH)_2$	3.07±0.05[e]	0.35±0.01[d]	3.83±0.03[d]	38.27±0.39[c]	1.03±0.03[a]	9.10±0.12[b]
T5 - MS	2.62±0.02[h]	0.33±0.03[de]	3.83±0.01[d]	38.21±0.10[c]	0.97±0.02[b]	9.26±0.13[b]
T6 - LF fertilizer	2.90±0.05[f]	0.49±0.01[a]	4.42±0.09[a]	39.75±0.04[a]	1.04±0.01[a]	9.72±0.08[a]
T7 - NPK fertilizer + $CaCO_3$	3.20±0.02[d]	0.34±0.02[de]	3.70±0.01[e]	37.40±0.10[d]	1.07±0.02[a]	9.13±0.13[b]
T8 - NPK fertilizer + $Ca(OH)_2$	4.23±0.07[a]	0.44±0.02[b]	4.13±0.02[b]	38.83±0.16[b]	0.81±0.03[c]	9.76±0.20[a]
T9 - NPK fertilizer + MS	3.32±0.08[c]	0.40±0.02[c]	3.38±0.02[g]	38.56±0.11[bc]	1.07±0.02[a]	9.81±0.18[a]
P value	***	***	***	***	***	***
LSD (0.05)	0.105	0.034	0.066	0.449	0.045	0.226

*means ± standard deviation; LSD - least significant difference; value followed by the same letter in a column is not significantly different at $P < 0.05$.

Table 5. Trace elements in Swiss chard biomass depending on the treatment applied

Treatments	Trace metals (mg kg^{-1} of dry biomass)*			
	Fe	Zn	Cu	Cd
T1 - control	302.33±2.08[c]	183.78±1.55[a]	22.61±0.03[a]	3.74±0.03[a]
T2 - NPK	465.33±17.93[b]	129.95±8.21[b]	13.50±0.18[d]	3.25±0.05[b]
T3 - $CaCO_3$	259.33±5.03[d]	76.27±8.76[ef]	13.46±0.16[d]	1.52±0.03[h]
T4 - $Ca(OH)_2$	236.33±12.50[d]	85.55±4.50[de]	15.25±0.16[b]	1.93±0.04[f]
T5 - MS	325.67±2.52[c]	67.49±7.79[f]	11.61±0.14[e]	2.04±0.06[e]
T6 - LF fertilizer	123.33±9.45[e]	91.88±5.41[cd]	11.33±0.10[f]	2.58±0.01[c]
T7 - NPK fertilizer + $CaCO_3$	528.67±16.50[a]	30.83±2.93[g]	7.39±0.15[h]	1.72±0.02[g]
T8 - NPK fertilizer + $Ca(OH)_2$	545.00±36.01[a]	41.96±7.17[g]	14.56±0.04[c]	1.31±0.01[i]
T9 - NPK fertilizer + MS	309.67±14.22[c]	100.00±10.33[c]	10.01±0.01[g]	2.41±0.09[d]
P value	***	***	***	***
LSD (0.05)	17.864	5.318	0.111	0.057
Reference value				
Normal	50[1]	15[3]	3[3]	<0.1-1[3]
Critical	250[1]	150[2]	15[2]	5[2]
Toxic	600[2]	200[2]	20[2]	10[2]

*means ± standard deviation; LSD - least significant difference; value followed by the same letter in a column is not significantly different at $P < 0.05$; Literature source: [1]Schulze et al. (2005), [2]Kastori et al. (1997), [3]Kloke et al. (1984).

CONCLUSIONS

Fertilizer management is of great importance in vegetable production. The results of the paper indicate that all Ca-materials studied, including metallurgical slag, along with the studied mineral and liquid fish fertilizers, showed positive effects on the content of main and beneficial biogenic macroelements in aerial biomass of Swiss chard. There is a statistically significant tendency of an increase in the content of P, K and C in tested plants in the variant with liquid fish fertilizer in relation to other treatments. Similar to the content of macroelements, the biomass yield was significantly higher in variants which included liquid fish fertilizer and NPK mineral fertilizer in combination with metalurgical slag and lime material [particularly in the Ca(OH) form]. The content of trace elements in Swiss chard plants was mainly in the range of normal and critical concentrations, except for Cu from control plants, where its content was a little above the toxic value of 20 mg kg^{-1} per dry biomass (Kastori et al., 1997). Nevertheless, there was not found higher accumulation of Fe in tested plants in the treatments where metallurgical slag was applied in spite of its significant content in this liming material. Generally, it was estimated that the studied metallurgical slag of the standardized chemical composition can be added to marginal soils toward their fertility amelioration/improving without adverse effects. It should be noted that liquid fish fertilizer of the stated chemical composition its self showed an excellent effect on all parameters tested and could be a part of an effective alternative to chemical fertilization in vegetable production under semi-controlled conditions.

ACKNOWLEDGEMENTS

This research work was carried out with the support of Ministry of Education, Science and Technological Development of the Republic of Serbia and also was financed from Project TR-37006.

REFERENCES

Al-Jassir, M.S., Shaker, A., Khaliq, M.A. (2005). Deposition of heavy metals on green leafy vegetables sold on roadsides of Riyadh city, Saudi Arabia. *Bulletin of Environmental Contamination and Toxicology*, *75*(5), 1020–1027.

Anderson, D.L. (1991). Soil and leaf nutrient interactions following application of calcium silicate slag to sugarcane. *Nutrient Cycling in Agroecosystems*, *30*(1), 9–18.

Ankerman, D., Large, R. (1977): Minor elements. In D. Ankerman (Ed.), *Soil and plant analysis* (pp. 34–45). Madison, Wisconsin, USA: Soil Science Society of America, Inc.

Đurđević, B. (2014a). Determination of available phosphorus and potassium using AL-method. In D. Miklavčić (Ed.), *Manual in plant nutrition* (pp. 25–28). Osijek, Croatia: Faculty of Agriculture.

Đurđević, B. (2014b). Spectrophotometric determination of phosphorus. In D. Miklavčić (Ed.), *Manual in plant nutrition* (pp. 60–61). Osijek, Croatia: Faculty of Agriculture.

Đurđević, B. (2014c). Determination of K, Ca, Mg and Na. In D. Miklavčić (Ed.), *Manual in plant nutrition* (pp. 62–63). Osijek, Croatia: Faculty of Agriculture.

Hadžić, V., Nešić, Lj., Belić, M., Furman, T., Savin, L. (2002). Potentional of soils in Serbia. *Tractors and Power Machines*, *7*(5), 43–51.

Huang, Y, Guoping, X, Huigao, C, Junshi, W, Yinfeng, W, Hui, C. (2012). An overview of utilization of steel slag. *Procedia Environmental Science*, *16*, 791–801.

IUSS Working Group WRB (2006). *World reference base for soil resources, 2nd edition*. World Soil Resources Reports No. 103, Rome, Italy: FAO. Retrieved June 21, 2011, from http://www.fao.org/3/a-a0510e.pdf.

Kastori, R., Petrović, N., Arsenijević-Maksimović, I. (1997). Heavy metals and plants. In R. Kastori (Ed.), *Heavy metals in the environment* (pp. 196–257). Novi Sad, Serbia: Institute of Field and Vegetable Crops.

Kloke, A., Sauerbeck, D.R., Vetter H. (1984). The contamination of plants and soils with heavy metals and the transport of metals in terrestrial food chains. In J. O. Nriagu (Ed.), *Dahlem Konferenzen: Changing metal cycles and human health* (pp. 113–141). Berlin, Heidelberg, New York, Tokyo: Springer-Verlag.

Lopez, F. A., Balcazar, N., Formoso, A., Pinto, M., Rodriguez, M. (1995). The recycling of Linz-Donawitz (LD) converter slag by use as a liming agent on pasture land. *Waste Management and Research*, *13*(5), 555–568.

Motz, H., Geiseler, J. (2001). Products of steel slags an opportunity to save natural resources. *Waste Management*, *21*(3), 285–293.

National Slag Association (2011). *Use of steel slag in agriculture and for reclamation of acidic lands*. Retrieved February 10, 2010, from http://www.nationalslag.org/sites/nationalslag/files/ag_guide909.pdf.

Naylor, S., Moccia, R.D., Durant, G.M. (1999). The chemical composition of settleable solid fish waste (manure) from commercial rainbow trout farms in Ontario, Canada. *North American Journal of Aquaculture*, *61*(1), 21–26.

Nelson, D.W., Sommers, L.E. (1996). Total carbon, organic carbon, and organic matter. In D. L. Sparks (Ed.), *Methods of soil analysis, part 3* (pp. 961–1010). Madison, Wisconsin, USA: SSSA.

OGRS (1994). *Official Gazette of Republic of Serbia: Regulation on limit values of pollutants, harmful and dangerous substances in soil*, 30/2018.

Olson, G.L. (1992). *The use of trout manure as a fertilizer for Idaho crops*. Idaho Falls, Idaho, USA: Center for Environmental Monitoring and Assessment, EG&G Idaho Inc.

Pajević, S., Arsenov, D., Nikolić, N., Borišev, M., Orčić, D., Župunski, M., Mimica-Dukić, N. (2018). Heavy metal accumulation in vegetable species and health risk assessment in Serbia. *Environmental Monitoring and Assessment*, 1*90*(8), 459. doi: 10.1007/S10661-018-6743-Y.

Pawłowski, L. (1997). Acidification: its impact on the environment and mitigation strategies. *Ecological Engineering*, *8*(4), 271–288.

Pivić, R., Stanojković, A., Maksimović, S., Stevanović, D. (2011). Improving the chemical properties of acid soils and chemical composition and yield of spring barley (*Hordeum vulgare* L.) by use of metallurgical slag. *Fresenius Environmental Bulletin*, *20*(4), 875–885.

Riesen, O., Feller, U. (2005). Redistribution of nickel, cobalt, manganese, zinc and cadmium via the phloem in young and maturing wheat. *Journal of Plant Nutrition*, *28*(3), 421–430.

Ristow, P.L., Foster, J., Ketterings, Q.M. (2010). *Lime guidelines for field crops: Tutorial workbook*. Ithaca, New York, USA: Cornell University, Department of Animal Science. Retrieved March 19, 2019, from http://nmsp.cals.cornell.edu/projects/curriculum.html

Schaetzl, R.J., Anderson, S. (2005). *Soils: genesis and geomorphology*. New York, USA: Cambridge University Press.

Schulze, E.D., Beck, E., Müller-Hohenstein, K., Lawlor, D., Lawlor, K., Lawlor, G. (2005). *Plant Ecology*. Berlin, Heidelberg, Germany: Springer-Verlag.

Sharma, R.K., Agrawal, M., Marshall, F.M. (2008). Heavy metal (Cu, Zn, Cd and Pb) contamination of vegetables in urban India: A case study in Varanasi. *Environmental Pollution*, *154*(2), 254–263.

Sipter, E., Rozsa, E., Gruiz, K., Tatrai, E., Morvai, V. (2008). Site-specific risk assessment in contaminated vegetable gardens. *Chemosphere*, *71*(7), 1301–1307.

Stanojković, A., Pivić, R., Maksimović, S. (2011). *Metallurgical slag use effects on soils and plant chemical properties*. Saarbrücken, Germany: LAP Lambert Academic Publishing.

Troeh, F.R., Thompson, L.M. (2005). *Soils and fertility, 6th edition*. New Jersey, USA: Blackwell Publishing.

Westerman, P.W., Hinshaw, J.M., Barker, J.C. (1993). Trout manure characterization and nitrogen mineralization rate. In J-K. Wang (Ed.), Techniques for modern aquaculture (pp. 35–43). St. Joseph, Michigan, USA: American Society of Agricultural Engineers.

White, J.W., Holben, F.J., Jeffries, C.D. (1937). *The agricultural value of specially prepared blast-furnace slag, bulletin No 341*. Pennsylvania, USA: Agricultural Experiment Station, Pennsylvania State College.

WRB (2014). *World reference base for soil resources: International soil classification system for naming soils and creating legends for soil maps*. World Soil Resources Reports No. 106, Rome, Italy: FAO. Retrieved March 22, 2019, from http://www.fao.org/3/i3794en/I3794en.pdf.

Wright R.J., Stuczynski T. (1996). Atomic absorption and flame emission spectrometry. In D. L. Sparks (Ed.), *Methods of soil analysis, part 3* (pp. 65–90). Madison, Wisconsin, USA: SSSA.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

ASSESS OF THE IMPACT OF FERTILIZATION ON WHEAT PROTEIN AND ENERGY NUTRITION

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Abstract

The purpose of this study is based of assessing the influence of leaf fertilizers on the two common wheat varieties. The fertilization was started with the liquid fertilizers Lactifrost, Lactofol base and Wuxal Grano. These are leafy liquid fertilizers that are used to nourish crops. A basic fertilization with ammonium nitrate was also carried out. The study options are as follows: 1. Without fertilization; 2. Ammonium nitrate (N14); 3. Lactifrost - l l/da; 4. Lactifrost + Lactofol base - 1.0 l/da + 0.5 l/da; 5. Lactofol base - 0.5 l/da .; 6. Wuxal Grano – 0.400 l/da; 7. Wuxal Grano - 0.400 l/da + 0.200 l/da. The results of study show that the crude protein content ranges from 128.50-143.94 g/kg DM in the Enola variety and from 115.93 to 127.34 g/kg DM in the Illico variety. The introduction of Wuxal Grano slurry increased the crude protein content by 9.1 and 12.0% relative to the control. As a result of the correlation analysis, a very high correlation (r = 0.947-0.993) was found between CP and PDI for both common wheat varieties.

Key words: *common wheat, energy nutrition, fertilization, protein feed.*

INTRODUCTION

Wheat is one of the traditional and economically important crops for many regions around the world. Protein is the main quantitative factor determining the quality of wheat grains. In this connection, factors that affect protein levels are of particular importance.

Increasing grain protein content is a topical issue in the world of nutrition (Uauy et al., 2006; Mangova et al., 2007). Main criteria for selection of varieties are resistance to abiotic and biotic stress combined with high productive potential and grain quality. (Panayotov et al., 2004; Yanchev et al., 2012).

A number of studies have found a positive correlation between nitrogen fertilization levels and protein content in the grain (Kelley & Sweeney, 2007; Guangkail et al., 2009). According to Kelley and Sweeney (2007), total nitrogen in the grain is strongly influenced by nitrogen fertilization levels and by the type of prior culture.

A positive correlation between productivity, grain protein content and fertilizer levels (N_{12} and N_{18}) has been found in post-flowering feeding (Montemurro et al., 2007). Results of experiments with different fertilizer norms to assess the impact of different levels on inorganic (0, 80, 160 and 240 kg nitrogen ha^{-1}) and organic (0, 30 and 60 mg municipal waste compost ha^{-1}) fertilizers on the yield of wheat and protein content have shown that high yields and levels of protein and gluten in the grain (Tayebeh et al., 2010) have been achieved with fertilization with 160 kg ha^{-1} nitrogen.

The current study has a purpose to assess the correlation between the chemical composition of wheat grain and the energy and protein feed of ruminant and non-ruminant wheat typical of the two common wheat varieties that under the influence of leaf fertilizers, imported through vegetation.

MATERIALS AND METHODS

For the purpose of the study, authors used two-year data from field experiments, conducted in the experimental field of the Faculty of Agriculture, Trakia University of Stara Zagora, Bulgaria. The experiments were carried out on a meadow cinnamon soil.

The trials were based on tripartite plotting, with three replicates of fertilization and liquid fertilizer feed. The fertilization was started with the liquid fertilizers Lactifrost, Lactofol base and Wuxal Grano.

These are leafy liquid fertilizers that are used to nourish crops. A basic fertilization with ammonium nitrate was also carried out. The study options are as follows: 1. Without fertilization, 2. Ammonium nitrate (N_{14}); 3. Lactifrost - 1 l/da; 4. Lactifrost + Lactofol base - 1.0 l/da + 0.5 l/da; 5. Lactofol base - 0.5 l/da; 6. Wuxal Grano - 0,400 l/da; 7. Wuxal Grano - 0.400 l/da + 0.200 l/da

Table 1. Content of macro and micro elements in leaf fertilizers

Foliar fertilizers	gL^{-1}					mgL^{-1}				
	N*	P_2O_5	K_2O	SO_3	MgO	B	Cu	Mn	Mo	Zn
Lactofol base	101	29.4	50.9	1.36	-	305	203	226	23	452
Lactifrost	13.8	42.4	37.9	2.12	-	477	106	106	2120	64
Wuxal Grano	219	-	-	365	29	-	0.0043	0.0043	-	0.0146
* NO_3-N + NH_4-N + NH_2-N ($g\ l^{-1}$): 22.6 and 13.8 + 11.3 and 6.4 + 67.8 + 0.3										

The experiment included two varieties of common wheat - *Enola* and the introduced *Illico* variety. The experience was brought out during the period 2015-2016. After grain harvesting, the grain chemistry of the common wheat varieties was analyzed. The chemical analysis of the grain was carried out by the Weende method. The levels of crude protein (CP), crude fibre (CF), ether extract (EE), nitrogen free extract (NFE), and mineral substances in the grain of the two studied varieties were determined.

The technology to extract the field survey is standard for the area, except for the fertilization and feeding of common wheat.

By the formulas of Todorov et al. (2004, 2007) the FUM, FUG and PDI content of ruminants were calculated.

$GE = 0,0242\ CP + 0,0366\ EE + 0,0209\ CF + 0,017\ NFE$

$ME = 0,0152\ DP + 0,0342\ DEE + 0,0128\ DCF + 0,0159\ DNFE$

$q = ME / GE$

$FUM = ME\ (0,075 + 0,039q)$

$FUG = ME\ (0,04 + 0,1q)$

$PDI = 1,11CP\ (1 - Deg)\ Dsi + 0,093\ FOM$

$FOM = DOM - DEE - FP - CP\ (1 - Deg)$

$FP = 250 – 0,5\ DM$

$BPR = CP(Deg – 0,1) – 0145\ FOM$

where: GE - gross energy, CP - crude protein, EE - ether extract, CF - crude fibre, NFE - nitrogen free extract, ME - metabolizable energy, DP - digestible protein, DEE - digestible ether extract, DF - digestible fibre, DNFE - digestible nitrogen free extract, Deg - degradability of dietary protein in the rumen, FOM - fermentable organic matter, DOM - digestible organic matter, PDI - protein digestible in (small) intestine, Dsi - digestibility in small intestine, Deg - degradability of dietary protein in the rumen.

Digestible energy (DE) and metabolizable energy (ME) values for pigs and poultry were calculated using the equations (Todorov et al., 2004):

$DEpg = 0.0242\ DP + 0.0394\ DEE+0.0184\ DCF + 0.0170\ DNFE$

$MEpg = 0.0210\ DP + 0.0374\ DEE+0.0144\ DCF + 0.0171\ DNFE$

$DEp = 0.0239\ DP + 0.0398\ DEE+0.0177\ DCF + 0.0177\ DNFE$

$MEp = 0.0178\ DP + 0.0397\ DEE+0.0177\ DCF + 0.0177\ DNFE$

Experimental data were processed by a correlation analysis, which established and evaluated the relationship between the studied indicators. The same is expressed by the correlation coefficient r, determined through the statistical program SPSS 13.

The correlation dependencies are a product of the mathematical and statistical processing of Genchev's output data and others (1975).

RESULTS AND DISCUSSIONS

The present material presents the data on the organic matter contents in the grain of two common wheat varieties obtained by Weende analysis. The analysis of the levels of the qualitative indicators in the different feeds of common wheat shows that the variance under the influence of the studied factors (liquid fertilizers) is poor for both varieties.

The content of crude protein in the grain of common wheat is a key quality indicator. For Enola variety, the content ranges from 128.50 to 143.94 g/kg DM, and Illico variety from 115.93 to 127.34 g/kg DM. The increase due to fertilizer use during crop vegetation is in a narrow range. The variation in the range of 109 to 130 g/kg DM was also found by Zilic et al. (2010). For bigger variation in the content of the CP reports Kim et al. (2003) in Australian wheat varieties - 9.7 to 19.1% CP and Dapoza (2006) in Spanish wheat varieties - 7.6 to 15.1% CP.

In Enola, crude protein levels are higher than the fertilizer control with 4.6-12.0%, while in Illico the increase is up to 9.1% relative to the control. From the studied factors, the fertilization with Wuxal Grano has a more significant impact on the crude protein yield (5.3-12.0 %).

The crude fat contents – 16.83-24.32 g/kg DM of the evaluated wheat varieties were similar to reported by Sauvant et al. (2004) and Lasek et al. (2011).

Crude fibre reduce the digestibility of the feed and thus reduce its nutritional value. The value of the CF was established in the range of 15.45 to 19.86 g/kg DM and they are consistent with the established values of Alijosius et al. (2016) and ~~are~~ lower than those established by Zijlstra et al. (1999) and Anjum et al. (2014).

The concentrations of NFE – 798.05-831.32 g/kg DM were in general agreement with the data of Anjum et al. (2014), who was reported, that for 19 different varieties of wheat the NFE in grain ranged from 78.78% to 82.92% DM.

A major indicator of the nutritional value of feed is their energy and protein nutrition. In ruminants, two units of energy nutrition assessment are used: feed unit for growth (FUG), feed unit for milk (FUM).

Protein nutrition is determined by the amount of protein, truly digestible in the intestine - protein digestible in (small) intestine (PDI). This indicator takes into account the contribution of feed to meet the animal's protein needs.

Table 2. Chemical composition of the grain of common wheat, g/kg DM

Var.	Rep.	CP	EE	CF	NFE
Enola	1	128.50	20.75	17.59	813.52
	2	134.46	20.57	18.73	809.77
	3	135.80	21.10	15.45	809.68
	4	137.53	24.32	17.37	798.05
	5	136.60	23.93	18.52	804.43
	6	143.76	21.16	20.07	799.23
	7	143.94	22.65	17.00	800.70
Illico	1	116.72	21.04	19.86	828.89
	2	115.93	17.25	20.36	831.32
	3	119.90	17.74	18.85	826.64
	4	117.10	18.66	17.38	829.49
	5	119.21	19.83	18.55	826.93
	6	122.94	19.38	18.90	824.18
	7	127.34	16.83	18.46	820.75

The FUM content in 1 kg DM ranges from 1.62 to 1.64 for Enola and 1.65 to 1.67 for Illico. FUG analysis for Enola shows slight variation at 1.46-1.47, and 1.41-1.49 at Illico.

The data show the weak influence of the liquid fertilizers studied on FUM and FUG. In fact, the content of raw nutrients in the grain of common wheat under the influence of feeding with various liquid fertilizers during vegetation changes in narrow limits.

For pigs and poultry, the feed content of digestible and metabolizable energy is used as an indicator of the feed energy feed.

Table 3. Energy and protein value of common wheat for ruminants, for pigs and poultry in 1 kg DM

Var.	Rep.	Ruminant animals			Non-ruminant animals			
		FUM	FUG	PDI	DEp	MEp	DEpg	MEpg
Enola	1	1.47	1.64	101.50	15.81	15.18	16.40	16.10
	2	1.47	1.64	102.65	15.86	15.21	16.47	16.15
	3	1.47	1.64	102.75	15.89	15.22	16.49	16.17
	4	1.46	1.63	102.26	15.82	15.15	16.43	16.11
	5	1.47	1.64	102.67	15.90	15.23	16.52	16.19
	6	1.46	1.62	103.99	15.89	15.18	16.50	16.16
	7	1.47	1.63	103.98	15.94	15.24	16.56	16.22
Illico	1	1.49	1.67	100.20	15.83	15.26	16.43	16.16
	2	1.48	1.66	100.25	15.77	15.20	16.35	16.09
	3	1.48	1.65	100.70	15.78	15.19	16.36	16.09
	4	1.48	1.66	100.20	15.79	15.22	16.37	16.10
	5	1.49	1.66	100.54	15.82	15.24	16.41	16.14
	6	1.48	1.66	101.20	15.84	15.24	16.43	16.15
	7	1.48	1.65	101.91	15.81	15.19	16.39	16.10

DEpg - digestible energy for pigs, MEpg - metabolizable energy for pigs,
DEp - digestible energy for poultry, MEp - metabolizable energy for poultry

The data on the PDI content (Table 3) show that the liquid leaf fertilizers studied did not affect the content. In the Enola variety it ranges from 101.50-103.99 g/kg DM, and in Illico the variation is in the range of 100.20-101.91 g/kg DM on average during the field experience. The increase in PDI content for both varieties ranged from 1.7-2.4 %.

Table 3 also provides the calculated digestate and exchange rate data for non-ruminant pigs and poultry in 1 kg DM. The digestible energy content of the birds varies between two wheat varieties ranging from 15.77 to 15.94 MJ/kg, and in the pigs range from 16.35 to 16.56 MJ/kg DM. Mollah et al. (1983), Wiseman (2000), McCracken et al. (2002) establish similar values for DEp - 8.49 to 15.9 MJ/kg DM.

The differences are extremely insignificant. The swine content is higher, but the differences are again negligible. The values of the exchange energy also vary within narrow limits. Again, the trend in digestible energy levels is higher in pigs (16.09-16.22) MJ/kg DM.

After the correlation analysis of the studied varieties of common wheat, a very high correlation (r = 0.947) was found between the CP and the PDI in the Enola variety and Illico variety (r = 0.993). High positive values of r (r = 0.809) are reported between NFE and FUG. We have found also a high correlation (r = 0.99, $P<0.01$) between CP and PDI and between NFE and FUG (r = 0.99, $P<0.01$) in our other research.

We have a negative correlation between the CP and FUG (r = -0.790). Negative correlation is established between CF and FUM/FUG (r = -0.420, and r = -0.4670) (Table 4). Stoyanova et al. (2016) also establish negative correlation between CF and FUG (r = -0.20, $P<0.05$). Mathematically unproven are the correlation relationships between EE and the other indicators considered.

In Illico variety high positive values of r (r = 0.794, r = 0.821) was obtained between EE and FUM/FUG (Table 5). A negative correlation between NFE and PDI (r = -0.964) was also obtained.

Table 4. Correlation coefficients between wheat grain chemistry, energy and protein nutrition of ruminant wheat for Enola variety 2015-2016

	CP	EE	CF	NFE	FUM	FUG	PDI
CP	1	0.288	0.242	-0.835*	-0.433	-0.790*	0.947**
EE		1	-0.072	-0.654	-0.294	-0.138	0.016
CF			1	-0.291	-0.420	-0.467	0.271
NFE				1	0.728	0.809*	-0.622
FUM					1	0.806*	-0.226
FUG						1	-0.686
PDI							1

*Correlation significant at the 0.05 level
**Correlation significant at the 0.01 level

Table 5. Correlation coefficients between wheat grain chemistry, energy and protein feed of wheat for ruminants in Illico variety 2015-2016

	CP	EE	CF	NFE	FUM	FUG	PDI
CP	1	-0.386	-0.322	-0.986**	-0.322	-0.634	0.993**
EE		1	0.076	0.251	0.794*	0.821*	-0.449
CF			1	0.345	0.208	0.362	-0.229
NFE				1	0.197	0.571	-0.964**
FUM					1	0.636	-0.369
FUG						1	-0.642
PDI							1

*Correlation significant at the 0.05 level
**Correlation significant at the 0.01 level

A high positive correlation is recorded between the CP and DEpg (r = 0.767). The correlation between CP and MEpg (r = 0.662) is also very positive (Table 6).
The correlation between the content of CF and DEp/MEp negative as logical. The content of CF in the feed affect digestibility and energy nutrition.
We have also found negative correlation between CF and DEpg and MEpg (r = - 0.600 ÷ 0.741) and positive correlation dependence (r = 0.660) between CP and DEpg (r = 0.619) was found (Stoyanova et al. 2018) in our previous studies. Positive correlation between the contents of the CP and DE and a negative correlation between the content of CF and DE tempting 15 samples of wheat in experiments with pigs establish and Zijlstra et al. (1999).
The negative correlation between the CP and NFE found in both wheat varieties (r = -0.835, and r = -0.986). Analogous results between the crude protein and NFE (r = -0.83) and establish Alijosius et al. (2016).
High positive correlation relationships are reported between EE and DEp (r = 0.970) and EE and DEpg/MEpg (r = 0.775, r = 0.899) (Table 7).

Table 6. Correlation coefficients between wheat grain chemistry, energy and protein nutrition of non-ruminant wheat for Enola variety 2015-2016

	CP	EE	CF	NFE	DEp	MEp	DEpg	MEpg
CP	1	0.288	0.242	-0.835*	0.731	0.212	0.767*	0.662
EE		1	-0.072	-0.654	0.094	-0.077	0.200	0.158
CF			1	-0.291	-0.029	-0.239	0.033	-0.066
NFE				1	-0.332	0.209	-0.419	-0.287
DEp					1	0.812*	0.990**	0.988**
MEp						1	0.781*	0.867*
DEpg							1	0.987**
MEpg								1

*Correlation significant at the 0.05 level
**Correlation significant at the 0.01 level

Table 7. Correlation coefficients between wheat grain chemistry, energy and protein feed of wheat for non-ruminants in Illico variety 2015-2016

	CP	EE	CF	NFE	DEp	MEp	DEpg	MEpg
CP	1	-0.386	-0.322	-0.986**	0.366	-0.332	0.244	-0.032
EE		1	0.076	0.251	0.691	0.970**	0.775*	0.899**
CF			1	0.345	-0.068	0.105	0.025	0.161
NFE				1	-0.468	0.220	-0.356	-0.086
DEp					1	0.753	0.989**	0.911**
MEp						1	0.824*	0.936**
DEpg							1	0.958**
MEpg								1

*Correlation significant at the 0.05 level
**Correlation significant at the 0.01 level

CONCLUSIONS

The results of the present study show that the crude protein content ranges from 128.50-143.94 g/kg DM in the Enola variety and from 115.93 to 127.34 g/kg DM in the Illico variety. The introduction of Wuxal Grano slurry increased the crude protein content by 9.1 and 12.0 % relative to the control.

The applied leaf fertilizers do not affect the contents of FUM, FUG and PDI.

As a result of the correlation analysis, a very high correlation correlation (r = 0.947-0.993) was found between CP and PDI for both common wheat varieties.

REFERENCES

Alijosius, S., Svirmickas, G.J., S. Bliznikas, Gruzauskas, R., Sasyte, V., Raceviciute-Stupeliene, A., Kliseviciute, V., Dauksiene, A. (2016). Grain chemical composition of different varieties of winter cereals. *Zemdirbyste-Agriculture*, *103*(3), 273–280.

Anjum M.I., Ghazanfar S., Begum I. (2014). Nutritional composition of wheat grains and straw influenced by differences in varieties grown under uniform agronomic practices. *International Journal of Veterinary Science*, *3*(3), 100–104.

Dapoza, C., 2006. How to manage the variability of protein and amino acid contents in raw materials. *Amino NewsTM, Degussa 7*(1), 1–12.

Genchev, G., Marinkov, E., Yovcheva, V., Ognqnova, A. (1975). Biometric methods in plant growing, genetics and selection. Zemizdat, Sofia, Bg.

Guang-cail Z., Xu-hong, C., Yu-shuang, Y., Shan-shan, L., Ming, F., Yu-ping, D., Xiu-hong, W., Tie-heng, Z. (2009). Grain yield and protein components responding to the amount and rate of nitrogen application in winter wheat. *Journal of Triticeae Crops*, 09–02.

Kelley, K. & Sweeney, D. (2007). Placement of preplant liquid nitrogen and phosphorus fertilizer rate affects no-till wheat following different summer crops. *Agronomy Journal*, *99*, 1009–1017.

Kim, J.C., Mullan, B.P., Simmins, P.H. and Pluske, J.R. (2003). Variation in the chemical composition of wheats grown in Western Australia as influenced by variety, growing region, season and post-harvest storage. *Australian Journal of Agricultural Research*, *54*, 541–550.

Lasek, O., Barteczko, J., Augustyn, R., Smulikowska, S., Borowiec, F. (2011). Nutritional and energy value of wheat cultivars for broiler chickens. *Journal of Animal and Feed Sciences*, *20*, 246–258.

McCracken, K.J., Preston, C.M., Butler, C. (2002). Effects of wheat variety and specific weight on dietary apparent metabolizable energy concentration and performance of broiler chicks. *Br. Poult. Sci*., *43*, 253–260.

Mollah, Y., Bryden, W.L., Wallis, I.R., Balnave, D., Annison, E.F. (1983). Studies on low metabolizable energy wheats for poultry using conventional and rapid assay procedures and the effects of processing. *Br. Poult. Sci*., *24*, 81–89.

Mungova, M., & Vasileva, E. (2007). Amendment of cereal grain quality depending on nitrogen fertilization. *International Scientific Conference "The Plant Gene Fund - The Base of Modern Agriculture*", 13-14.06.2007, Bg.

Montemurro, F., Convertini, G., Ferri, D. (2007). Nitrogen application in winter wheat grown in Mediterranean conditions: effects on nitrogen uptake, utilization efficiency and soil nitrogen deficit. *Journal of Plant Nutrition, 30*(10), 1681–1703.

Panayotov, I., & Atanasova, D. (2004). New results in wheat breeding and their use in a global scale. Proceedings of the 17th EUCARPIA General Congress *"Genetic variation for plant breeding", 8-11 Sept., Tulln, Austria*, 181–184.

Sauvant, D., Perez, J.M., Tran, G. (2004). Tables of Composition and Nutritional Value of Feed Materials. Pigs, Poultry, Cattle, Sheep, Goats, Rabbits, *Horses and Fish.* 2nd revised Edition. INRA, Wageningen Academic Publishers.

Stoyanova, A., Ganchev, G., Kuneva, V. (2016). Nutrition value of two grain common wheat. *Scientific Papers. Series A. Agronomy*, *LIX*. 421–425. ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785.

Stoyanova, A., Kuneva, V., Ganchev, G., Georgiev, M. (2018). Evaluation of energy and protein nutrition of common valuation wheat varieties treated whit leaf fertilizers valuation. *2nd International Conference on Food and Agricultural Economics 27-28th April, Alanya, Turkey*, 298–304.

Tayebeh A., Alemzadeh, S., Kazemeini, A., Abdolreza, S. (2010). Effect of Organic and Inorganic Fertilizers on Grain Yield and Protein Banding Pattern of Wheat. *Australian Journal of Crop Science., 4*(6), 384–389.

Todorov, N., Ilchev, A., Georgieva, V., Girginov, D., Djouvinov, D., Penkov, D., Shindarska, Z. (2004). Animal nutrition. Textbook, UNISCORP, Sofia, 312 p.

Todorov, N., Krachunov, I., Djouvinov, D., Alexandrov, A. (2007). Handbook of Animal Feeding. Matkom Sofia, 400, Bg.

Uauy, C., Distelfeld, A., Fahima, T., Blechl, A., Dubcovsky, J. (2006). A NAC Gene Regulating Senescence Improves Grain Protein, Zinc, and Iron Content in Wheat. *Science, 314*(5803), 1298–1301, DOI: 10.1126/science.1133649.

Wiseman, J. (2000). Correlation between physical measurements and dietary energy values of wheat for poultry and pigs. *Animal Feed Science and Technology*, *84*, 1–11.

Yanchev, I. & Ivanov, U. (2012). Comparative study of physical, chemical and technological properties of the Greek and Bulgarian common wheat varieties. *FCS*, *8*(2), 219–226, Bg.

Zijlstra, R.T., , De Lange C.F.M., Patience, J.F. (1999). Nutritional value of wheat for growing pigs: chemical composition and digestible energy content. *Canadian Journal of Animal Science, 79*(2), 187–194.

Zilic, S., Dodig, D., Hadzi-Taskovic S., V., Maksimovic, M., Saratlic, G., Skrbic, B. (2010). Bread and durum wheat compared for antioxidants contents, and lipoxygenase and peroxidase activities. *International Journal of Food Science and Technology, 45*, 1360–1366.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOME ASPECTS OF BAKERY INDUSTRY QUALITY FOR ORGANIC AND CONVENTIONAL WHEAT

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Abstract

The objective of these researches was to analyze the quality for bakery industry of some organic and conventional wheat samples on the agricultural market in Romania. Analysis was performed on grains, flour and bread of the organic wheat by comparison with the conventional wheat. For wheat grains we tested: moisture, gluten, starch, proteins, Zeleny index, ash and fiber. For integral wheat flour we performed the following: organoleptic examination; wet gluten content; the index of gluten deformation; gluten index; Falling Number. For bread, we analysed: organoleptic examination; the ratio height/diameter; crumb porosity; crumb elasticity. The chemical composition of organic wheat grains was: 10.73% moisture; 13.25% proteins; 66.27% starch; 1.53% fibre; 1.87 ashes; 35 Zeleny Index. By comparison, for conventional wheat grains were: 11.52% moisture; 12.96% proteins; 67.12% starch; 2.31% fibre; 1.62 ash; 32 Zeleny Index. The results of integral wheat flour include: specific color of wheat milled grist, with smell pleasant, not specific smell of mold, hot or other special smell; the taste was normal, slightly sweet, not bitter or sour, with no mineral impurities; the wet gluten content varied between 30.74% for organic flour and 27.12% for conventional; deformation index was between 5 for organic flour and 3 for conventional flour; 56.43 gluten index was for organic flour and 50.07 for conventional; Falling Number was 247 seconds for organic flour and 234 seconds for conventional flour. Another aspect that was highlighted by determinations organoleptic bread quality assessment where it obtained 23 points for conventional bread and 30 points for organic bread. Thus, the bread obtained good fall into the category (18.1 ... 24 points) for conventional bread and very good, especially in terms of smell and taste for organic bread.

***Key words**: wheat, bakery industry, organic agriculture, yields quality, chemical composition.*

INTRODUCTION

Bread is a commonly consumed grain product in numerous countries and societies. Nowadays, there is a great variety of organic and “natural” breads, along with conventional ones. Can choose from white bread, wheat, whole or sprouted wheat, multi-grain, and gluten-free (Kucińska, 2012).
A clear understanding of the relationships between farming systems and crop nutritional quality is very important for designing agricultural management strategies which enhance environmental quality and sustainability while improving consumer’s health.
Agricultural production systems may differ greatly in terms of amount and sources of fertilisers, crop protection strategies and crop rotation. As such, a relationship between food quality and farming systems could be expected (Mazzoncini et al., 2007).
Introduction European policy to promote quality food products is an important component of the current Common Agricultural Policy (CAP). By this policy, European Union encourages farmers in order to obtain the agricultural products with superior quality. Currently, while the initial goal was achieved (in many agricultural products European Union provides the use of domestic over 100%), CAP was shifted to the qualitative aspect of food products, rather than to the quantity (Toader, 2008).
Grain quality at the mill is the result of the interaction of genotype with environmental conditions from sowing to delivery to the mill, and this interaction is potentially different for

each aspect of grain quality (Cauvain, 2003). The baking potential of wheat flours is influenced by many factors, most notably protein content (MacRitchie, 1987). Protein content is in turn influenced mainly by nitrogen fertilization, while the protein quality is determined primarily by the wheat genotype (Samaan et al., 2006). On the other hand, both the quality and the content of the wheat protein are affected by the climatic conditions during wheat maturation (Huebner, 1999). Vitreousness is considered to be related to the endosperm microstructure whereas hardness is suggested to influence the adhesion forces between starch granules and protein matrix (Greffeuille et al., 2006).

EU quality policy on the products obtained in the organic farming system is part of wider agricultural product quality policy. This policy meets the demand for specific products, increasingly stronger European consumers towards standardization in the development of conventional products. Producers are allowed if their products meet the conditions imposed by European regulation, to engage in a quality approach to enable better marketing. In order to qualify, depending on the geographical area will be enhanced, two indications: protected designation of origin and protected geographical indication. There is also the possibility that products produced using traditional methods to obtain certification of specificity (under endorsement traditional specialty guaranteed) (www.organic-europe.net).

This means that modern organic products, are not only fresh, delivered straight from the farm, but they also cover products every day involving sophisticated methods of processing products including wine, beer, pasta, yoghurt, prepared meals, cheeses etc. Like their counterparts from conventional agriculture, organic farmers and processors monitor trends in food consumption to ensure their products keep up with any changes in consumer tastes or demand, with the guarantee system certification on these products.

As a result of fewer irrigation practices and no synthetic fertilizers and pesticides being used in crops' growth, a higher amount of significant health-promoting nutrients, minerals, vitamins, and antioxidants can be found in organic grains. Eating organic bread can also lower your exposure to pesticides. Organic farming is significantly reflected not only in quality and safety of grain and grain-based products; it also has a great impact on our environment.

Furthermore, wheat is the most important cultivated plant, which is obtained mainly bread, staple food for about 40% of world population. The wheat flour is used for the preparation of various bakery products and manufacturing of pasta, etc. The grains of wheat are included in mixtures for breakfast cereals. Wheat grains are used in animal feed as such or ground, for the production of starch, gluten, alcohol, spirits (vodka, whiskey) beer, biofuel (ethanol). Straw have multiple uses, such as raw material in the pulp and paper industry; bedding; roughage; organic fertilizer into the soil after harvest incorporation or composting; producing energy by burning ends in fiery heat recovery.

In Romania there are favorable conditions for wheat cultivation and its role in food security strategy is determined by the possibilities of conservation inexpensively compared to other foods, cold chains unnecessary or costly installation (Roman et al., 2012).

MATERIALS AND METHODS

The main objective of this research was to determine the chemical composition and yield quality for organic and conventional wheat from the market of agricultural products in Romania. At the same time, there were tested samples of wheat flour for bakery industry quality. The material includes 20 of wheat samples, 10 for each type, organic and conventional. For results we present the average results.

Chemical analyses were performed to Yield Quality Laboratory of Crops Sciences Department of the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest, in 2017.

The devices used for analyses of wheat grains was spectrophotometer infrared NIR Inframatic 9200 Product Instalab-Analizer (Figure 1). For wheat grains we tested: moisture, gluten, starch, proteins, Zeleny index, ash and fibre.

For integral wheat flour performed according the STAS Methods (SR 877-95-Wheat Flour),

the following: organoleptic examination; wet gluten content; the index of gluten deformation; gluten index; Falling Number (Figure 2).
For bread, we analysed: organoleptic examination; the ratio height/diameter; crumb porosity; crumb elasticity (Table 1, Figure 3).

Figure 1. NIR Inframatic 9200 Product Instalab-Analizer (Yield Quality Laboratory of Crops Sciences Department)

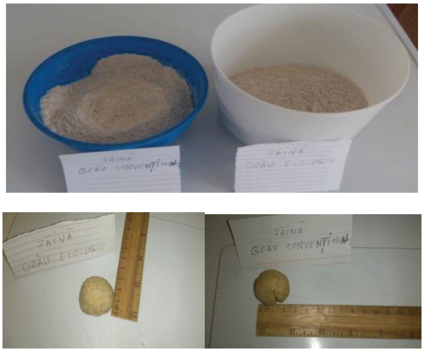

Figure 2. Aspects of gluten index deformation analysis (Yield Quality Laboratory of Crops Sciences Department)

Figure 3. Aspects of bread analysis (Yield Quality Laboratory of Crops Sciences Department)

Table 1. Descriptive vocabulary and definitions used by assessors to evaluate bread

Attribute	Definition
Appearance	
Crust colour	Degree of colour darkness in the crust ranging from pale to dark brown
Crumb colour	Degree of colour darkness in the crumb ranging from creamy to white
Crumb appearance	Degree of porosity and its uniformity from non uniform to uniform
Odour	
Yeasty	Odour associated with aromatic exchange from yeast fermentation
Grainy	An aromatic impression of cereal derived products like wheat, barley and corn
Texture	
Manual	Force required snapping sample by hand
Oral	Force required biting completely through sample placed between the molars
Flavour	
Sweet	Fundamental taste sensation of which sucrose is typical
Salt	Fundamental taste sensation elicited by sodium chloride
Sour	Fundamental taste sensation evoked by acids

RESULTS AND DISCUSSIONS

Results of wheat grains chemical compositon. Knowing, the moisture content plays an important role in storage of wheat grains. Moisture percentage is very important in the practice of obtaining and preserving agricultural products, as increased moisture favors grains firing and infestation. In Romania, the STAS moisture for wheat seeds is 14% (SR ISO 13548:2013).
According to the moisture content, a higher amount of dry matter was obtained from organic wheat, about 90% compared with conventional wheat which had a value of about 88% (Table 2).

Table 2. Comparative analysis on chemical composition of conventional and organic wheat seeds

Compounds (%)	Conventional wheat	Organic wheat
Moisture	11.52	10.73
Dry matter	88.38	89.27
Proteins	12.96	13.25
Starch	67.12	66.27
Fibre	2.31	1.53
Ash	1.62	1.87
Zeleny index (number)	32	35

Regarding the biochemical content of other components, one can highlight content slightly higher in proteins, over 13% for organic wheat, compared with conventional wheat samples.

Because minerals are distributed mainly to the periphery of the grain, the degree of extraction of the flour is higher, the higher ash content and flour is darker but richer in minerals, fibre and enzymes. Ash content for organic wheat flour had a value of 1.87%. For conventional wheat flour ash content was 1.62%. The starch was higher of conventional wheat, respectively 67.12%, by comparison with organic wheat.

Results on flour (whole meal). Organoleptic examination of the flour was: appearance (colour), smell and taste of flour for bakery.

Both types of flour (organic and conventional) had the characteristics of healthy product. After infestation determination, it was observed that after about an hour, the cone made with a funnel did not collapse, and there were no signs indicating the presence of mites or other insects on the surface for any of the analyzed samples.

The obtained results showed an ash content falling standards for integral flours in Romania for both types of sample. The wet gluten and proteins contents have the main role in the production of bread from wheat flour. Sufficiently flexible and extensible gluten ensures well-developed, smooth, uniform porosity with thin walls. In our experiments, wet gluten content varied between 30.74% for organic wheat and 27.12% for conventional wheat (Table 3). Thus, the obtained values frame the organic wheat and the conventional wheat in the Romanian standards (22-32%) for high quality flour for bakery. It highlights the wheat flour made from organic wheat, where gluten was over 30%.

The deformation of gluten index indicates the proteolytic activity of the flour. The deformation of the gluten is high if it exceeds 15 mm. If gluten deformation is below 5 mm, proteolytic activity is very low, gluten is highly elastic, and flour requires amelioration with proteolytic or reducing enzymes. For baking flours Standards, gluten deformation index ranges from 3 to 25 mm. In this experiment it was obtained 3 mm for conventional wheat flour and 5 mm of organic wheat flour.

Gluten index calculated for the two types of flour ranged between 50.07 in the case of conventional wheat flour and 56.43 respectively for organic wheat flour. The standards gluten index ranges oscillated between 40 and 50.

The nutritional content of breads is related to the chemical composition of the bread, hence protein content and starch composition are of importance when considering the dietary impact of breads.

Table 3. Comparative analysis on flour of conventional and organic wheat

Compounds	Conventional wheat	Organic wheat	Standards Values
Organoleptic examination	Product specific healthy	Product specific healthy	Product specific healthy
Wet gluten (%)	27.12	30.74	22
Deformation index (mm)	3	5	3-15
Gluten index	50.07	56.43	40-50

Falling Number test was determined in laboratory by Falling Number device, according to standard ISO 3093-2005. In these circumstances it was 228 seconds organic flour and conventional flour was 247 seconds (Table 4). This index measuring amylase activity involved in starch degradation, which can be excessive in the presence of germinating seeds or emerging germination. The method is widely used commercially to measure the germination of grain. The results show the two types of flour are suitable to use in bakery industry.

Table 4. Falling Number of conventional and organic wheat flour

Values			Quality of flour
Conventional wheat flour	Organic wheat flour	Standards Values	
-	-	< 120 seconds	Unusable
-	-	120- 180 seconds	Satisfactorily
228	247	180- 260 seconds	Suitable
-	-	>260 seconds	Usable with the addition of enhancers

Results on bread. Organoleptic characteristics after 4 hours of rest from cooking were following:

- Type of product: bread analyzed has regular shape at both flour types, and volume was sufficiently developed and flattened to any of the samples.

• Bark:
- Appearance: shell surface was matt (has cracks on upper and side shell surface over 1 cm long, the skin is crispy, slightly soft to hear a sound hitting its open, clean).
- Color: nice browned crust was yellowish-gold coloration is uniform and attractive.

• Core:
- Sectional appearance: the product was sufficiently ripe, fairly uniform crumb porosity, cracks vertical, lateral or horizontal core.
- Colour: crumb feature was the assortment of bread.
- Consistency: pressing with the finger slowly enough bread crumb returns to its original state and cutting the slice of bread, knife remains clean, it is not crumbles core was wet and tacky to the touch, and elastic.
- Smell: the product was especially pronounced flavor of organic wheat flour.
- Taste: product has a satisfactory taste.
- The total number of points by scoring method (Table 5) was 23 points for the conventional bread by comparison with organic bread (30).

Quality bread falling in the good category (18.1-24 points) for conventional bread and very good for organic bread, especially in terms of smell and taste.

Table 5. Organoleptic quality assessment by method points of bread

Organoleptic characteristics	Maximum points for conventional bread	Maximum points for organic bread
Product form	4	5
Crust:		
aspect	3	3
colour	3	3
Crumb:		
sectional appearance	3	3
colour	3	3
consistency	3	3
Smell	2	5
Taste	3	5
Total score	23 points	30 points

Besides other issues as determined by organoleptic examination, little can comment on defects in, namely:
- Cracks in the shell, even if they were not large, were determined through gas outlet fermentation of the dough, giving it specifies that poor aesthetics and a low volume, so once the output of these gases fermentation were lost and some the flavour substances of bread.
- The core was wet, probably due to elastic baking at elevated temperature, a short time.

The evolution of the core and shell after 24 hours storage. Bread immediately after cooling began to aging, the first signs appeared after 10-12 hours. Bark has become matt, it was first soaked, then it became hard core was brittle. Flavor and aroma of fresh bread disappeared, replaced by a bland taste, especially stale bread from conventional wheat. The core brittle occurred due to insufficient gelatinisation of starch. Taste of the core and shell fad after 24 hours of storage arose due to insufficient fermentation and leaven dough.

To remember is that not all defects importance for the quality of bread. Very important are: ripeness, volume, porosity, taste and smell. They can decide the acceptability of bread as the final product.

CONCLUSIONS

The concept of organic farming is aimed to produce food with the taste, texture and authentic qualities and attractive corresponding to culinary preferences and skills of the modern consumer.

The production of certified organic wheat allowed to maintain a premium price compared with the conventional market.

Consumers are increasingly aware of social issues and the environment related to food production as evidenced by the significant increase in sales of food certificates, both in stores and in natural food supermarket chains.

All these values are exponent climatic conditions and culture technology applied for the two types of wheat analyzed and reveal their importance on the chemical composition or the quality of the harvest.

ACKNOWLEDGEMENTS

This article was financed by the Faculty of Agriculture, University of Agronomic Sciences and Veterinary Medicine of Bucharest.

REFERENCES

Cauvain, S.P. (2003). *Bread making. Improving quality*. Woodhead Publishing Limited. Cambridge. England.

Greffeuille, V., Abecassis, J., Rousset, M., Oury, F., Faye, A., Lullien-Pellerin, A. (2006). Grain characterization and milling behavior of near-isogenic lines differing by hardness. *Theor. Appl. Genet*., *114*, 1–12.

Huebner, F.R., Bietz, J.A., Nelson, T., Bains, S.G., Finney, P.L. (1999). Soft wheat quality as related to protein composition. *Cereal Chem*., *76*, 650–655.

Kucińska, M. (2012). Organic bread versus natural and convetional bread. Archive at https://planet save. com/2012/09/20/organic-read-versus-natural-and-conventional-bread/.

MacRitchie, F. (1987). Evaluations of contributions from wheat protein fractions to dough mixing and bread making. *J. Cereal Sci*., *6*, 259–268.

Mazzoncini, M., Belloni, P., Risaliti, R., Antichi D. (2012). Organic Vs Conventional Winter Wheat Quality and Organoleptic Bread Test. Archived at http://orgprints.org/view/projects/int_conf_qlif2007.html

Roman, Gh.V., Duda, M., Imbrea, F., Matei, Gh., Timar, A.V. (2012). *Conditioning and preserving agricultural products.* Bucharest, RO: University Publishing House.

Samaan, J., El-Khayat, G.H., Manthey, F., Fuller, M., Brennan, C.S. (2006). Durum wheat quality: II The relationship of kernel physicochemical composition to semolina quality and end product utilisation. *Int. J. Food Sci. Technol*., *41*, 47–55.

Toader, M. (2008). Research on chemical composition and quality of the harvest of grain and pseudocereales species under the influence of natural and technological factors. PhD Thesis, USAMV Bucharest.

http://ISO Standards of wheat: http://old.madr.ro/pages/proiecte_interne2012/ordin-manual-gradare-seminte-consum.pdf.

http://www.organic-europe.net.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF SOWING PERIOD AND TREATMENT WITH VARIOUS FOLIAR FERTILIZERS ON THE PRODUCTIVITY OF RAPESEED

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Abstract

The experiment is conducted in the Experimental Base of the Department of Plant Growing at Agricultural University of Plovdiv in the period 2012-2015. The experiment is set in block mode, in 4 repetitions, with cultivated plot size of 20 m^2, with winter rapeseed – the Visby hybrid. The aim of the experiment is to establish rapeseed yield treated with the following leaf fertilizers: Lactofol B - 4000 ml/ha, Litovit® - 2000 g/ha, and Fertiactyl Starter - 3000 ml/ha, and one untreated control. Rapeseed is sown during the following sowing dates - 1-10.IX, 1-10.X, 10-20.X, 20-30.X. The leaf fertilization is carried out in the 2-4 leaf stages for the first two sowing periods and in 1-2 leaf stages for the second two sowing periods. The studied leaf fertilizers have positively influenced the yield of rapeseed. The highest grain yield was recorded in 2014, and the lowest in 2013. Both during the three-years period and averagely for the study period, the highest grain yield for the different sowing periods was obtained from variants treated with Fertiactyl Starter - 3000 ml/ha.

Key words: *rapeseed, sowing period, leaf fertilization, yield.*

INTRODUCTION

The problem of shortage of moisture during sowing – germination period of rapeseed in Bulgaria in recent years has been steadily increasing. The small amount of precipitation in this period leads to the later germination of the crop and its inability to enter the optimum phase necessary for wintering. One way to stimulate plant growth and overcome unfavourable weather conditions is to use leaf fertilizers. The direct treatment of the leaves leads to a faster and more efficient supply of the plants with the necessary nutrients and minerals.

Experiments with leaf fertilizers have been carried out in many countries around the world and in our country on various agricultural crops (Sekar S. et al., 2010; Soare M. et al., 2010; Stoyanova A. et al., 2010; Toader C., 2010; Sanmueang A. et al., 2011), however those concerning rapeseed are scarce.

That is why we set ourselves the objective to find out the effect of some foliar fertilizers on the productivity of the oil producing rapeseed.

MATERIALS AND METHODS

The experiment was carried out within the period of the years 2012-2015, on the land of the Training, experimental and implementation base of the Department of Plant growing of the Agricultural University of Plovdiv. The experiment was carried out according to the block method, with 4 reiterations, with the experimental plot having the size of 20 m^2, using Visby hybrid that originates from Germany.

Experimental variants:

Factor A - Sowing dates

Factor B - Foliar fertilizers

Factor C - Development phases

I. Date of sowing 1-10.IX

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4th leaf
- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4th leaf
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4th leaf

II. Date of sowing 1-10.X

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4th leaf
- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4th leaf
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4th leaf.

III. Date of sowing 10 – 20.X

- Untreated variant

- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4th leaf
- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4th leaf
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4th leaf.

IV. Date of sowing 20-30. X

- Untreated variant
- Spraying with Lactofol B - 4000 ml/ha - phenophase - 2-4th leaf
- Spraying with Litovit® - 2000 g/ha - phenophase - 2-4th leaf
- Spraying with Fertiactyl Starter - 3000 ml/ha - 2-4th leaf.

Rapeseed is planted after wheat being the precursor. After its harvesting, the land was ploughed at a depth of 18-20 cm, and then cultivated twice by cross-disking and rolling before sowing. With the main soil cultivation 100 kg/ha phosphorus and 80 kg/ha potassium were inserted. From a total of 170 kg/ha nitrogen - 30 kg/ha have been imported in the autumn in the course of pre-sowing treatment, and the rest of the quantity - as early as early spring.

Sowing was carried out by means of a seed drill, at a 12-15 cm distance between the rows, with a sowing rate of 6 kg/ha, which delivers a density of 60 plants/m^2. The depth of sowing is 2-3 cm. After sowing, the land is rolled.

In the course of the experiment the productive capacity of rapeseed is observed and studied, the one resulting from different sowing dates and treated with different foliar fertilizers.

The main meteorological factors that have influenced crop productivity over the years are air temperature and quantity of rainfall during the periods that are of critical importance for the development of rapeseed. (Figures 1, 2, 3).

The study of these factors shows specifics with regard to the average monthly temperatures and especially with respect to moisture (quantity and distribution of rainfall during vegetation), which is one of the risk factors for obtaining high yields of rapeseed.

Significant deviations from the values of the average day and night temperature during the three years of the experiment carried out on the lands of the Training, experimental and implementation base versus the multi-year period were not observed.

More unevenly distributed during vegetation, however the highest quantity of rain in the course of the three experimental years was reported in 2014-2015 (856.4 mm).

Less, but better distributed are rainfalls reported in 2013-2014 (574.7 mm), while in the period 2012-2013 (541.0 mm) the reported rainfall is the least.

The coldest months during the winter period are December and January, with minimum temperatures being recorded in January 2014-2015 (-14.2°C), December 2012-2013 and 2013-2014 (-9.8°C; -10.4°C) (Figure 2).

RESULTS AND DISCUSSIONS

The indicators related to seed yield are given in Tables 1, 2 and 3. The data shows that, depending on the sowing dates, weather conditions, and foliar fertilization, these indicators change during the years of the experiment.

All structural elements with the exception of fruit length and pods weight of a plant have the highest values in 2012-2013, and the lowest in 2014-2015.

During the three years of sowing, all structural elements, averagely in the course of the experimental period, have higher values in the variants sown in the period 20-30.X and 1-10. IX, followed by those sown from 1 to 10.X and from 10 to 20.X.

Plant height from all sowing dates, and averagely for the studied period had the highest values for the plants treated with Fertiactyl Starter (from 159.9 to 166.5 cm), followed by Lactofol B treated (from 159.1 to 164.4 cm), Litovit® (from 158.7 to 163.7 cm), while and the lowest height is registered for the control plants (from 158.0 to 163.5 cm).

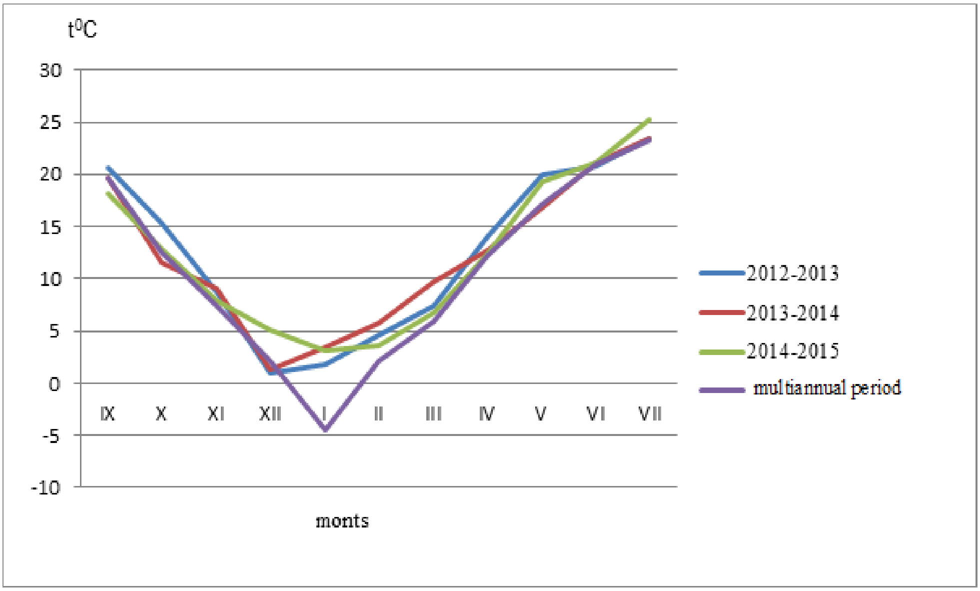

Figure 1. Average monthly temperatures in the region of the Training, experimental and implementation base

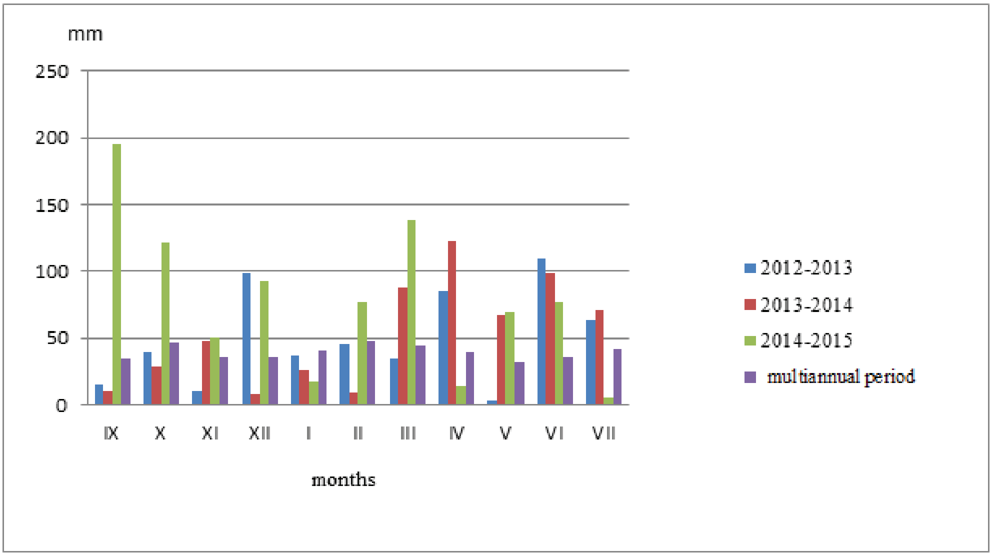

Figure 2. Quantity of rainfall during the years of survey in the region of the Training, experimental and implementation base

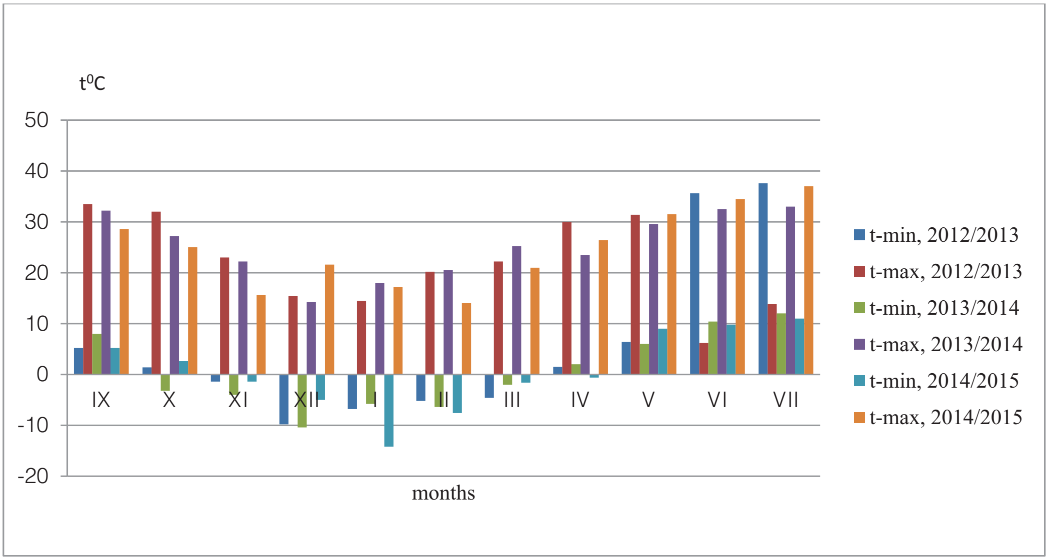

Figure 3. Absolute minimum and maximum temperatures by months 2012/2015

Averagely for the experimental period, Fertiactyl treated variants again have the highest values with regards to the number of branches (from 7.9 to 9.5 pieces), followed by the variants treated with Lactofol B (from 7.9 to 9.3 pieces), Litovit® (from 7.8 to 9.2 pieces), and finally the control group of plants (from 7.8 to 9.2 pieces).

In terms of the number of fruits of a plant, higher values during the three years and averagely for the study period were reported for the leaf fertilizer treated variants versus the control group.

Of the foliar fertilized variants, averagely for the experimental period, the number of fruits was greatest (from 203.7 to 218.0) in Fertiactyl Starter treated plants, followed by those treated with Lactofol B (202.2 to 215.6) and Litovit® (from 200.7 to 213.7 pieces). The smallest number of fruits was reported in the control variants (from 199.8 to 212.4 pieces).

The length of the fruits from the different sowing dates and between the control
and leaf treated variants varies within a small range.

With regards to the number of seeds in a single fruit, the highest number of seeds averagely for the period was reported for Fertiactyl Starter treated variants (27.0 to 28.2 pieces).

The number of seeds averagely for the experimental period for the remaining variants ranged from 26.8 to 27.7 for Lactofol B, 26.6 to 27.5 for Litovit® and from 26.6 to 27.3 for the control plants.

The main structural elements that have the greatest impact on seed yield are the weight of the fruits and seeds of a single plant.

Highest values of these structural elements in leaf-fertilized variants were reported for Fertiactyl Starter, while the lowest - for Litovit®.

On average during the experimental period, the values of these indicators in leaf-fertilized variants ranged from 25.1 to 26.2 g for Fertiactyl Starter treated plants, followed by Lactofol B treated plants (from 24.8 to 25.7 g), Litovit® (from 24.7 to 25.6 g), and the lowest in the control plants (24.6 to 25.5 g) in terms of fruit weight, and within 14.1 to 15.0 g for Fertiactyl Starter treated plants, followed by Lactofol B treated variants (13.9 to 14.8 g), Litovit® (from 13.8 to 14.7 g), and the lowest for the control plants (from 13.7 to 14.6 g) in terms of the weight of the seeds of a plant.

Both at all sowing dates, and for all variants, the values of these two indices are the lowest in the control.

Table 1. Plant height and structural elements of the seed yield

	Plant height (cm)				Number of branches in a single plant (pcs)				Number of fruits in a single plant (pcs)			
1-10.IX												
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	165.4	163.1	162,0	163.5	9.6	9.0	8,9	9.2	218.3	211.6	207,3	212.4
Fertiactyl Starter	167.3	165.1	163,5	165.3	9.8	9.3	9,1	9.4	225.7	217.2	212,0	218.3
Litovit®	165.7	163.3	162,1	163.7	9.6	9.1	9,0	9.2	219.5	213.0	208.5	213.7
Lactofol B	166.3	164.2	162,8	164.4	9.7	9.2	9,1	9.3	221.4	215.2	210.2	215.6
1-10.X												
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	162.2	161.1	160.3	161.2	8.4	8.3	8.2	8.3	210.7	205.9	203.0	206.5
Fertiactyl Starter	164.4	163.2	161.7	163.1	8.7	8.6	8.4	8.6	216.8	211.9	207.3	212.0
Litovit®	162.8	161.5	160.6	161.6	8.5	8.4	8.3	8.4	212.4	207.4	204.1	208.0
Lactofol B	163.7	162.3	161.1	162.4	8.6	8.5	8.3	8.5	214.3	209.3	206.5	210.0
10-20.X												
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	160.1	157.2	156.8	158.0	8.0	7.8	7.7	7.8	203.7	198.7	197.1	199.8
Fertiactyl Starter	162.2	159.4	158.1	159.9	8.1	7.9	7.8	7.9	207.5	203.2	200.5	203.7
Litovit®	160.9	157.9	157.2	158.7	8.0	7.8	7.7	7.8	204.7	199.6	197.7	200.7
Lactofol B	161.9	158.3	157.2	159.1	8.0	7.8	7.8	7.9	206.5	201.1	199.0	202.2
20-30.X												
	2012/ 2013	2013/ 2014	2014/ 2015	Average	012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	-	163.3	162.2	-	-	9.1	9.0	-	-	213.5	209.9	-
Fertiactyl Starter	168.5	166.2	164.7	166.5	9.7	9.5	9.4	9.5	221.3	218.4	214.3	218.0
Litovit®	-	164.1	163.0	-	-	9.3	9.3	-	-	215.9	212.1	-
Lactofol B	-	165.5	164.2	-	-	9.4	9.3	-	-	217.1	213.2	-

Table 2. Structural elements of the seed yield

	Fruit length (cm)				Number of seeds in one fruit (pcs)				Weight of the fruit in a single plant (g)			
1-10.IX												
	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average
Control	7.7	7.6	7.6	7.6	27.6	27.2	27.0	27.3	26.0	25.6	24.8	25.5
Fertiactyl Starter	7.8	7.8	7.7	7.8	28.3	28.0	27.8	28.0	26.5	26.1	25.2	25.9
Litovit®	7.7	7.7	7.8	7.7	27.8	27.4	27.3	27.5	26.1	25.7	24.9	25.6
Lactofol B	7.8	7.7	7.7	7.7	28.1	27.6	27.4	27.7	26.3	25.8	25.0	25.7
1-10.X												
	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average
Control	7.6	7.5	7.5	7.5	27.0	26.9	26.8	26.9	25.0	24.9	24.7	24.9
Fertiactyl Starter	7.7	7.6	7.6	7.6	27.8	27.5	27.3	27.5	26.0	25.7	25.4	25.7
Litovit®	7.6	7.6	7.6	7.6	27.1	27.0	26.9	27.0	25.2	25.1	24.9	25.1
Lactofol B	7.6	7.6	7.5	7.6	27.3	27.1	27.0	27,1	25.7	25.4	25.2	25.4
10-20.X												
	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average
Control	7.6	7.5	7.5	7.5	26.8	26.5	26.4	26.6	24.8	24.5	24.4	24.6
Fertiactyl Starter	7.7	7.7	7.6	7.7	27.3	27.0	26.7	27.0	25.3	25.1	24.9	25.1
Litovit®	7.6	7.6	7.5	7.6	26.9	26.6	26.4	26.6	24.9	24.6	24.5	24.7
Lactofol B	7.7	7.6	7.5	7.6	27.0	26.8	26.6	26.8	25.1	24.8	24.6	24.8
20-30.X												
	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average	2012/2013	2013/2014	2014/2015	Average
Control	-	7.6	7.5	-	-	27.4	27.2	-	-	25.7	25.1	-
Fertiactyl Starter	7.8	7.7	7.6	7.7	28.4	28.2	27.9	28.2	26.6	26.4	25.5	26.2
Litovit®	-	7.6	7.6	-	-	27.6	27.5	-	-	25.8	25.2	-
Lactofol B	-	7.7	7.6	-	-	27.8	27.6	-	-	26.0	25.3	-

Table 3. Structural elements of the seed yield

	Weight of the seeds in a single plant (g)				Weight of pods in a single plant (g)			
1-10.IX								
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	15.0	14.6	14.1	14.6	11.0	11.0	10.7	10.9
Fertiactyl Starter	15.3	15.0	14.4	14.9	11.2	11.1	10.8	11.0
Litovit®	15.1	14.7	14.2	14.7	11.0	11.0	10.7	10.9
Lactofol B	15.2	14.8	14.3	14.8	11.1	11.0	10.7	10.9
1-10.X								
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	14.0	14.0	13.9	14.0	11.0	10.9	10.8	10.9
Fertiactyl Starter	14.9	14.6	14.4	14.6	11.1	11.1	11.0	11.1
Litovit®	14.2	14.1	14.0	14.1	11.0	11.0	10.9	11.0
Lactofol B	14.7	14.4	14.2	14.4	11.0	11.0	11.0	11.0
10-20.X								
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	13.9	13.7	13.6	13.7	10.9	10.8	10.8	10.8
Fertiactyl Starter	14.2	14.1	14.0	14.1	11.1	11.0	10.9	11.0
Litovit®	14.0	13.8	13.7	13.8	10.9	10.8	10.8	10.8
Lactofol B	14.1	13.9	13.8	13.9	11.0	10.9	10.8	10.9
20-30.X								
	2012/ 2013	2013/ 2014	2014/ 2015	Average	2012/ 2013	2013/ 2014	2014/ 2015	Average
Control	-	14.6	14.2	-	-	11.1	10.9	-
Fertiactyl Starter	15.3	15.1	14.5	15.0	11.3	11.3	11.0	11.2
Litovit®	-	14.7	14.3	-	-	11.1	10.9	-
Lactofol B	-	14.9	14.4	-	-	11.1	10.9	-

The weight of the pods for the different variants and dates of sowing do not differ significantly.

Averagely for the experimental period, the highest values for pod weight were reported in the Fertiactyl Starter treated variant (from 11.0 to 11.2 g).

All structural elements at different sowing dates and experimental variants have the highest values in the case of treatment with Fertiactyl Starter foliar fertilizer, while the lowest - for the control variant.

Seed yield

Data on the seed yield from the experimental variants and sowing dates are presented in Table 4.

The results obtained show that, depending on the year, the meteorological conditions, the sowing date and the treatment with foliar fertilizers, the seed yield varies.

The more favourable combination and distribution of meteorological factors in 2013-2014 create a prerequisite for obtaining higher yields during the years and cultivation variants followed by the yields of 2014-2015. The lowest yield was obtained in 2012-2013.

In the second or third year of the experiment, the highest seed yields were obtained during the first sowing date, whereas in the following dates yields gradually decreased.

The optimal combination of moisture and temperature at the second and third sowing dates in 2012-2013 (from 1 to 10.X and 10 to 20.X) are a prerequisite for obtaining higher yields in all cultivation variants compared to the first sowing date.

Lower temperatures during the sowing period from 20 to 30.X create a prerequisite for the inability of the plants to reach the phase that is optimal for wintering during the three experimental years.

Therefore the yields of seeds are the lowest at this sowing date, and in 2013, yields are only reported for the variants treated with Fertiactyl Starter.

Averagely for the experimental period, the highest seed yield was obtained by Fertiactyl Starter treated variants at all sowing dates (1 to 10.IX - 3.774 t/ha; 1 to 10.X - 3.946 t/ha; 10 to 20.X - 3.703 t/ha; 20 to 30.X - 0.981 t/ha), followed by Lactofol B (from 1 to 10.IX - 3.713 t/ha; from 1 to 10.X - 3.823 t/ha , from 10 to 20.X - 3.604 t/ha), Litovit® (from 1 to 10.IX - 3.617 t/ha; from 1 to 10.X -3.706 t/ha; from 10 to 20.X - 3.496 t/ha), and the control (from 1 to 10.IX - 3.527 t/ha; from 1 to 10.X - 3.598 t/ha; from 10 to 20.X - 3.335 t/ha).

Foliar fertilizer treatment compared to the control plants is of greatest importance for sowing period from 10 to 20.X, followed by sowing period 1-10.X and 1-10.IX.

Fertiactyl Starter-treated variants have the highest percentage increase in yield compared to the control plants (from 7.0 to 11.0%), followed by Lactofol B treated plants (from 5.3 to 8.1%) and Litovit® treated plants (from 2.4 to 4.8%).

On the basis of the mathematical processing, during the three experimental years, at all sowing dates, statistical proof in terms of seed yield data, at LSD 5%, was established between the leaf-fertilized variants and the control.

CONCLUSIONS

Foliar fertilization has a beneficial effect on structural elements and seed yield for all variants and dates of sowing.

Compared to the control plants, the structural elements producing the seed yield have higher values for the leaf-fertilizer treated variants.

The highest values of all structural elements forming the seed yield were reported in the variant treated with Fertiactyl Starter.

For leaf-treated variants, seed yield is from 2.4 to 11.0% higher than the control.

Averagely during the experimental years, the highest seed yield was obtained from Fertiactyl Starter treated variant (0.981-3.946 t/ha), which exceeds the control with 7.0 to 11%.

Table 4. Seed yield t/ha

Variants / Sowing dates	Seed yield t/ha				
	1-10.IX				
	2012/2013	2013/2014	2014/2015	Average	%
Control	3.058[a]	3.908[a]	3.614[a]	3.527	100.0
Fertiactyl Starter	3.236[c]	4.148[c]	3.939[d]	3.774	107.0
Litovit®	3.132[b]	4.007[b]	3.712[b]	3.617	102.4
Lactofol B	3.202[bc]	4.102[bc]	3.836[c]	3.713	105.3
LSD 5%	0.0701	0.0988	0.0890	-	-
	1-10.X				
	2012/2013	2013/2014	2014/2015	Average	%
Control	3.553[a]	3.662[a]	3.579[a]	3.598	100.0
Fertiactyl Starter	3.903[d]	4.023[d]	3.911[d]	3.946	109.7
Litovit®	3.659[b]	3.784[b]	3.674[b]	3.706	103.0
Lactofol B	3.784[c]	3.902[c]	3.799[c]	3.828	106.4
LSD 5%	0.079	0.080	0.079	-	-
	10-20.X				
	2012/2013	2013/2014	2014/2015	Average	%
Control	3.285[a]	3.383[a]	3.337[a]	3.335	100.0
Fertiactyl Starter	3.647[d]	3.752[d]	3.709[d]	3.703	111.0
Litovit®	3.437[b]	3.546[b]	3.505[b]	3.496	104.8
Lactofol B	3.544[c]	3.655[c]	3.613[c]	3.604	108.1
LSD 5%	0.073	0.075	0.074	-	-
	20-30.X				
	2012/2013	2013/2014	2014/2015	Average	%
Control	-	1.000[a]	0.754[a]	-	-
Fertiactyl Starter	0.889	1.154[d]	0.901[d]	0.981	-
Litovit®	-	1.047[b]	0.817[b]	-	-
Lactofol B	-	1.101[c]	0.863[c]	-	-
LSD 5%	-	0.030	0.028	-	

REFERENCES

Sanmueang, A., Detpiratmongkol, S., Yoosukyingsataporn, S., Ubolkerd, T. (2011). Influence of foliar application of potassium fertilizers on growth and yield of sweet sorghum. *Proceedings of the 49th Kasetsart University Annual Conference, Kasetsart University, Thailand,* 1-4 February. *Subject: Plants*, *1*, 458–464.

Sekar, S., Amanullah, M., Manoharan, S., Muthukrishnan, P., Subramanian, K., Vincent, S. (2010). Influence of fertilizer levels and growth substances on yield and economics of hybrid maize. *Madras Agricultural Journal, 97*(1/3), 58–61.

Soare, M., Sîrbu, C., Cioroianu, T., Oprică, I., Mihalache, D., Grigore, A., Anton, I., Marin, N. (2010). The influence of nitrogen chemical sources from complex foliar fertilisers on the penetration, uptake and the distribution of the potassium in the unteached leaves of sunflower plants from the solutions applied on leaves. *Annals of the University of Craiova, Agriculture, Montanology, Cadastre Series, 40*(1), 587–591.

Stoyanova A., Gospodinov, I., Petkova, R. (2010). Economic evaluation of winter wheat leaf fertilization. *Agricultural Science and Technology, 2*(3), 136–138.

Toader, C., Rusu, M., Mărghitaş, M. (2010). Foliar fertilization effect of supplementing the fertilization of the soil on the production of potato tubers. *Bulletin of University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca. Agriculture, 67*(1), 279–285.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF LEAF FERTILIZERS ON THE CHEMICAL COMPOSITION OF THE GRAIN OF SOLID DURUM

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Abstract

During the 2014-2016 period, in the experimental field of the Department of Plant Growth at Agricultural University of Plovdiv were carried out field experiments. The aim of the study was to investigate the influence of foliar fertilizers on the chemical composition of the grain of durum wheat varieties Predel and Elbrus. The plants were treated in the tillering, jointing and heading phases of growth with Megafol (300 ml/da) and Megafol-protein (300 ml/da). The results were compared with an untreated control. The experiments were based on a predecessor chickpea on the block method in four replications with a plot size of 15 m^2.
As a result of the experiments, the use of Megafol-protein had the most significant effect on the crude protein content of the Predel grain when it was applied in the tillering phase and the Elbrus grain when applied in the heading phase. The use of Megafol had a significant positive effect on the gluten content of the Elbrus variety only after application in the jointing phase.

Key words*: gain chemical content, durum wheat, leaf fertilizers.*

INTRODUCTION

The factors of the environment have an enormous influence on the processes of the plants, the formation of the yield and its quality. The protein content of wheat grains also varies strongly (from 9 to 24%) depending on the growing conditions. The role of nutrition and the importance of individual elements in plant life are primarily determined by the fact that the nutrients entering the plant cells are compounded by important organic compounds. Nitrogen in plants is quickly converted to amino acids, which are the starting compounds for the synthesis of proteins, nucleic acids, alkaloids and others.
The use of foliar fertilizers during vegetation can greatly influence the formation of yields and the quality of the grain obtained. There is much work to do in this direction. For example, the application of leaf fertilizers during jointing phase has been found to increase grain yield for durum wheat: 9.1% for Lactofol O and 9% for Devifert M + M due to their positive influence on the structural elements of the yield and the mass per 1000 grains (Delchev, 2000). The Ramil Stimulator, applied alone or in combination with complex foliar fertilizers, increases the protein content, wet and dry gluten, as compared to Tritimil and its combinations (Delchev, 2004). Applied through vegetation, leaf microbes provide a better diet for wheat. The combined spring feed with Vouxal Microplant (100 ml / da) + Kodice (250 ml/da) contributes to obtaining the highest number of grains in class and a larger grain mass - 63% higher than untreated crops. The application of leaf fertilizers during the period of intense growth increases the protein content, the raw fats, the carbohydrate compounds (starch) and the cellulose. The amount of crude protein is highest (17.4%) when applying the mineral fertilizers Vouxal and Kodice - 14.4% higher protein content than in the wheat, once fertilized with ammonium nitrate at 30 kg/da (Stoyanova, 2010). The combination of the Microelements for cereals with Amalgerol Premium leads to an increase in yield (by 59.4 kg/da) and increases the resistance of durum wheat to unfavorable climatic conditions (Delchev, Stoyanova, 2013).
The purpose of this study was to investigate the effects of two foliar fertilizers (Megafol and Megafol-protein) and the time of their application on grain quality and yields of two varieties of durum wheat in the conditions of the Plovdiv region.

MATERIALS AND METHODS

The experiments were carried out in the experimental field of the Department of Plant Growth at the Agricultural University of Plovdiv in the period 2014-2016. The effects of the Megafol (300 ml/da) and Megafol-protein (300 ml/da) were studied. The influence of the fertilizers was compared to control plants that were not treated. The experiments were based on a predecessor chickpea on the block method in four replicates with a plot size of 15 m^2. The sowing of durum wheat is carried out in the optimal time from 20th of October to 10th of November with a sowing rate of 500 seeds/m^2 and mineral fertilization with 12 kg/da nitrogen and 8 kg/da phosphorus. All phosphorus fertilizer was imported prior to sowing together with 1/3 of the nitrogen fertilizer. The rest of nitrogen fertilizer was imported as spring feed. All units of the established technology for growing durum wheat were followed.

The foliar treatment preparations used contain valuable plant growth stimulating ingredients. Megafol contains amino acids and a complex of biologically active substances (sugar beet extract, algae extract, and organic ingredients required for growth). Megafol-protein also contains a complex of biologically active substances that activate the genes involved in protein synthesis (GEA 166).

The following grain parameters were determined: wet and dry gluten yield (by weight), fiber, crude fat (by residual method), nitrogen content and crude protein (Kjeldahl), carbohydrate substances (by the residual method).

RESULTS AND DISCUSSIONS

Table 1 shows the occurrence of phenophases in the tested varieties.

The duration of the period from sowing to germination is determined by the amount of precipitation before sowing, but is mainly limited by the air temperature, with the higher average air temperature guaranteeing the faster and joint growth of wheat. Prior to sowing of durum wheat on 10th November 2014, soil moisture reserves increased, which combined with the higher temperature compared to the multiannual period of time created good conditions for the germination of the plants. Higher temperatures in mid-November and falling precipitations contributed to the normal germination phase at the end of the month. The germination of plants is followed by abundant rainfall in the first ten days of December - 32.3 mm more than the multiannual period of time. In the early stages of development no significant differences were observed between the two tested varieties (Table 1). The Elbrus variety enters one to two days earlier, respectively, in phase of germination and third leaf compared to Predel variety. Predel variety entered the phase of 3rd leaf on 17.12.2014, and Elbrus variety on 15.12.2014. Gradually, with decreases in temperatures in December, the growth processes of durum wheat also subsided. In January, rainfall was 23.6 mm less, and in February and March it was 44.6 and 100.0 mm higher than the norm for the multiannual period. These two months were also characterized by higher average monthly temperatures of 3.10°C and 3.70°C (versus -0.4°C for January and 2.2°C for February on average over a thirty year period), but the plants are in hibernation. In the second ten days of March, with temperatures rising, the plants entered the tillering phase. The tillering phase of Elbrus variety is on 26th March 2015, and in the Predel variety it is three to four days later on 30th March 2015. The precipitation in April was 27.0 mm less than the norm, and this had a retaining effect on the growth of durum wheat. Temperatures for the same period were within the normal range for the month, and a shooting up phase occurred on 20.04.2015 for Elbrus variety and three days later on 23.04.2015 for Predel variety.

The rainfall in May and June was 12.5 mm and 15.7 mm less than the norm, respectively, while the average monthly temperature in May and June was higher than the norm, which led to an accelerated development of durum wheat. The stages of filling and ripening of the grain occurred under favourable rainfall conditions. The ear formation phase of Elbrus variety took place on 15.05.2015, while on 18.05.2015 for Predel variety. Full maturity for durum wheat was recorded on 01.07.2015 and on 03.07.2015 respectively for Elbrus and Predel varieties. Harvest of durum wheat was carried out on 09.07.2015.

Table 1. Phenological phases of the durum wheat varieties tested

Development phases	2014-2015	
	Predel	Elbrus
Sowing	10.11	10.11
Germination	26.11	25.11
3rd leaf	17.12	15.12
Tillering	30.03	26.03
Shooting up	23.04	20.04
Ear formation phase	18.05	15.05
Full maturity	03.07	1.07

Table 2. Duration, temperature sum, and precipitation during interfacial periods

Year	Interfacial periods	Number of days		∑ *t* active, (°C)		*t* average, (°C)		The amount of rainfal, mm/m^2	
		Predel	Elbrus	Predel	Elbrus	Predel	Elbrus	Predel	Elbrus
2014 - 2015	Sowing - germination	17	16	144.0	144.0	8.47	9.0	48.7	48.7
	Germination – 3rd leaf	22	21	77.3	77.3	3.51	3.68	78.8	78.7
	3rd leaf - tillering	104	102	394.6	345.4	3.79	3.38	246	239.3
	Tillering - shooting up	25	26	297.6	311.6	11.90	11.98	13.6	20.3
	Shooting up - ear formation phase	26	26	451.0	420.5	16.17	17.34	66.0	64.7
	Ear formation - ripening	47	48	976.2	993.1	20.77	20.68	87.3	79.5

The data in Table 2 on the characteristics of the interphase periods shows similar numbers of days, the sum of the active and average t°C for both varieties. The germination of the plants takes place for 16 and 17 days at an equal sum of active temperature of t°C - 144°C for Predel and Elbrus varieties and at average temperatures of 8.47-9.0°C with an equal amount of precipitation for the period - 48.7 mm. The interphase period "germination - third leaf" runs for 21 and 22 days also at the same temperature sums of 77.3°C, with average temperature for the period 3.5-3.7°C, and precipitation quantity with close values 78.7-78.8 mm for both varieties. The longest period is the period of the
3rd leaf - tillering, i.e. 102 days for Elbrus and 104 days for Predel variety, the sum of the active temperatures for Predel variety being greater than that of Elbrus variety, respectively 394,6°C and 345,4°C. The average t°C for the period of Elbrus variety is 3.38°C, and for Predel variety is 3.79°C, and the precipitation quantity is 239.3 mm and 246.0 mm, respectively. The interphase period of tillering - shooting up occurs in shorter time in Predel variety, 25 days, and in Elbrus variety for 26 days. The sums of active temperatures are slightly different - 297.6°C for Predel and 311.6°C for Elbrus. Average temperatures are close to 12.0°C, and the amount of rainfall in Elbrus variety is greater, 20.3 mm, versus that of Predel, 13.6 mm.

The same is the duration of the interphase period tillering - ear formation equalling to 26 days for both varieties. It occurs at sum of the active temperature of 451.0°C and precipitation quantity of 66.0 mm for Predel variety, the values of which are greater than those of the Elbrus, namely 420.5°C, and 64.7 mm. The value of the average t°C for Elbrus variety period is 17.34°C, being higher than that of Predel variety for the same period - 16.17°C. "Ear formation - ripening" takes place for 48 days for Elbrus variety, and 47 days for Predel variety. The sum of the active temperatures is greater for Elbrus variety 993.1°C, compared with the Predel variety, 976.2°C, while the average temperature for the period and the amount of precipitations are higher for Predel, 20.77°C and 87.3 mm, compared to the Elbrus variety, 20.68°C and 79.5 mm.

In the second experimental year, the sowing of durum wheat was carried out on 21th October 2015. This year, the soil moisture reserves due to fallen rainfall combined with the higher temperature compared to the multiannual period, create good conditions for the germination of the plants.

Higher temperatures at the end of October and the amount of fallen rainfall contributed to the normal germination phase at the end of the month. In the early stages of development no significant differences were observed between the two tested varieties.

Elbrus variety enters one day earlier, respectively, in phase germination and third leaf compared to Predel variety (Table 3).

Table 3. Phenological phases of the durum wheat varieties tested

Development phases	2015-2016	
	Predel	Elbrus
Sowing	21.10	21.10
Germination	31.10	30.10
3rd leaf	18.11	17.11
Tillering	09.03	07.03
Shooting up	15.04	13.04
Ear formation phase	12.05	09.05
Full maturity	02.07	01.07

Predel variety enters 3rd leaf phase on 18.11.2015, while Elbrus variety on 17.11.2015. Plants enter the beginning of the tillering phase on 20.11.2015.

Gradually, with the fall in temperatures in December, the growth processes of durum wheat subside. In January, the amount of precipitation was more, and in February and March and April less than the multiannual period norm. These four months were also characterized by higher average monthly temperatures. The occurrence of the tillering phase of Elbrus variety is on 07.03.2016, while in Predel variety it is two days later on 09.03.2016. Temperatures and precipitation in April were higher compared to a multiannual period of time and shooting up phase occurred on 13.04.2016 for Elbrus variety and two days later on 15.04.2016 for Predel variety.

The phases of filling and ripening of the grain took place in favourable rainfall conditions and optimal temperatures in May and June. The shooting up phase occurred on 13 April 2016 for Elbrus variety, and two days later on 15 April 2016, for Predel variety.

Ear formation phase of Elbrus variety took place on 09.05.2016, while for Predel variety on 12.05.2016. Full maturity for durum wheat was reported on 01.07.2016 and on 02.07.2016 respectively for Elbrus and Predel varieties. Harvest of durum wheat was carried out on 11.07.2016.

In conclusion, it can be said that Elbrus variety in both years of research is developing faster and entering the different stages of development earlier than Predel variety.

Varieties that can complete the vegetation more quickly are advantageous in avoiding later the summer heat and could be an indication of a good adaptability of Elbrus variety for growing in the Plovdiv region.

According to Gandee et al. (1997), a longer vegetation period means higher yields unless grain filling occurs in a time of extreme drought. Others, Giordani et al. (1989) found that at high temperature and water deficiency, factors that are known to accelerate the phenophase development, less number of days are required for going through the respective phenophase.

According to the same author (1989), the longer duration of grain formation time is reflected in a larger number of grains. Plants are most sensitive to the impact of unfavourable factors in the ear formation phase when the processes of gametogenesis are completed and pollen and ova are formed.

Table 4. Duration, temperature sum, and precipitation during interfacial periods

Year	Interfacial periods	Number of days		∑ *t* active, (°C)		*t* average, (°C)		The amount of rainfal, mm/m²	
		Predel	Elbrus	Predel	Elbrus	Predel	Elbrus	Predel	Elbrus
2015 - 2016	Sowing - germination	11	10	100.8	92.1	9.16	9.21	39.6	39.6
	Germination – 3rd leaf	19	19	215.5	216.0	11.34	11.36	0	0
	3rd leaf - tillering	114	113	628.9	618.5	5.51	5.47	139.4	135.9
	Tillering - shooting up	38	38	397.6	402.4	10.46	10.58	45.7	48.5
	Shooting up - ear formation phase	28	27	414,0	396.6	14.78	14.68	41.9	38.7
	Ear formation - ripening	51	54	1117	1168	21.91	21.62	103.9	97.7

The germination of plants in the second year occurs for 10 and 11 days with sum of the active temperature t°C, 100.8°C for Predel variety and 92.1°C for Elbrus, and at average temperatures of 9.16-9.21°C with the same amount of precipitation for the period - 39.6 mm (Table 4). The interphase period "germination - 3rd leaf" occurs for 19 days for both varieties also at close temperature sums of 215.5-216.0°C, average temperature for the period 11.34-11.36°C, and the same precipitation amount 0.0 mm. The longest is the period "3rd leaf - tillering, consisting of 113 days for Elbrus and 114 days for Predel, the sum of the active temperatures for Predel variety - 628.9°C being greater than that of Elbrus variety - 618.5°C. The average t°C for the period of Elbrus is 5.47°C, while for Predel variety, 5.51°C, and the precipitation quantity, 135.9 mm and 139.4 mm, respectively. The interphase period "tillering - shooting up" takes place for 38 days in both varieties of durum wheat. The sums of active temperatures are slightly different - 397.6°C for Predel and 402.4°C for Elbrus.

The average temperatures are close to 11.0°C, while the amount of rainfall for Elbrus variety is 48.5 mm being higher than that of Predel - 45.7 mm. The interphase period "shooting up - ear formation" occurs for 27-28 days. It occurs at sum of the active temperature, 414.0°C, and rainfall quantity, 41.9 mm for Predel variety, the values of which are higher than those of Elbrus, 396.6°C and 38.7 mm, respectively. The value of the average t°C for the period for Elbrus variety is 14.68°C being lower than that of Predel variety for the same period - 14.78°C. "Ear formation - ripening" takes place for 54 days for Elbrus variety, and 51 for Predel variety. The sum of the active temperatures is higher for Elbrus variety, 1168°C, versus that of Predel variety, 1117°C, while the average temperature for the period and the precipitation quantity are higher for Predel, 21.91°C and 103.9 mm, versus Elbrus variety, 21.62°C and 97.7 mm, respectively.

Soil climatic conditions are an important factor for grain quality in cereals. Plant feeding, however, in the form of soil fertilizer application or use of leaf fertilizers can compensate for adverse abiotic factors and strongly affect the chemical composition of the grain and increase its quality.

The Table 5 presents the results of the surveyed indicators, which are expressed as a percentage and represent an average of three repetitions for the two consecutive years. From the analysis of grain chemistry, it has been found that application of Megafol-protein results in an increase in the crude protein content of the Predel grain. This was observed in the three phases of development, with the increase being most pronounced after the foliar fertilizer application (14% crude protein) compared to the control (12.5%). These results were in positive correlation with the nitrogen content

(2.5%) and gluten (wet - 31.2% and dry - 14.7%) during the tillering phase treatment. Elevated crude fat content of the Predel variety was also observed as a result of the use of Megafol-protein, the highest value being reported after treatment in the heading phase (3.6%).

In the Elbrus variety, the application of Megafol-protein in the heading phase only leads to an increase in the nitrogen content (2.4%) and protein (13.8%). Its application in the remaining phases does not result in significant differences in these indicators. The application of Megafol during the tillering and jointing phases in the Elbrus variety leads to an increase in the gluten content (27.8% - tillering, 26.1% - jointing). The crude fat content of the Elbrus variety was increased after administration of Megafol-protein in the tillering phase (3.1%) and jointing (2%), while using Megafol - only in the heading phase (2.3%).

Another indicator that is affected by application of leaf fertilizers is the content of carbohydrate substances (in particular starch accumulation). The carbohydrates and protein content are in reverse correlation. The highest content of carbohydrate substances was recorded in the variants with the lowest protein content, and this was observed in both tested varieties and the two tested products.

Table 5. Chemical composition of the grain of two varieties of durum wheat (Predel and Elbrus) treated with leaf fertilizers (Megafol and Megafol-protein) at different stages of their development

Variety	Variants	% WGY	% DGY	% N	% Crude protein	% Carbohydrates	% Fibre	% Crude fats
Predel	$A_1B_1C_0$	29,5	13,1	2,2	12,5	84,1	1,0	2,4
	$A_1B_1C_1$	24,2	11,5	2,4	13,4	82,3	3,1	1,2
	$A_1B_1C_2$	**31,2**	**14,7**	**2,5**	**14,0**	82,0	1,4	**2,6**
	$A_1B_2C_0$	28,1	12,1	2,3	12,9	82,1	3,1	1,9
	$A_1B_2C_1$	23,8	10,1	2,3	13,0	81,6	3,1	2,3
	$A_1B_2C_2$	**32,0**	**14,3**	**2,4**	**13,6**	81,7	1,4	**3,3**
	$A_1B_3C_0$	23,4	10,7	2,2	12,7	84,6	1,5	1,2
	$A_1B_3C_1$	23,6	10,9	2,2	12,4	82,4	1,7	3,5
	$A_1B_3C_2$	**27,2**	**12,4**	**2,3**	**13,2**	78,3	4,9	**3,6**
Elbrus	$A_2B_1C_0$	25,5	12,7	2,1	12,0	85,1	1,2	1,8
	$A_2B_1C_1$	**27,8**	**13,3**	2,1	11,7	**85,7**	1,3	1,3
	$A_2B_1C_2$	26,6	12,1	2,1	11,8	82,0	3,1	**3,1**
	$A_2B_2C_0$	22,7	10,1	2,1	12,1	83,0	3,6	1,4
	$A_2B_2C_1$	**26,1**	**12,7**	2,1	12,2	**85,4**	1,4	1,1
	$A_2B_2C_2$	19,6	8,8	2,1	11,7	83,9	2,4	**2,0**
	$A_2B_3C_0$	28,9	15,4	2,0	11,4	**84,8**	2,3	1,5
	$A_2B_3C_1$	19,4	8,9	2,2	12,5	83,0	2,2	**2,3**
	$A_2B_3C_2$	27,5	12,5	**2,4**	**13,8**	82,7	1,7	1,8

Legend: Factor A (variety) - A_1 - Predel, A_2 - Elbrus; factor B (development phases used for the treatment) - B_1 - tillering, B_2 - jointing, B_3 - heading; Factor C (leaf fertilizer) – C_0 - control (untreated), C_1 - Megafol 300 ml / ha, C_2 - Megafol protein 300 ml / ha. % WGY - percentage of wet gluten yield, % DGY - yield of dry gluten.

Regarding the fiber content, no clear relationship was found between the type of leaf fertilizer, the application phase and the variety.
Similarly to our results, were the findings of other authors worked with wheat (Delchev et al., 2004; Blandino et al., 2015; Smith et al., 2005). For example, late leaf nourishment (in flowering phase) with nitrogen-containing fertilizers increases the grain protein content of durum wheat and bread wheat (Blandino et al., 2015).
Synthesis of protein is a complex and multi-step process requiring a lot of energy that is provided by solar radiation. A large amount of water is required to form carbohydrates, causing water deficiency to disturb the synthesis of carbohydrates. In cereals, there is a reverse relationship between the amount of protein and starch: with increased protein content in the grain, the amount of starch decreases and vice versa. In order to improve the quality of cereals, the nitrogen fertilizers are also important (Delchev, 2000). Nitrogen feed fertilization by spring increases not only yield but also improves grain quality. Of great importance for the quality are the nitrogen-containing complex fertilizers applied in the late stages of development - for example, during the period of jointing, heading and flowering (Delchev, 2010). The nitrogen introduced in these terms is used by the plant, not for the growth of the vegetative organs, but it reaches the reproductive organs, increases the concentration of nitrogen in them and during the period of grain development and growth, the protein synthesis processes are much more intense (Delfine et al., 2005; Stoyanova, 2010). In leaf fertilizers, nitrogen uptake faster into plants and transported into seeds, creating excellent conditions for intense protein synthesis. This can be used with great effect in unfavorable climatic conditions to increase yields and improve quality. In excessively humid years, when conditions are created to produce grains with degraded properties (low protein and high starch content), it is advisable to use nitrogen containing fertilizers (soil or leaf applied) even in the late stages of plant growth. In very dry years, when plants need to be grown under irrigation conditions to increase yields and quality, additional fertilization may also be used.

CONCLUSION

The use of leaf fertilizers containing nitrogen and/or amino acids increases the quality of wheat of Predel and Elbrus varieties. The phase in which fertilizers are applied is essential. In this study, we found that the use of Megafol-protein had the most significant effect on the crude protein content of the Predel grain when applied in the tillering phase, and in the Elbrus grain when applied in the heading phase. The use of Megafol has a significant positive effect on the gluten content of the Elbrus variety only after application in the jointing phase.

REFERENCES

Blandino, M., Vaccino, P., Reyneri, A. (2015). Late-Season Nitrogen Increases Improver Common and Durum Wheat Quality. *Agronomy Journal*, *107*(2), 680–690.

Delchev, G. (2000). Exploration of the timing of import of certain complex foliar fertilizers in durum wheat. *Plant Growth Sciences,* BG, *37*(9), 738–742.

Delchev, G., Ivanova, I., Nenkova, D. (2004). Study of some combinations of growth regulators and complex foliar fertilizers in durum wheat. *Plant Growth Sciences, XLI*(6), 552–555.

Delchev, G. (2010). Grain yield and quality of durum wheat when treated with some complex and organic foliar fertilizers. *Soil Science, Agrochemistry and Ecology, XLIV*(3), 46–51.

Delchev, G., and Stoyanova, A. (2013). Changes in the sowing properties of durum wheat seed in the use of leaf fertilizers, stimulants and antitranspirants. *Plant studies, III*(6), 215–216.

Delfine, S., Tognetti, R., Desiderio, E., Alvino, A. (2005). Effect of foliar application of N and humic acids on growth and yield of durum wheat. *Agron. Sustain. Dev*., *25*, 183–191.

Gandee, C.M., Davies, W.P., Goodind, M.J. (1997). Interactions between nitrogen and growth regulator on wheat and spelt. *Aspects of Applied Biology*, *50*, 219–224.

Giordani, G., Baldoni, G., Rapparini, L. (1989). Influenza di trattamenti fitoregulatori su varieta di frumento duro. *Informatore Agrario*, *45*(15), 115–118.

Giordani, G., Rapparint, L. (1989). Influenza della concimacione azotata e dei trattamenti fitoregulatori su varieta di frumento tenero e duro. *Informatore Agrario*, *45*(16), 83–90.

Smith, G.H., Chaney, K., Murray, Ch., Le, M.S. (2015). The Effect of Organo-Mineral Fertilizer Applications on the Yield of Winter Wheat, Spring Barley, Forage Maize and Grass Cut for Silage. *Journal of Environmental Protection*, *6*, 103–109.

Stoyanova, A. (2010). Foliar fertilizers and wheat grain quality. *Agriculture Plus*, *5*, 33–35.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

HEAT STRESS TOLERANCE OF SOME GREEN BEAN (*Phaseolus vulgaris* L.) GENOTYPES

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Abstract

The seedlings of 3 green bean genotypes ('Balkız', 'Ferasetsiz', 'Local Genotype') were grown for 4 weeks at 30/15°C (day/night) temperature with 65% humidity and then transferred to a growth chamber. The temperature in the growth chamber was increased stepwise to 35, 40, 45 and 50°C to create a heat-stressed environment. In addition, to determine HST (LT_{50}), the leaf discs of each genotype were exposed to 35, 40, 45, 50, 55 and 60°C. According to results, 'Ferasetsiz' and 'Local Genotype' had the highest leaf relative water content (RWC), while the 'Balkız' had the lowest. However, 'Ferasetsiz' and 'Local Genotype' had the lowest loss of turgidity and malondialdehyde (MDA), while 'Balkız' had the highest. The leaf area and chlorophyll content (Chl) were not associated with the HST of genotypes. Many protein bands, in mass range 27–157 kDa, were detected depending on the genotypes and heat treatments. Besides, a few new proteins appeared in response to heat stress. Based on the data collected, 'Ferasetsiz' and 'Local Genotype' were determined to heat tolerant genotypes, while 'Balkız' was relatively heat sensitive genotype.

Key words*: green bean, Phaseolus vulgaris L., high temperature, malondealdehyde, SDS-PAGE, stress tolerance.*

INTRODUCTION

Higher growing period temperatures can have impressive affects on agricultural productivity, farm revenues, and food safety (Battisti & Naylor, 2009). According to the report of the Intergovernmental Panel on Climate Change (IPCC), 0.3-0.7°C of warming is expected globally between 2016 and 2035 and to 0.3-4.8°C by the end of this century (IPCC, 2014).
Heat stress generally damages photosynthetic activity, and the reduced water content caused by heat has negative effects on cell division and growth (Hasanuzzaman et al., 2013). Heat stress also causes impaired to the proteins, lipid liquefaction or perturbation of membrane integrity (Levitt, 1980).
Plants have evolved delicate mechanisms to combat stressful environments. A gradual increase in temperature informs organisms that they should ready themselves in case they undergo even warmer conditions (Salomé, 2017). This process has significant effect on inducement of tolerance to lethal high temperatures (Hasanuzzaman et al., 2013).
Legumes are valued worldwide as a primary food source and low-cost meat alternative and considered to important rotational crop used to improve soil nitrogen status (Maphosa & Jideani, 2017). Because of the growing request for plant products and environmental pressures on agro-eco systems, it comes out that legumes would play an important function in here after cropping systems (Stagnari et al., 2017). On the other hand, an increase in global temperature, as a result of climate change, increments the risk of a heat stress-induced diminution in legume crop yield (Ozga et al., 2017). Hence, to ameliorate heat tolerance in legume crops contributing to improved food security. In this research, genotypic variations in 3 green bean genotypes and their relationship to heat-stress tolerance were investigated. The main aim of the study is to provide data for afterwards, more comprehensive works on green bean heat-tolerance.

MATERIALS AND METHODS

The seeds of green bean genotypes, 'Balkız', 'Ferasetsiz', and 'Local Genotype' were sown in pots 14 cm × 12 cm containing a mixture of peat, perlite and soil (1: 1: 1). The plants were grown for 4 weeks (3-4 trifoliate leaves) in a controlled greenhouse with day/night temperature of 30°C/15°C and approximately 65% relative humidity. The plants were watered as needed to avoid any water stress.

The plants were transferred to a growth chamber (DAIHAN WGC-1000, South Korea) with a relative humidity of 65%, a 16/8 h (light/dark) photoperiod regime and a 450 $\mu mol/m^2/s$ light intensity. Then, the temperature was increased stepwise, 5°C every 24 h from 35 to 40, 45 and 50°C to impose heat stress gradually. Plants were kept in the growth chamber for 24 h at each temperature. Control plants were held in the greenhouse during the treatment with a 30/15°C day/night temperature. All temperature treatments were replicated three times with all genotypes. Samples that were obtained at each treatment temperature were analyzed for leaf relative water content (RWC), loss of turgidity, leaf area, chlorophyll content (Chl), lipid peroxidation (MDA) and protein content.

Leaf RWC (%) and loss of turgidity were measured according to Gulen and Eris (2003). Briefly, 1.5 cm leaf discs were taken and the fresh weight (FW) was recorded. Then, the leaf discs were floated on distilled water in a petri dish for 4 h at room temperature then leaf discs were taken out from the petri dish, blotted and turgid weight (TW) was recorded. The leaf discs were oven-dried for 48 h at 70 °C for the dry weight (DW). The leaf RWC and loss of turgidity were calculated as follows:

RWC (%) = [(FW -TW)/(TW -DW)]*100 and

Loss of turgidity (%) = [(TW-FW)/ TW]*100

The leaf area was determined for three plants per replicate by a portable area meter (LI-3000, Li-Cor, Lincoln, NE, USA).

Changes in Chl of green bean leaves were analyzed spectrophotometrically as described by Moran and Porath (1980). Leaf samples subjected to dimethylformamide (DMF) extraction were incubated at +4ºC for 72 hours. The absorbance of supernatants was measured at 652 nm by a spectrophotometer (Perkin Elmer Lambda 25, USA). The content was calculated as mg/g FW.

Malondialdehyde content was determined in the leaves as described by Rajinder et al. (1981). Briefly, 100 milligram of leaf sample was homogenized in 5 ml of 0.1% trichloroacetic acid (TCA) and centrifuged at 10000×g for 5 min at 4°C (Beckman Coulter Allegra 64R, USA). An aliquot supernatant (0.3 ml) was mixed with 1.2 ml of 0.5% thiobarbituric acid (TBA) prepared in TCA 20% and incubated at 95°C for 30 min. After stopping the reaction in an ice bath for 5 min, samples were centrifuged at 10000×g for 10 min at 25°C. The supernatant absorbance was read at 532 and 600 nm using a spectrophotometer (Perkin Elmer Lambda 25, USA). Malondialdehyde content was determined using the extinction coefficient 155 mM/cm.

To determine total soluble protein (TSP) content, fully expanded leaf material was collected from each plant group at each temperature application step. Triplicate samples of leaf tissues were immediately frozen and ground in liquid N_2 and stored at - 80 °C until used. Total soluble proteins were extracted in the light of Shen et al. (2003) method with few modifications. In brief, 0.25 g of each sample was homogenized in 1 ml homogenate buffer containing 25 mM Tris-base (pH 7.8), 275 mM sucrose, 2 mM EDTA, 10 mM Dithiothreitol (DTT), 0.5 mM phenylmethylsulfonyl fluoride (PMSF), and 1% polyvinylpolypyrolidone (PVPP). The homogenate was transferred into an eppendorf tube and then centrifuged at 10000 rpm for 10 min at 4°C. Protein content was measured using the Bradford assay method (Bradford, 1976).

Discontinuous sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) was performed with a PROTEAN tetra vertical electrophoresis unit (Bio-Rad, Hercules, CA, USA) using 0.04 stacking gel and 0.125 separating gel. An equal amount of total protein (7.5 mg) was loaded for each sample and gels were stained with Coomassie Brilliant Blue G-250.

Cell membrane injury and HST (LT_{50}; denoted by electrolyte leakage) were determined as previously described by Arora et al. (1998) to assess the six temperature steps-35, 40, 45, 50, 55, 60°C. Briefly, green bean leaf discs of 1.5 cm diameter from each treatment group were placed in test tubes containing 500 µl of deionized water. Tubes were capped and placed in a thermostatically controlled water bath then the temperature was increased by 5°C at 30 min intervals up to 45°C, and then the bath temperature was increased by 1°C at 5 min intervals up to 60°C. Samples were allowed to equilibrate and then were kept for 30 min at each temperature (35, 40, 45, 50, 55 and 60°C). Three tubes per temperature were removed and placed in an incubator. Three tubes of the control (un-stressed) discs remained at 30°C as controls. One hour after the last sample was removed from the bath, 10 mL of deionized water was added to each test tube, and it was incubated overnight. The electrical conductivity of each solution was measured using a conductivity meter (YSI 3200, USA) then immediately autoclaved. Total conductivity was determined once more when the solution in test tubes cooled down to room temperature. Cell membrane injury was defined as the percentage of total ions present in the tissue (Gulen & Eris, 2003). In addition, HST (LT_{50}) was calculated as the mid-point between the maximum injury and the control, which expresses the temperature causing half-maximum percentage injury.

The experiment was arranged in a randomized block design, with three replications. The data were tested with SPSS 20.0 for Windows (SPSS Inc., Chicago, IL, USA) and the means were evaluated by the Duncan test at P<0.05.

RESULTS AND DISCUSSIONS

The changes of leaf RWC and loss of turgidity values of the genotypes are shown in Figure 1A and Figure 1B, respectively. The data from the present study showed that leaf RWC slightly increased at temperatures up to 45°C in 'Ferasetsiz' and 'Local Genotype', while it decreased significantly at 45ºC and 50ºC. However, leaf RWC value was greater only at 35ºC than that of control treatment and a remarkable decrease was observed at temperatures from 40 ºC and up in 'Balkız' genotype (Figure 1A).

In contrast to RWC results, loss of turgidity of leaf samples slightly decreased at temperatures up to 45°C in 'Ferasetsiz' and 'Local Genotype', while it increased significantly at 45ºC and 50ºC. On the other hand, loss of turgidity value was lower only at 35ºC than that of control treatment and it increased significantly at temperatures from 40ºC and up in 'Balkız' genotype (Figure1B). A two-way ANOVA revealed significant effects of temperature, genotype and the interaction between temperature and genotype on leaf RWC and loss of turgidity (Table 1).

Leaf RWC represents an indicator of plant water balance because it expresses the absolute water amount the plant requires to reach artificial full saturation (Siddiqui et al., 2015). A decrease in RWC in response to high temperature was also reported in strawberry (Gulen & Eris, 2003; Kesici et al., 2013), olive (Cansev, 2012) and lablab bean (Myrene & Devaraj, 2013). The differences between the leaf RWC in genotypes can be a sign of difference in leaf hydration, leaf water deficiency and physiological water conditions. First of all heat stress causes a decrease in leaf RWC and the loss of turgidity, which is a consequence of elevated transpiration (Cansev, 2012; Turhan et al., 2014). Besides, De Belie et al. (2000) pointed out that the cause of turgor loss in high temperature stress conditions may be related to the permeability of the cell membrane at high temperature.

Table 1. Results of analysis of variance (ANOVA) of genotype (G), temperature (T) and their interactions with RWC, Loss of turgidity, Leaf Area, Chl, MDA and TSP content in leaf tissues. Numbers represent F values relative to a significance level of 0.05

Dependent Variable	**Independent Variable**		
	G	T	GxT
RWC	9.054*	350.439*	16.422*
Loss of Turgidity	5.461*	188.007*	12.084*
Leaf Area	58.115*	60.589*	3.308*
Chl	2.216^{ns}	74.421*	10.284*
MDA	257.973*	687.561*	62.923*
TSP	38.555*	25.543*	10.400*
LT_{50}	265.542*	1032.070*	45.495*

* Significant at p<0.05, ^{ns} Non-significant.

The changes in leaf area values of green bean genotypes are given in Figure 2A. Although there is no significant change in the leaf areas of genotypes up to 45ºC, a remarkable change was observed when it comes from 45°C to 50ºC. According to the average values, the maximum leaf area value occurred in 'Ferasetsiz' (42.09 cm^2) and 'Balkız' (37.86 cm^2), whereas the minimum leaf area value occurred in 'Local Genotype' (27.69 cm^2). When the treatments were compared, the highest leaf area value was determined in the control treatment with 47.66 cm^2, while the lowest leaf area value was found at 50ºC treatment with an average of 23.09 cm^2 (Figure 2A). A two-way ANOVA revealed significant effects of temperature, genotype and the interaction between temperature and genotype on leaf area (Table 1).

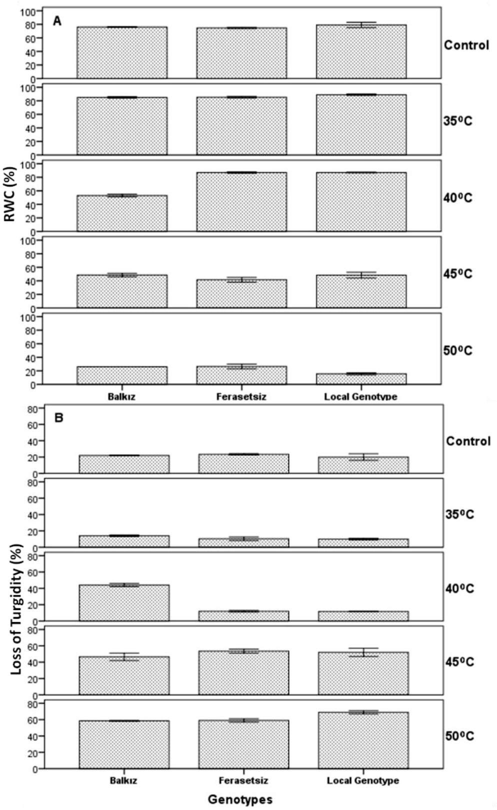

Figure 1. Leaf RWC (Panel A) and loss of turgidity (Panel B) of three green bean genotypes in heat-stressed and control samples. Vertical bars indicate ±S.E. of three replications

According to the results obtained from this study, it was determined that leaf areas of the genotypes generally decreased during high temperature applications. It was reported that the leaf area significantly reduced in sorghum (Djanaguiraman et al., 2010) and potato (Beetge & Krüger, 2019) depending on the high temperature applications. This situation may have thought to be because of the decrease in leaf area and decreases in photosynthetic activity parallel to the slowing of plant growth and development due to high temperature.

The changes in Chl in the green bean genotypes are shown in Figure 2B. According to the average values, the lowest Chl occurred in 'Balkız' (4.28 mg/g FW) and 'Ferasetsiz' (4.31 mg/g FW), whereas the highest Chl occurred in 'Local Genotype' (4.83 mg/g FW). In general, the average Chl in the control treatment (4.4 mg/g FW) was significantly lower than that in the 50 ºC treatment (8.35 mg/g FW). A two-way ANOVA revealed significant effects of temperature and the interaction between temperature and genotype on Chl but no significant effect of the genotypes (Table 1).

According to the results obtained from this study, it was determined that 50°C treatment increased total Chl in all genotypes. There are different results in the literature about this issue. For example, high temperature stress reduced the total Chl in mulberry (Chaitanya et al., 2001) and citrus (Guo et al., 2006). Besides, Cansev (2012) reported that high temperature applications did not affect total Chl up to 45°C in olives but decreased Chl from 50°C. Liu and Huang (2000) and Kesici et al. (2013) reported that a relationship between Chl and heat-tolerance in *Agrostis palustris* and strawberry, respectively. This can be explained by the fact that the physiological responses of plants to the stress conditions vary greatly depending on the species, cultivar, severity and the source of the stress.

The changes in MDA content in the leaf tissues are shown in Figure 3. The data from the five sampling treatments showed that 'Ferasetsiz' had the lowest MDA content with 46.8 nmol/g FW. 'Local Genotype' followed this genotype with 49.8 nmol/g FW. 'Balkız' had the highest MDA content with 90.3 nmol/ g FW. When the heat treatments were compared, the lowest MDA content was determined with an average of 24.7 nmol/g FW at 35ºC, while the highest MDA content was determined at 50ºC with 139.7 nmol/g FW. A two-way ANOVA revealed significant effects of temperature, genotype and the interaction between temperature and genotype on MDA content (Table 1).

Malondialdehyde is a final decomposition product of lipid peroxidation and is often used as an indicator for the status of lipid peroxidation (Smirnoff, 1995). High temperature exposure produced increased MDA content in all genotypes (Figure 3). This result is consistent with previous findings in rice (DongGi et al., 2007) and strawberry (Kesici et al., 2013). Gaschler and Stockwell (2017) also reported that the increase in lipid peroxidation (MDA content) is associated with cell membrane damage. Therefore, this result may indicate that the accumulation of ROS caused membrane damage.

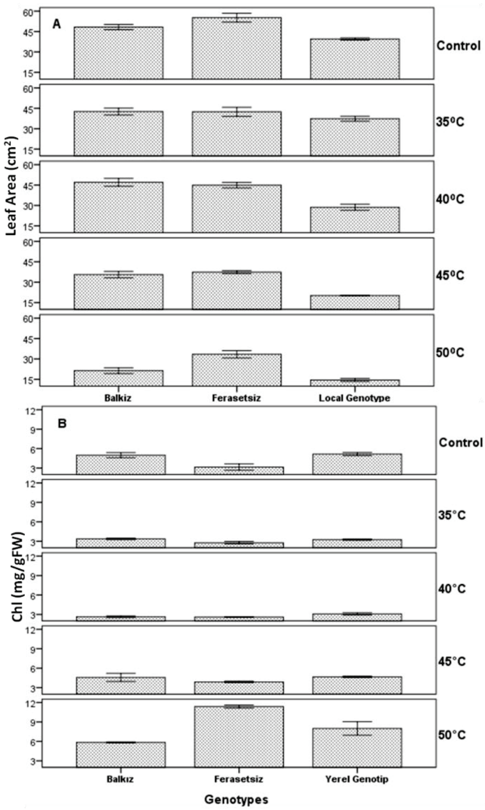

Figure 2. Leaf area (Panel A) and Chl (Panel B) of three green bean genotypes in heat-stressed and control samples. Vertical bars indicate ±S.E. of three replications

In general, it was observed that the effect temperature applications on the total soluble protein (TSP) content of green bean genotypes in leaf tissues was different (Figure 4). Total soluble protein content of genotypes decreased significantly compared to control treatment at 40ºC. A clear increase at 45ºC was observed TSP content of the 'Balkız' and 'Ferasetsiz' genotypes. There was a decrease in the TSP content of the 'Balkız' and 'Ferasetsiz' genotypes, at 50ºC. In the 'Local Genotype', the most significant reduction in TSP content was observed at 45ºC. A two-way ANOVA revealed significant effects of temperature, genotype and the interaction between temperature and genotype on TSP content (Table 1).

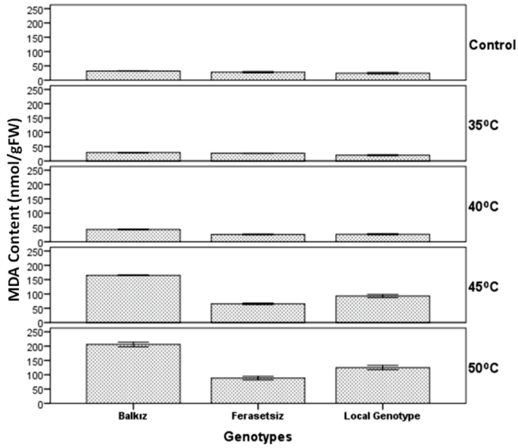

Figure 3. MDA content of three green bean genotypes in heat-stressed and control samples. Vertical bars indicate ±S.E. of three replications

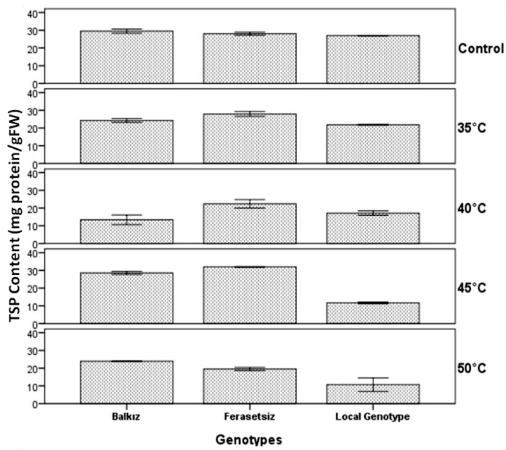

Figure 4. Total soluble protein content of three green bean genotypes in heat-stressed and control samples. Vertical bars indicate ±S.E. of three replications

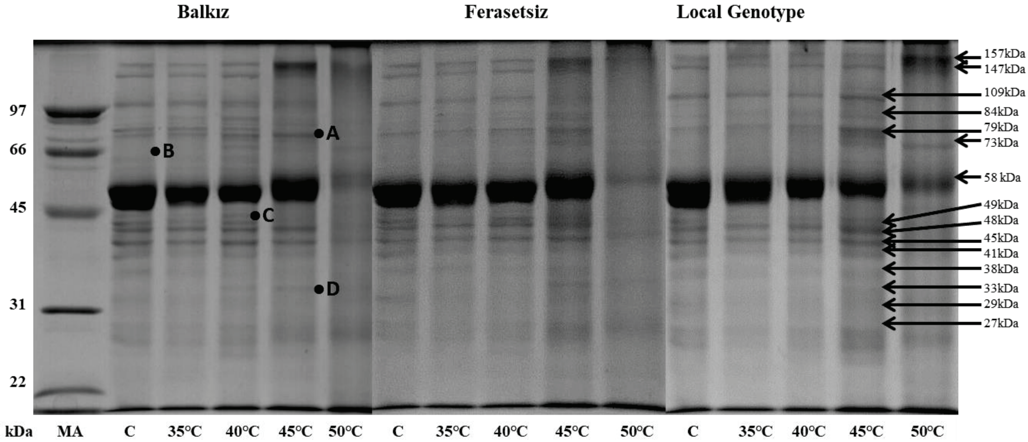

Figure 5. Effect of heat stress on protein profiles of green bean genotypes. MA: Marker, C. Control, A: 78 kDa; B: 65 kDa, C: 45 kDa, D: 34 kDa

Ledesma et al. (2004) reported that TSP content decreased, while new proteins had been synthesized depending on increased temperatures in "Nyoho" and "Toyonaka" strawberry varieties. High temperature stress also reduced the TSP content and increased the total amino acid and proline accumulation in mulberry (Chaitanya et al., 2001). Similarly, Turhan et al. (2015) determined that high temperature applications reduced the TSP content up to certain temperatures in pepper cultivars. High temperature stress leads to structural changes of proteins. Thus, proteins become denatured, proteolytic enzymes become sensitive (Levitt, 1980).

Figure 5 shows the total protein profiles in green been genotypes depending on high temperature applications. It has been determined that many protein bands, in mass range 27-157 kDa, were detected depending on the genotypes and heat treatments. Unlike the 'Ferasetsiz' and 'Local Genotype', in the 'Balkız' genotype, protein bands of 45, 65 and 78 kDa were determined and 34 kDA were different from the 'Local Genotype'. The density of the bands decreased in all three genotypes due to increasing temperature applications and this reduction was especially significant at 50°C. The 58 kDa protein band was the most frequently observed protein band in all three genotypes. It was determined that the 58 kDa protein band was probably a dominant band consisting of structural proteins and this band decreased in all genotypes at 50°C. On the other hand, some protein bands were completely lost at 50°C. In addition, it was determined that the protein band of 45 kDa was synthesized at 40°C in 'Balkız' genotype and this band lost again at 45-50°C. This decrease in protein bands due to high temperature is thought to be due to the deterioration of protein structure at increasing temperatures. Similarly, it was reported that new proteins are synthesized or reduced and almost complete lost in tomatoes (Heckathorn, 1998) and strawberries (Ergin et al., 2016; Ledesma et al., 2004) in response to high temperature stress. On the other hand, Ergin et al. (2016) determined that two protein bands of 40 kDa and 23 kDa accumulated at high temperatures (from 46°C) in strawberry leaves in gradual and shock high temperature applications. Similarly, Turhan et al. (2015) found that strips of 7-54 kDa in the pepper plant under high temperature stress conditions and 40 kDa protein band may be associated with high temperature tolerance.

The calculated stress tolerance point (LT_{50}) values of three green bean genotypes are shown in Figure 6. Heat-acclimation caused an increase in heat-tolerance of all genotypes, which was the highest in 'Ferasetsiz' (LT_{50}; 50.8ºC) and 'Local Genotype' (LT_{50}; 49.9ºC). Meanwhile, 'Balkız' had the lowest heat-tolerance with a LT_{50} value of approximately 45.5ºC. A two-way ANOVA revealed significant effects of temperature, genotype and the interaction between temperature and genotype on LT_{50} value (Table 1).

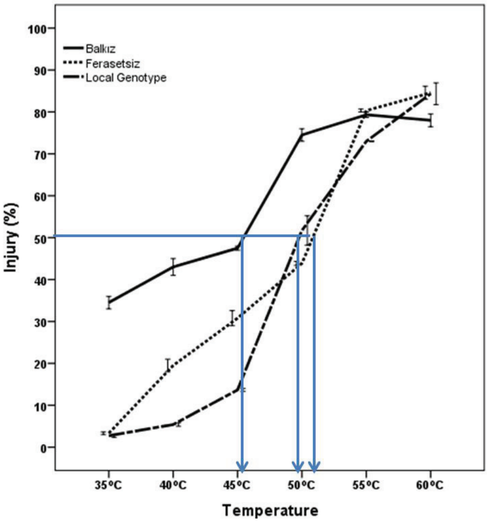

Figure 6. Heat stress tolerance (LT_{50}) of three green bean genotypes. Vertical bars indicate ±S.E. of three replications

In plants, measuring electrolyte leakage is a common evaluation way for cell membrane thermostability and membrane thermostability can be a valuable selection criterion for HST (Gulen & Eris, 2003). Electrolyte leakage is affected by the age, sampling part, development period, growing season, hardening and species of plant (Wahid et al., 2007). In this study, from the HST degrees of the genotypes, it can be suggested that 'Ferasetsiz' and 'Local Genotype' are relatively more tolerant to high temperatures than 'Balkız' genotype. Similar results have been found in strawberry cultivars by Kesici et al. (2013). Besides Cansev (2012) suggested that 'Gemlik' olive cultivar has moderate HST according to its LT50 value. Lethal temperature (LT_{50}) is the temperature at which 50% of individuals in a population are exposed to high damage or die. In a study, conducted on the *Deschampsia antarctica* Desv lawn variety it was showed that LT_{50} value as 48.3ºC due to membrane damage (Reyes et al., 2003). Turhan et al. (2015) demonstrated that, the LT_{50} values were determined as 36.9, 37.0 and 42.3 in 'Çarli', 'Çoban' and 'Demre' pepper varieties, respectively in the high temperature stress. They suggested that 'Demre' pepper cultivar was more tolerant to heat stress according to its LT_{50} value.

CONCLUSIONS

The determination of the tolerance of genotypes for the breeding of high temperature or tolerant genotypes in beans is important in terms of obtaining efficient genotypes. This will be possible by explaining the mechanisms developed by the plants against heat stress. In summary, leaf RWC, loss of turgidity and MDA content were found to be effective in determining the tolerances of some green bean genotypes to high temperatures. The LT_{50} showed the heat-tolerance between 45.5 and 50.8ºC in the green bean genotypes. The genotypes 'Ferasetsiz' and 'Local Genotype' were determined as relatively heat-tolerant, while 'Balkız' was relatively heat-sensitive among the 3 green bean genotypes evaluated. The results obtained from this study could shed light on the molecular mechanism of heat stress and future studies on the development of heat-tolerant green bean genotypes.

ACKNOWLEDGEMENTS

This study is part of Asli Tokyol's Master of Science Thesis Project, which was completed at Eskisehir Osmangazi University's Graduate School of Natural and Applied Sciences, Department of Horticultural Science. We would like to thanks Deniz Susan Muti for the English editing.

REFERENCES

Arora, R., Pitchay, D.S., Bearce, B.C. (1998). Water-stress-induced heat tolerance in geranium leaf tissues: A possible linkage through stress proteins. *Physiologica Plantarum*, *103*. 24–34.

Battisti, D.S., Naylor, R.L. (2009). Historical warnings of future food insecurity with unprecedented seasonal heat. *Science, 323*. 240–244.

Beetge, L., Krüger, K. (2019). Drought and heat waves associated with climate change affect performance of the potato aphid *Macrosiphum euphorbiae*. *Scientific Reports*, 9:3645, Retrieved March 25, from https://www.nature.com.

Bradford, M.M. (1976). A Rapid and sensitive method for quantitation of microgram quantities of protein using the principle of protein-dye binding. *Ann. Biochem*., *72*. 248–254.

Cansev, A. (2012). Physiological effects of high temperature treatments on leaves of olive cv. Gemlik. *Plant Arch., 12*(1), 521–525.

Chaitanya, K.V., Sundar, D., Masilamani, S. Reddy, A.R. (2001). Mulberry leaf metabolism under high temperature stress. *Biologia Plant*, *44*. 379–384.

De Belie, N., Herppich, W., De Baerdemaeker, J. (2000). A kinetic model for turgor loss in red cabbage cells during mild heat treatment. *Journal of Plant Physiology*, *157*(3), 263–272.

Djanaguiraman, M., Prasad, P.V.V., Seppanen, M. (2010). Selenium protects sorghum leaves from oxidative damage under high temperature stress by enhancing antioxidant defense system. *Plant Physiol. Biochem.*, *48*. 999–1007.

DongGi, L., Ahsan, N., Young, K. (2007). A proteomic approach in analyzing heat-responsive proteins in rice leaves. *Proteomics*, *7*(18), 3369–3383.

Ergin, S., Gülen, H., Kesici, M., Turhan, E., Ipek, A.N., Köksal, N. (2016). Effects of high temperature stress on enzymatic and nonenzymatic antioxidants and proteins in strawberry plants. *Turk. J. Agric. For.*, *40*. 908–917.

Gaschler, M.M., Stockwell, B.R. (2017). Lipid peroxidation in cell death. *Biochem Biophys Res. Commun., 482*(3), 419–425.

Gulen, H., and Eris, A. (2003). Some physiological changes in strawberry (*Fragaria x ananassa* cv. Camarosa) plants under heat stres. *J. Hort. Sci. Biotech., 78*. 894–898.

Guo, Y.P., Zhou, H.F., Zhang, L.C. (2006). Photosynthetic characteristics and protective mechanisms against photooxidation during high temperature stress in two citrus species. *Scienta Hort., 108*. 260–267.

Hasanuzzaman, M., Nahar K., Alam, M.M., Roychowdhury, R., Fujita, M. (2013). Physiological, biochemical, and molecular mechanisms of heat stress tolerance in plant. *Int. J. Mol. Sci*., *14*. 9643–9684.

Heckathorn, S.A., Downs, C.A., Sharkey, T.D., Coleman, J.S. (1998). The small, methionine-rich chloroplast heat shock protein protects photosystem II electron transport during heat stress. *Plant Physiol., 116*. 439–444.

IPCC Climate Change (2014). Synthesis Report Core Writing Team, R.K. Pachauri, L.A. Meyer (Eds.), Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change, IPCC, Geneva, Switzerland, 151.

Kesici M., Gulen, H., Ergin, S., Turhan, E., Ipek, A., Koksal, N. (2013). Heat-stress tolerance of some strawberry (*Fragaria × ananassa*) cultivars. *Not. Bot. Horti. Agrobo*., *41*. 238–243.

Ledesma, N.A., Kawabata, S., Sugiyama, N. (2004). Effect of high temperature on protein expression in strawberry plants. *Biol. Plant., 48*(1), 73–79.

Levitt, J. (1980). *Responses of plants to environmental stresses.* Academic Pres, pp. 497.

Liu, X., Huang, B. (2000). Heat stress injury in relation to membrane lipid peroxidation in creeping bentgrass*. Crop Sci., 40*. 503–510.

Maphosa, Y., Jideani, V.A. (2017). The role of legumes in human nutrition. *Functional Food-Improve Health through Adequate Food,* 103–122.

Moran, R., Porath, D. (1980). Chlorophyll determination in intact tissues using N,N-Dimethylformamide. *Plant Physiol*., *65*(3), 478–479.

Myrene, R. D'souza, Devaraj, V.R. (2013). Induction of thermotolerance through heat acclimation in lablab bean (*Dolichos lablab*). *African J. Biotech., 12*(38), 5695–5704.

Ozga, J.A., Kaur, H., Savada, R.P. Reinecke, D.M. (2017). Hormonal regulation of reproductive growth under normal and heat-stress conditions in legume and other model crop species. *J. Exp. Bot., 68*(8),1885–1894.

Rajinder, S.D., Dhindsa, P.P., Thorpe, T.A. (1981). Leaf senescence: Correlated with increased levels of membrane permeability and lipid peroxidation, and decreased levels of superoxide dismutase and catalase. *J. Exp. Botany.*, *32*. 93–101.

Reyes, M.A., Corcuera, L.J., Cardemil, L. (2003). Accumulation of HSP70 in *Deschampsia Antarctica* Desv. leaves under thermal stress. *Antarctic Science*, *15*. 345–352.

Salomé, P.A. (2017). Some like it hot: Protein translation and heat stress in plants. *The Plant Cell*, *29*. 2075.

Shen, S.H., Jing, Y.X., Kuang, T.Y. (2003). Proteomics approach to identify wound-response related proteins from rice leaf sheath. *Proteomics*, *3*. 527–535.

Siddiqui, M.H., Al-Khaishany, M.Y., Al-Qutami, M.A., Al-Whaibi, M.H., Grover, A., Hayssam M.A., Al-Wahibi, M.S. (2015). Morphological and physiological characterization of different genotypes of faba bean under heat stress. *Saudi Journal of Biological Sciences*, *22*. 656–663.

Smirnoff, N. (1995). Antioxidant systems and plant response to the environment, 217-243 p. In Smirnoff, N. (Ed.). Environment and plant metabolism: flexibility and acclimation. UK:Bios Scientific Publishers, Oxford.

Stagnari, F., Maggio, A., Galieni, A., Pisante, M. (2017). Multiple benefits of legumes for agriculture sustainability: an overview, C*hem. Biol. Technol. Agric*., *4*(2), 1–13.

Turhan, E., Aydogan, C., Ergin, S., Ozturk, N. (2015). Proteins in pepper plants (*Capsicum annuum* L.) under heat stress. *Journal of Biotechnology, 208*. 31.

Turhan, E., Aydogan, C., Ergin, S. and Ozturk, N. (2014). Variation in heat stress-induced some physiological changes and peroxidase activities among three tomato (*Lycopersicon esculentum* Mill.) cultivars. *Turkish J. Agric. Natural Sci., 2*. 1492–1498.

Wahid, A., Gelani, S., Ashraf, M., Foolad, M.R. (2007). Heat tolerance in plants: an overview. *Environ. Exper. Botany., 61*. 199–223.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

RESPONSE OF SOME CORN HYBRIDS TO DROUGHT STRESS

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Abstract

Corn (Zea mays. L), the most widespread agricultural crop in Romania, is frequently affected by drought. In order to investigate and quantify the effect of drought stress (hydric and thermal stress) on grain yield at seven hybrids of corn, an experiment was conducted in a randomized block with three replications during 2017 and 2018 at ARDS Simnic. Year 2017 was considered a dry year (with water and thermal deficiency), and 2018 a year favorable to corn (without stress). Six stress tolerance indices, including mean productivity (MP), geometric mean productivity (GMP), Yield Index (YI), Yield stability index (YSI), stress susceptibility (SSI) and stress tolerance index (STI) were used. The ANOVA test showed significant differences between the two culture conditions for the production. All hybrids experimented under favorable crop conditions (without stress) have achieved higher yields (7.37-10.16 tons/ha) and under drought conditions had a lower yield of 38% (4.32-6.16 tons/ha). The grain yield under drought conditions (stress) (Ys) was having significant positive correlation with GMP, YI, YSI and significant negative correlation with SSI. The yield under favorable field conditions (no-stress) (Yp) had a strong positive correlation only with MP index The verification of drought-tolerant crops using this rankings method indicated the hybrids HSF 158-14, HSF 734-13 and F376 as the most tolerant of drought.

Key words: *drought, corn, stress tolerance index.*

INTRODUCTION

The Oltenia area is often affected by drought and heat that strongly influence plant development and yield (Bonea & Urechean, 2015; Bonea, 2016).

Decrease of grain yield in corn due to drought, depends on genotype, stage of plant development at the moment of drought installation, intensity and duration of drought etc. Mi et al. (2018) showed that the drought manifested in the corn reproductive stage led to a reduction in grain yield of 41.6-46.6% and the drought installed in the vegetative stage at a reduction of 18.6-26.2%.

Bonea and Urechean (2017) reported that the drought during blooming phase has determined a significant reduction of the grain yield with 60.5%.

Although it is believed that the intensity of physiological processes decreases after reaching the physiological maturity, it seems that the august rainfall has a decisive role in defining production capacity, both in dry years and in favorable years (Urechean et al., 2010).

To reduce these negative effects, it is important to identify and use drought-tolerant hybrids in the culture.

Using plant genotypes adaptable to drought stress is an optimal strategy in sustainable agriculture (Asgarinia et al., 2017). According to Shiri et al. (2010) direct selection of drought tolerant hybrids by yield only has low selection effectively because of environmental influences sometimes are greater than genetic influence.

Therefore, the researchers proposed various techniques for assessing genetic differences between hybrids on their drought tolerance, such as stress tolerance indices based on a mathematical relation between stress and optimum conditions (Bouslama & Schapaugh, 1984; Fernandez, 1992; Fischer & Maurer, 1978; Gavuzzi et al., 1997; Rosielle & Hamblin, 1981).

The present study was conducted to identify corn hybrids with drought tolerance by using different selection indices and the ranking method. In order to investigate and quantify the effect of drought stress (hydric and thermal stress) on grain yield using different tolerance indices and rankings method.

MATERIALS AND METHODS

Seven Romanian corn hybrids were studied for production performance under two different environmental conditions, namely in 2017 and 2018, respectively.

The years of the investigation as regards weather conditions, were characterized as follows: 2017 was considered a dry year and 2018 was considered a favorable to corn.

Both experiences were placed in a randomized block (latin rectangle) with three replications at ARDS Simnic, Craiova.

Sowing was made on 10.04.2017 and 24.04.2018 respectively. The plant sown in the previous year was wheat.

Plowing was done in autumn and in spring preparing the suitable soil for germination was done with the disc and the combiner.

Fertilization was done with 250 kg/ha ($N_{20}P_{20}K_0$) complex fertilizers before sowing and in vegetation (phase 8-10 leaves) with ammonium nitrate 250 kg/ha.

The herbicide was made with DUAL GOLD 960-1.5 l/ha immediately after sowing and with EQUIP 1.5 l/ha + BUCTRIL 1.0 l/ha in vegetation (phase 6-8 leaves).

Two mechanical and two manuals weeding were applied.

The six stress tolerance indices: MP, GMP, YI, YSI, SSI and STI have been calculated on the basis of grain yield obtained in two conditions: without stress (Yp) and drought stress (Ys) using the following formulas:

Mean productivity (MP) (Rosielle & Hamblin, 1981), the genotypes with high value of this index will be more desirable:

$$MP = \frac{Y_S + Y_P}{2}$$

Geometric mean productivity (GMP) (Fernandez, 1992), the genotypes with high GMP value will be more desirable:

$$GMP = \sqrt{(Y_S)(Y_P)}$$

Yield Index (YI)) (Gavuzzi et al., 1997), the genotypes with high value of this index will be suitable for drought stress condition:

$$YI = \frac{Y_S}{\bar{\bar{Y}}_P}$$

Yield stability index; YSI (Bouslama & Schapaugh, 1984), the genotypes with high YSI values can be regarded as stable genotypes under drought and non-stress conditions:

$$YSI = \frac{Y_S}{Y_P}$$

Stress susceptibility index (SSI) (Fischer & Maurer, 1978), the genotypes with SSI < 1 are more resistant to drought stress conditions:

$$SSI = \frac{1-(\frac{Y_S}{Y_P})}{SI} \quad , SI = 1-(\frac{\bar{Y}_S}{\bar{\bar{Y}}_P})$$

Where SI = intensity of stress

Stress tolerance index (STI) (Fernandez, 1992), the genotypes with high STI values will be tolerant to drought stress:

$$STI = \frac{(Y_S)(Y_P)}{(\bar{Y}_P)^2}$$

For the evaluation of tolerant genotypes by the rankings method, was used the formula proposed by Farshadfar and Elyasi (2012):

Rank sum (RS) = Rank mean + Standard deviation of rank (SDR)

The data on the grain yield have been computed by variance analysis using ANOVA program for both environmental condition.

The phenotypical correlation between studied traits as well as the correlation between yields in both environmental conditions (Yp and Ys) and the six indices of stress tolerance have been interpreted by using simple correlation coefficients (r).

RESULTS AND DISCUSSIONS

ANOVA results showed that there were significant differences between hybrids for grain yield in both environmental conditions (p = 0.05) (Table 1).

All hybrids experimented under favorable crop conditions (without stress) have achieved higher yields (7.37-10.16 tons/ha) and under drought conditions had a lower yield of 38% (4.32-6.16 tons/ha).

Hybrids HSF 158-14 and HSF 153-14 achieved the highest yield under drought conditions and hybrids HSF 734-13 and F376 had the highest yield under without drought conditions (Table 2).

Photo 1. Corn during blooming phase

Photo 2. Corn at physiological maturity

The calculation of stress tolerance indices (Table 3) is not always eloquent and sufficient to identify stress tolerance hybrids. For example, based on MP and GMP, the most tolerant hybrid was HSF 734-13; based on YI and STI the most tolerant was the HSF 158-14 hybrid, and on the basis of YSI and SSI the most tolerant hybrid was HSF 154-14.

In a previous paper Bonea and Urechean (2011) confirm that using all tolerance indices there cannot be selected genotypes with similar tolerance.

Therefore, to determine the best indices in establishing stress tolerance, correlation coefficients were calculated between YP, Ys and the six indices used in this study (Table 4).

Numerous researchers (Kumar et al., 2015; Urechean & Bonea, 2017) believe that analyzing these correlations is a much better criterion for assessing drought tolerance of genotypes.

Table 1. Analysis of variance for grain yield under non-stress (Yp) and stress (Ys) conditions

Source of variations	Degree of freedom	Ys (MS)	YP (MS)
Genotype	6	1.213*	1.446*
Error	7	0.054	0.046

MS = mean square; * = significant at p=0.05

Table 2. Mean comparisons of grain yield under non-stress (Yp) and stress (Ys) conditions

Hybrid	Drought condition (Ys) tons/ha	Non-stress condition (Yp) tons/ha
F376	5.36	9.45*
F423	5.56	8.03^{0}
Oituz	4.32^{0}	8.46
Iezer	4.48^{0}	8.08
HSF 734-13	5.52	10.16*
HSF 153-14	5.91*	7.37^{0}
HSF 158-14	6.16*	8.48
Mean (CT)	5.33	8.57
LSD5%	0.45	0.41
% reduction	**38%**	

*, ^{0} = significant positive and negative, respectively at p = 0.05

Grain yield under without drought conditions (Yp) has significantly positive correlated only with the MP index (r = 0.80*).

Yield under drought stress (Ys) recorded a significant positive correlation with: GMP (r = 0.75*), YI (r = 1.00**); YSI (r = 0.75*) and a significant negative correlation with SSI (r = -0.77^{0}).

Similar results have been reported by Ghobadi et al. (2012) in bread wheat genotypes under post anthesis drought stress. These was observed the highest correlation positive (r = 1.00**) between Ys and YI and the significant negative correlations between Ys and SSI (r = -0.47*).

Ceceareli et al. (1987) reported that there was a negatively significant correlation between grain yield cereal under drought stress condition (Ys) and SSI.

Other significant positive correlations were recorded between MP and GMP (r = 0.98**), STI (r = 0.98**), between GMP and YI (r = 0.75*), STI (r = 1.00**); between YI and YSI (r = 0.78*), STI (r = 0.74*).

Significant negative correlations were recorded between YI and SSI (r = -0,77^{0}) and between YSI and SSI (r = -1.00^{00}). Similar results have been reported by Sayyah et al. (2011) in bread wheat genotypes under post-anthesis drought stress.

According to Mitra (2001) some suitable indices must have a significant correlation with grain yield under both the conditions.

Because, in our study, have not been identified indices of drought tolerance that correlate significantly with the production obtained in both environmental conditions, the rankings method was used (Table 5).

The same situation was observed by Sio-Se Mardeh et al. (2006) and Bonea and Urechean (2017) in severe drought conditions.

According to Farshadfar and Elyasi (2012) the genotypes with the lowest RS (sum of the ranks) value are the most stable.

In our case, hybrids HSF 158-14, HSF 734-13 and F376 which recorded the lowest value for the sum of the ranks (RS), were identified as the most tolerant drought tolerant.

Numerous researchers have used a rankings method for the quantitative evaluation of all corn drought tolerance indices (Farshadfar & Sutka, 2002; Bonea et al., 2018).

Table 3. Drought tolerance indices (at SI = 0.38)

Indices / Hybrid	Yp	YS	MP	GMP	YI	YSI	SSI	STI
F376	9.45	5.36	7.40	7.11	1	0.56	1.15	0.68
F423	8.03	5.56	6.79	6.68	1.04	0.69	0.81	0.6
Oituz	8.46	4.32	6.39	6.04	0.81	0.51	1.28	0.49
Iezer	5.08	4.48	6.28	6.02	0.84	0.55	1.18	0,49
HSF 734-13	10.16	5.52	7.84	7.48	1.03	0.54	1.21	0.76
HSF 153-14	7.37	5.91	6.64	6.59	1.1	0.8	0.52	0.59
HSF 158-14	8.48	6.16	7.32	7.22	1.15	0.72	0.73	0.71

Yp = yield in non-stress condition; Ys = yield in stress condition; MP = Mean Productivity; GMP = Geometric Mean Productivity; YI =Yield Index; YSI=Yield stability index; SSI = Stress susceptibility index STI = Stress tolerance index; SI = intensity of stress

Table 4. Correlation coefficients between drought tolerance indices

	Yp	Ys	MP	GMP	YI	YSI	SSI	STI
Yp	-	-0.01	0.80*	0.65	-0.01	-0.63	0.64	0.66
Ys			0.59	0.75*	1.00**	0.78*	-0.77^{0}	0.74
MP				0.98**	0.59	-0.05	0.06	0.98**
GMP					0.75*	0.17	-0.16	1.00**
YI						0.78*	-0.77^{0}	0.74*
YSI							-1.00^{00}	0.16
SSI								-0.15
STI								

Yp = yield in non-stress condition; Ys = yield in stress condition; MP = Mean Productivity; GMP = Geometric Mean Productivity; YI =Yield Index; YSI=Yield stability index; SSI = Stress susceptibility index STI = Stress tolerance index; *$^{;0}$ and **$^{;00}$ - significant at 0.05 and 0.01 level of probability, respectively

Table 5. Ranking, average, standard deviation (SDR) and sum of rankings (SR) of tolerance indices

Hybrids / Indices	F376	F423	Oituz	Iezer	HSF 734-13	HSF 154-14	HSF 158-14
Yp	2	6	4	5	1	7	3
Ys	5	3	7	6	4	2	1
MP	2	4	6	7	1	5	3
GMP	3	4	6	7	1	5	2
YI	5	3	7	6	4	2	1
YSI	4	3	7	5	6	1	2
SSI	4	3	7	5	6	1	2
STI	3	4	6	7	2	5	1
Aveage	3.50	3.75	6.25	6.00	3.13	3.50	1.88
SDR	0.42	0.37	0.37	0.33	0.77	0.80	0.29
RS	3.92	4.12	6.62	6.33	3.90	4.30	2.17

CONCLUSIONS

The ANOVA test showed significant differences between the two culture conditions for the production.

All hybrids experimented under favorable crop conditions (without stress) have achieved higher yields (7.37-10.16 tons/ha) and under drought conditions had a lower yield of 38% (4.32-6.16 tons/ha).

The grain yield under drought conditions (*stress*) (Ys) was having significant positive correlation with GMP, YI, YSI and significant negative correlation with SSI.

The yield under favorable field conditions (no-stress) (Yp) had a strong positive correlation only with MP index.

The verification of drought-tolerant crops using this rankings method indicated the hybrids HSF 158-14, HSF 734-13 and F376 as the most tolerant of drought.

ACKNOWLEDGEMENTS

This research work was carried out with the support of Ministry of Agriculture and Rural Development and also was financed from Project ADER 1.1.2. Partnership No. 112/22.10.2015.

REFERENCES

Asgarinia, P., Mirlohi, A., Saeidi, G., MohamadiMirik, A.A., Gheysari, M., Razavi, V.S. (2017). Selection criteria for assessing drought tolerance in a segregating population of flax (*Linum usitatissimum* L.). *Canadian Journal of Plant Science, 97*(3), 424–437.

Bonea, D., Urechean, V. (2011). The evaluation water stress in maize (*Zea mays* L.) using selection indices. *Romanian Agricultural Research*, 28. 80–86.

Bonea D., Urechean V. (2015). The evaluation of heterosis for Romanian maize germplasm. *Pakistan. J. Bot.*, *47*(6), 2387–2390.

Bonea, D. (2016). The effect of climatic conditions on the yield and quality of maize in the central part of Oltenia. *Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series*, *XLVI* (1), 48–55.

Bonea, D., Urechean, V. (2017). Study on the selection for drought tolerance of some semi-late maize hybrids cultivated at A.R.D.S. Simnic. *Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series*, *XLVII*(1), 41–49.

Bonea D., Urechean V., Niculescu M. (2018). Yield and nutritional quality of different maize hybrids under drought stress. *Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series*, *48*(1), 44–53.

Bouslama, M., Schapaugh, T. (1984). Stress tolerance in soybean. Part I: Evaluation of three screening techniques for heat and drought tolerance. *Crop Science, 24*. 933–937.

Ceceareli, S., Nachit, M.M., Ferara, G.O., Mckin, M.S., Tahir, M., Leur, J.V., Serivastava, J.P. (1987). Breeding strategies for improving cereal yield and stability under drought. *Crop Science, 20*. 101–114.

Farshadfar, E., Sutka, J. (2002). Multivariate analysis of drought tolerance in wheat substitution lines. *Cereal Research Communication*, *31*. 33–39.

Farshadfar, E., Elyasi, P. (2012). Screening quantitative indicators of drought tolerance in bread wheat (*Triticum aestivum* L.) landraces. *European J. Exp. Biol.*, *2*(3), 577–584.

Fernandez, G.C.J. (1992). Effective selection criteria for assessing plant stress tolerance. Kuo C. G. (Ed.). *Proceedings of the international Symposium on Adaptation of Vegetables and other Food Crops in Temperature and Water Stress.* Taiwan, 257–270.

Fischer, R.A., Maurer, R. (1978). Drought resistance in spring wheat cultivars: I. Grain yield responses. *Aust. J. Agric. Res*., *29*. 897–912.
Gavuzzi, P., Rizza F., Palumbo, M., Campaline, R.G., Ricciardi, G.L., Borghi, B., (1997). Evaluation of field and laboratory predictors of drought and heat tolerance in winter cereals. *Can. J. Plant Sci*., *77*(4), 523–531.
Ghobadi, M, Ghobadi, M.E., Kahrizi, D., Zebarjadi, A.R., Geravandi, M. (2012). Evaluation of drought tolerance indices in dryland bread wheat genotypes under post-anthesis drought stress. *World Academy of Science, Engineering and Technology*, *67*. 1257–1261.
Kumar R., Kaul, J., Dubey, R.B., Singode, A., Chikkappa G. K., Manivannan, A., Debnath, M.K. (2015). Assessment of drought tolerance in maize (Zea *mays* L.) based on different indices. *SABRAO Journal of Breeding and Genetics, 47*(3), 291–298.
Mi, N., Cai, F., Zhang, Y., Ji, R., Zhang, S., Wang, Y. (2018). Differential responses of maize yield to drought at vegetative and reproductive stages. *Plant Soil Environ.*, *64*(6), 260–267.
Mitra, J., (2001). Genetics and genetic improvement of drought resistance in crop plants. *Curr. Sci*., *80*(6), 758–763.
Rosielle, A.A., Hamblin, J. (1981). Theoretical aspects of selection for yield in stress and non-stress environments. *Crop Sci.*, *21*(6), 943–946.
Sayyah, S.S., Ghobadi, M., Mansoorifar, S., Zebarjadi, A.R. (2011). Evaluation of Drought tolerant in some wheat genotypes to post-anthesis drought stress. *Journal of Agricultural Science*, *4*(11), 248–256.
Shiri, M., Aliyev, R.T., Choukan, R**. (**2010). Water stress effects on combining ability and gene action of yield and genetic properties of drought tolerance indices in maize. *Research Journal of Environmental Sciences*, *4*(1),75–84.
Sio-Se Mardeh, A., Ahmadi, A., Poustini, K., Mohammadi, V. (2006). Evaluation of drought resistance indices under various environmental conditions. *Field Crops Res*., *98*. 222–229.
Urechean, V., Bonea, D., Borleanu, I.C. (2010). The influence of climate on maize production in the centre of Oltenia. *Maize Genetics Cooperation Newsletter*, *84*. 14–15.
Urechean, V., Bonea, D. (2017). Estimate of drought tolerance at some maize hybrids grown in the central Oltenia zone with using stress tolerance indices. *17th International Multidisciplinary Scientific GeoConference SGEM, Conference Proceedings*, *17*(61), 681–688.
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Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

Bacillus amyloliquefaciens STRAINS WITH BIOCONTROL POTENTIAL AGAINST *Fusarium* spp. WHEAT PATHOGENS

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Abstract

Wheat contamination with fungi leads to quantitative losses and qualitative depreciation due to mycotoxins accumulation. Currently there are no Fusarium-resistant wheat cultivars. There are only medium tolerant varieties to such infections. In the present study, several Fusarium spp. pathogens were isolated from wheat debris and kernels harvested in 2017 from different lines of Triticum aestivum obtained in the experimental field of NARDI Fundulea. Fungal cultures were identified at genus level based on their microscopic and cultural characteristics on different nutrient media. Fungal growth inhibition of these pathogens was studied in vitro, using strains of Bacillus amyloliquefaciens (BIR, BPA, OS17 and BW) with biological control potential. The biocontrol efficacy, established by biometric measurements, was in the range of 61.9 to 84.4%. Further studies are required to determine the in vivo effects of these biocontrol bacteria on wheat plants.

***Key words**: biocontrol, Bacillus amyloliquefaciens, Fusarium sp., wheat.*

INTRODUCTION

Wheat contamination with *Fusarium* species leads to both quantitative loses and qualitative depreciation due to mycotoxin contamination. In Romania, the most common species of *Fusarium* found in wheat and other cereal grains are *F. graminearum*, *F. culmorum*, *F. roseum*, *F. avenaceum*, *F. poae, F. moniliforme* (sin *F. verticillioides*), *F. langsethiae* and *F. nivale* (BASF, 2012; 2019; Șopterean et al., 2012).
In wheat, fusariosis is more virulent on kernels and mature plants, but pathogens can also infect young plants or seeded kernels before emergence.
Currently, there are only tolerant or medium resistant wheat cultivars to fusariosis. Intensive growth of cereal crops, with poor variability in crop rotation, increase *Fusarium* infection pressure.
In special cases, such as plant breading fields and organic farming, where chemical pesticides are forbidden, the infection pressure is very high. Therefore, the only way to reduce pathogen attack is by applying some agrotechnical measures and natural plant protection products (PPPs), as an alternative to chemicals. The list of PPPs, approved by the European Commission, include several microorganisms with antifungal activity as active substances (http://ec.europa.eu).
Among these, *Bacillus amyloliquefaciens* can be used for biological control, either as single strain or as consortia.
Considering these, we focused our study on the characterization of *Fusarium* contaminants isolated from several wheat lines.
The biological control of these fungal pathogens was also analyzed, using four selected strains of *Bacillus*.

MATERIALS AND METHODS

Vegetal material

In order to analyze *Fusarium* contaminants of wheat we used severely infected kernels and spikelet's selected from a breeding field, placed in Călărași County, Romania. *Fusarium* infected samples are presented in Table 1. Samples were collected in 2017 and analysis during 2017 and 2018.

Table 1. *Fusarium* infected samples

Sample code	Vegetal material
E18A 2-4/ Sp4 / 2017 A	Kernels
E18A 2-4/ Sp4 / 2017 B	spikelets constituents
E18A 5-7/ Sp5 / 2017	Kernels
E25A F1-10/ Sp2 /2017 A	Kernels
E25A F1-10/ Sp2 /2017 B	spikelets constituents
E25A F1-10/ Sp3	Kernels
E28A F2-1/ Sp7+Sp8 P2 A	Kernels
E28A F2-1/ Sp7+Sp8 P2 B	spikelets constituents
E24A+F132 = R7S6	Kernels

Biocontrol bacteria

The biocontrol bacteria used in this study were previously isolated and identified as *Bacillus amyloliquefaciens* through Biolog GEN III system and 16S rDNA sequencing (Sicuia et al., 2016). These strains (BIR, BPA, OS17 and BW) were selected due to their antagonistic activity against different fungal pathogens, including *Fusarium* species (Sicuia, 2013; Grosu et al., 2014, 2015).

The Biolog Gen II technique for filamentous fungi was used to establish the biochemical fingerprint of the *Fusarium* isolates.

Fungal isolation

In order to isolate *Fusarium* spp. fungi from naturally infected wheat we use kernels and spikelet constituents presenting clear symptoms of fusariosis. The kernels used were shrivelled, wrinkled, matte, whitish or rosy shades, sometime covered with sporulated mycelia (Figure 1).

Figure 1. *Fusarium* infected wheat samples
a) kernels and
b) spikelet (arrow reveals the infection site)

From the whole wheat spikes were collected only spikelets with sporulated mycelia growing over its constituents (rachilla, glumes, palea and lemma).

The infected kernels and spikelet parts were inoculated on two semi-selective media for *Fusarium* isolation: Nash Snyder medium (also known as Peptone PCNB agar or PPA) and Malachite Green Agar (MGA) supplemented with antibiotics (50 ppm streptomycin and 100 ppm chloramphenicol) (Leslie & Summerell, 2006).

Typical growth of *Fusarium* was subcultured for several times on Potato-Dextrose-Agar (PDA) with antibiotics in order to assure pure cultures.

Fungal characterization

Fusarium cultures were macroscopically characterized when grown on PDA, Maltz extract agar (MA) and Potato-Sucrose-Agar (PSA). Some microscopic aspects were also studied on PDA cultures.

The Biolog Gen II technique for filamentous fungi was used to establish the biochemical fingerprint of some *Fusarium* isolates.

Mycotoxins analysis

The micotoxigenic potential of some fungal isolates was evaluated using TLC analysis (Ursan et al., 2018). Extraction was made from three weeks old *Fusarium* cultures grown on PSA medium.

Biocontrol activity

The biocontrol activity was analyzed by double culture technique. Bacterial strains were placed in spots at 2.5 cm distance from the fungal plugs inoculated in the center of the PDA plates. Incubation was carried out at 28°C. The antifungal efficacy was evaluated compared to the fungal growth obtained in the control plates (Islam et al., 2009).

RESULTS AND DISCUSSIONS

Nine *Fusarium* isolates were obtained from the naturally infected wheat, one from each sample subjected into the analysis.

During isolation better results were obtained on MGA semi-selective agar (Figure 2).

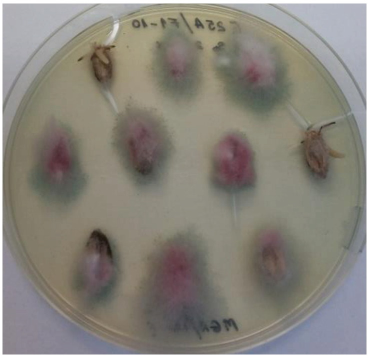

Figure 2. Wheat kernels covered in *Fusarium* growth after incubation on MGA semi-selective medium

MGA medium allowed typical growth of *Fusarium* spp., with less contaminants than PPA. Several other authors also recommend MGA as a better solution that PPA (Alborch, 2010; Bozac et al., 2014). Moreover, due to the fact that PCNB (pentachloronitrobenzene) is carcinogenic, it is less available.

Comparative analysis of the purified stains of *Fusarium* sp. grown on PDA, PSA and MA showed differences in pigment production depending on the growth substrate. Carbohydrate concentration and simple sugars positively affected pigment production in all strains. Red pigments were produced by all Fusarium isolates. Darker red color was obtained on PSA and PDA media, and light red pigmentation was obtained on MA medium (Figure 3).

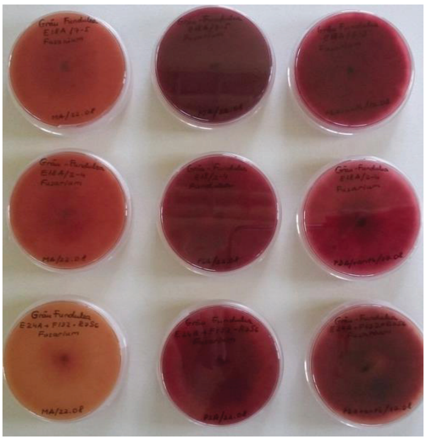

Figure 3. Red pigmentation of three *Fusarium* isolates grown on MA (left), PSA (middle) and PDA (right)

In comparison, in the two weeks old cultures, all *Fusarium* isolates presented the darkest red color on PSA medium, on PDA the pigments were faster developed compared to PSA medium (Figure 4).

Figure 4. *Fusarium* growth on MA (left), PSA (middle) and PDA (right) after two days of incubation

The highest mycelia growth rate was recorded on PDA medium, followed by PSA. On MA substrate, all fungal isolates had a slower growth rate, with less pigmentation (Figure 4).

The surface color of the cultures varied with strain, and depending on the growth substrate. Pinkish color (mostly on MA) to reddish mycelia (especially on PDA) were noticed, sometime with yellow to orange aerial hyphae on top of the basic red pattern (on PSA and PDA media) (Figure 5).

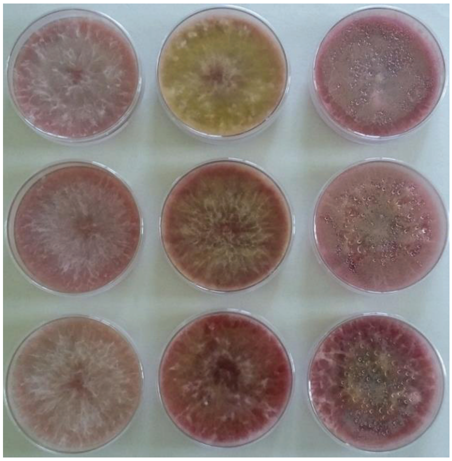

Figure 5. *Fusarium* cultures of E18A/7-5 (top line), E18A/2-4 (center) and E24A+F132=R7S6 (bottom line) grown on MA (left), PSA (middle) and PDA (right)

Studies regarding *Fusarium* pigments mention aurofusarin, neurosporaxanthin, and possibly rubrofusarin, as the most common pigments responsible for surface color in *F. graminearum* (Cambaza, 2018)*, F. culmorum, F. avenaceum*, *F. poae* and others, like: *F. acuminatum*, *F. crookwellens*, *F. pseudograminearum*, *F. sambucinum*, *F. sporotrichioides* and *F. tricinctum* (Frandsen et al., 2006).

Microscopic evaluation of the selected fungal isolates was performed only for PDA cultures. Analyzed under light microscopy (40×) all *Fusarium* isolates presented septate filaments, branched in acute angles, hyaline or pale pink colored. Older filaments presented rough surface cell walls (Figure 6a), and the young branches were colorless and smooth. Generally, the abundance of macroconidia was weak. However, in some cultures more than three weeks old, orange sporodochia were formed (Figure 6b), in which abundant macroconidia were present (Figure 6c). Microconidia were detected only in some of the strains. Chlamydospores were produced in both mycelia and macroconidia (Figure 6 d, e). In the mycelia, chlamydospores were found single or in chains.

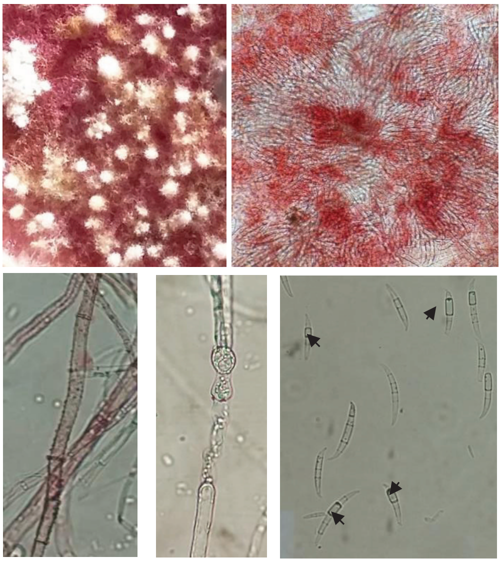

Figure 6. *Fusarium* culture characteristics: sporodochia macroscopic view (a) and microscopic aspects (b), rough filaments (c), chlamydospores within mycelia (d) and macroconidia (e) with chlamydospores (arrow)

Although the isolated fusaria were microscopically analyzed, the identification at specie level was not concluded. However, some of the strains are similar with *Fusarium graminearum*, *F. crookwellense*, *F. chlamydosporium* and *F. tricinctum.* Similar results were also revealed with the Biolog Gen II phenotypic assay for filamentous fungi identification (Figure 7).

Figure 7. Biochemical fingerprint of *Fusarium* sp. E18A/2-4 in Biolog Gen II microplates for filamentous fungi identification

Fusarium infected cereal grains are mostly contaminated with fumonisins and zearalenone (Kim & Vujanovic, 2016). Analyzing the mycotoxigenic potential of E18A/2-4, E18A/5-7 and E24A+F132 = R7S6, it was revealed that the isolated fungi are zearalenone producers (Figure 8).

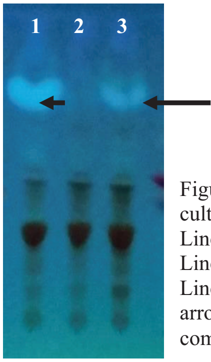

Figure 8. Mycotoxins in *Fusarium* cultures obtained on PSA medium: Line 1- E24A+F132 = R7S6, Line 2 - E18A/2-4 and Line 3 - E18A/5-7 extract, arrow indicates zearalenone compound

The most promising way to reduce mycotoxin contamination is to prevent plants from fungal infection, currently with plant protection products. Microbial based biopesticides such as *Bacillus amyloliquefaciens* can be used in biological control. In the European Union all active ingredients of chemical and biological pesticides must be approved by the European Commission and the commercial product must be authorized for release on the market (http://ec.europa.eu). Currently, in the EU there are four strains of *B. amyloliquefaciens* approved as active substance in pesticide formulations, QST 713, MBI 600, FZB24 and D747. Among these, *B. amyloliquefaciens* (former *B. subtilis*) QST 713 strain is the most common, being approved in 22 countries (BE, CY, CZ, DE, DK, EE, EL, ES, FI, FR, IE, IT, LT, LU, LV, NL, PL, PT, SE, SI, SK, UK).

Considering the biocontrol potential of *B. amyloliquefaciens* strains, we tested four Romanian native strains (BPA, BIR, OS17 and BW) for their antagonistic potential against the isolated *Fusarium* spp (Table 2).

Table 2. *B. amyloliquefaciens* efficacy in reducing *Fusarium* spp. growth

Fungal strain	Biocontrol strains			
	BPA	BIR	OS17	BW
E18A 2-4/ Sp4 / 2017 A	70.5%	70.5%	72.7%	68.2%
E18A 2-4/ Sp4 / 2017 B	74.5%	70.2%	72.5%	70.2%
E18A 5-7/ Sp5 / 2017	75%	71.6%	72.7%	72.7%
E25A F1-10/ Sp2 /2017 A	84.4%	77.8%	77.8%	75.6%
E25A F1-10/ Sp2 /2017 B	66.7%	61.9%	61.9%	61.9%
E25A F1-10/ Sp3	72.5%	67.5%	75%	70%
E28A F2-1/ Sp7+Sp8 P2 A	80.5%	75.6%	73.2%	75.6%
E28A F2-1/ Sp7+Sp8 P2 B	83.3%	83.3%	83.3%	83.3%

Among the tested strains BPA revealed the highest potential in reducing *Fusarium* spp. growth. During *in vitro* trials a synergic effect was noticed between BPA and OS17 strains in reducing fungal growth (Figure 9).

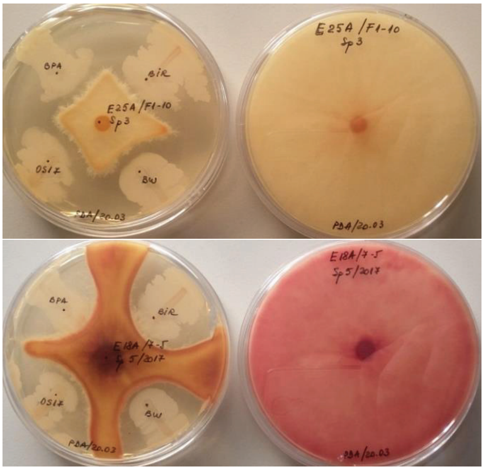

Figure 9. Antifungal activity of *B. amyloliquefaciens* against *Fusarium* spp.

These bacterial strains presented a wide spectrum of antifungal activity. Previous studies mentioned these strains as promising biocontrol agents against mycotoxigenic *Fusarium graminearum* and *F. culmorum* (Grosu et al., 2014), and several other fungal pathogens, like: *Alternaria* spp., *Botrytis cinerea*, *Fusarium oxysporum*, *F. solani*, *Macrophomina phaseolina*, *Penicillium* spp., *Rhizoctonia solani*, *Sclerotinia sclerotiorum* (Dinu et al., 2012; Sicuia et al., 2011; 2012a, b; Grosu et al., 2014; Sicuia et al., 2013).

CONCLUSIONS

Wheat samples (plant debris and grains) severely contaminated with *Fusarium* were purchased from a breeding field, in Călărași County, Romania.

New strains of phytopatogenic fungi belonging to *Fusarium* spp. were detected and characterized. Among the tested *Fusarium* semi-selective media, MGA was found to be more appropriate than PPA. The isolated *Fusarium* spp. were highly virulent, and some proved to be mycotoxin producers.

Biological control methods can be used in order to reduce the fungal growth of such pathogens. Four Romanian native strains of *Bacillus amyloliquefaciens* (BPA, BIR, OS17 and BW) prove to have good inhibitory activity against the newly isolated fusaria, with an efficacy of 61.9 to 84.4%.

REFERENCES

BASF, 2012. Cereale. Tehnologia BASF de protecție pentru o cultură de succes. www.agro.basf.ro.

BASF, 2019. Catalog 2019. www.agro.basf.ro.

Cambaza, E. (2018). Comprehensive Description of *Fusarium graminearum*. Pigments and Related Compounds. *Foods (Basel, Switzerland)*, *7*(10), 165.

Dinu, S., Sicuia, O.A., Constantinescu, F., Lazureanu, C., Oancea, F. (2012). Efficacy of some bacterial bioproducts against *Sclerotium cepivorum*. *Scientific Papers, Series B Horticulture, 56*. 81–88.

Frandsen, R.J.N., Nielsen, N.J., Maolanon, N., Sørensen, J.C., Olsson, S., Nielsen, J., Giese, H. (2006). The biosynthetic pathway for aurofusarin in *Fusarium graminearum* reveals a close link between the naphthoquinones and naphthopyrones. *Mol. Microbiol., 61*. 1069–1080.

Grosu, I.A., Sicuia, O.A., Dobre, A., Voaides, C., Cornea, C.P. (2015). Evaluation of some *Bacillus spp.* strains for the biocontrol of *Fusarium graminearum* and *F. culmorum* in wheat. *Agriculture and Agricultural Science PROCEDIA, 6*. 559–566.

Grosu, I., Israel-Roming, F., Sicuia, O.A., Constantinescu, F., Popa, G., Cornea, C.P. (2014). Effect of some bacterial antagonists on growth and mycotoxin production of *Fusarium graminearum* and

F. culmorum isolates. *Scientific Bulletin. Series F. Biotechnologies, 18*. 26–31.

http://ec.europa.eu/assets/sante/food/plants/pesticides/lop/index.html#inline_content

http://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/public/?event=activesubstance.selection&language=EN

Islam, R., Jeong, Y.T., Ryu, Y.J., Song, C.H., Lee, Y.S. (2009). Isolation, identification and optimal culture conditions of *Streptomyces albidoflavus* C247 producing antifungal agents against *Rhizoctonia solani* AG2-2. *Mycobiology. Springer. Stuttgart. 37*(2), 114–120.

Kim, S.H., Vujanovic, V. (2016). Relationship between mycoparasites lifestyles and biocontrol behaviors against *Fusarium* spp. and mycotoxins production. *Applied Microbiology and Biotechnology, 100*(12), 5257– 5272.

Leslie, J.F., Summerell, B.A. (2006). *The Fusarium Laboratory Manual.* Blackwell Publishing, Oxford, UK

Sicuia, O.A., Constantinescu, F., Dinu, S. (2012). Bacterial biocontrol strains that reduce *Rhizoctonia* damping off in tomato seedlings. *Scientific Papers, Series B Horticulture, 56*. 175–180.

Sicuia, O.A., Oancea, F., Cornea, C.P. (2012). New screening methods for evaluation of *Fusarium sporulation* inhibition by *Bacillus* biocontrol strains. *Scientific Papers, Series F. Biotechnologies, 16*. 67–72.

Sicuia, O.A., Olteanu, V., Ciuca, M., Cirstea, D.M., Cornea, C.P. (2011). Characterization of new *Bacillus* spp. isolates for antifungal properties and biosynthesis of lipopeptides. *Scientific Papers, Series A. Agronomy, 54*. 482–491.

Sicuia, O.A. (2013). Cercetări la nivel molecular şi celular privind mecanisme de interacţiune între microorganisme din rizosferă şi plante de interes economic. PhD Thesis. University of Bucharest.

Sicuia, O. A., Poli, A., Constantinescu, F., Cornea, C.P., Spadaro, D. (2016). Molecular differentiation of plant beneficial *Bacillus* strains useful as soil agro-inoculants. *Proceedings of the III International Symposium on Organic Greenhouse Horticulture ISHS Acta Horticulturae, 1164*. 257–264

Şopterean, M.L., Suciu, A.L., Nagy, E., Haş, V., Haş, I., Puia, C. (2012). The influence of the climatic conditions on *Fusarium* spp. attack on corn hybrids tested in natural and artificial infection conditions at A.R.D.S Turda in the period of 2009-2011. *Research Journal of Agricultural Science*, 44.

Ursan, M., Boiu-Sicuia, O.A., Voaides, C., Stan V., Bubueanu, C., Cornea, C.P. (2018). The potential of new streptomyces isolates as biocontrol agents against *Fusarium* spp. *Agriculture for Life, Life for Agriculture, Conference Proceedings, 1*(1), 594–600.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SEED VIGOUR INDEX ESTIMATION OF SOME ROMANIAN WINTER BARLEY BREEDING LINES

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Abstract

*A uniform emergence of barley plants in the field depends on their seed vigour potential and this index define the seed quality. The aim of this paper is to assess the germinative capacity and elements of seed vigour of some Romanian winter barley lines, estimated by two methods used at NARDI Fundulea. Thirty-five winter barley lines were tested under controlled conditions for different physiological traits to determine seed vigour index. The obtained results showed that at the second level of temperature (6°C), the length of shoot and seedling is significantly correlated with germination faculty (0.44**) and also the length of root and shoot with length of seedling (0.44** and 0.95***). The seed index vigour varied from 13.4 to 29.2 when was evaluated at 25°C and from 7.2 to 26.1 at 6°C. An increase tendency of root, shoot and seedling length at 6°C was noticed in average. The different level of temperature could be used for screening the winter barley lines developed in barley breeding program in order to promote genotypes which assure a fast and uniform emergence.*

Key words: *germination, seed vigour index, winter barley lines.*

INTRODUCTION

The success of the winter barley emergence in the field depends first of all by seed vigour. This seed quality trait determines a fast and uniform emergence of plants but in the same time is influenced by environmental conditions as soil temperature and soil moisture (Perry, 1973).
Winter barley breeding efficiency can increase by early selection of new obtained biological material (barley lines in different generations). During field selection are noticed many barley genotypes and a reduction of biological material volume can be possible by seed vigour early selection criteria.
These genotypes can be studied by different methods in order to characterize seed vigour, in laboratory and under many conditions in the field.
In the latest, climate changes (particularly high oscillating of temperature) have an increased influence on behaviour of winter barley, especially when this crop is sown after optimal period.
In Romania, cultivated area with winter barley varieties, is usually sown in September when is warm (20-25°C), before the recommended period or at the beginning of November (when the temperature usually decreases). In order to predict field barley emergence could be use different types of test for malting and naked barley (Kim et al., 1989) as follow: seed aging treatment, warm germination test, cold germination test, tetrazolium vigour test, conductivity test, field test and also percent of germination, length of seedling and seed vigour index (Kim et al., 1994). For example, in the case of rice, germination and field emergence, were identified as the main contributors to seed yield per plant (Adebisi et al., 2010). Recently, a succesful fast estimation of seed vigour was made by using the absolute seed protein content for wheat and maize, tested under different conditions. The results of this research showed

that, both in wheat and maize, can be use to estimation of seed vigour (Wen et al., 2018).

MATERIALS AND METHODS

Thirty-five winter barley lines developed at NARDI Fundulea (21 lines in F_8, 8 lines in F_7 and 6 lines in F_6 generation) and one specific winter barley variety were used as check (Simbol variety), to evaluate the behaviour under different temperature regarding the germination and the component of seed vigour. We adopt a standard treatment suggested by the Association of Official Seed Analysts for maize, namely Cold test method at 6°C and beside that, was used a warm germination test conducted at 25°C (Stan et al., 2008), under laboratory condition in order to simulate field conditions during optimal sowing (October) and later (in the first decade of November).

The principle of the warm method is to determine the germination faculty in optimal conditions of temperature and humidity in a well-established time.

The seeds are placed on rolls of industrial filter paper moistened with 60% water of its retaining capacity in four repetitions of one hundred seeds. Rolls are placed in the germination chamber at a temperature of 25°C for seven days. Determination of germination is performed at 4 and 7 days, respectively, according to internal rules and the evaluation of the germs is done after the ISTA evaluation manual (2006). Normal seedlings were counted and the obtained results are expressed in percent.

The principle of the Cold test method is to create in the laboratory, similar conditions to those in the soil, conditions that allow the microflora of the soil and seeds to participate in a competition from which those seeds, germs, or those individuals lacking sufficient resistance either due to heredity or to some physical, physiological or biochemical damage.

The cold test method (6°C) applied to study winter barley genotype is genuine, being improved at NARDI Fundulea, namely Fundulea Test Seed (FTS) by the Biology, pathology and seed quality control laboratory (Stan et al., 2016).

According to this method, the seed is placed in a 1/1 soil/sand mix, wetted with 60% water of its retaining capacity, in four repetitions of 100 seeds. The germination temperature is 6°C for seven days and after that, the seed is transferred to the growth chamber at a temperature of 25°C for four days.

The evaluation of the germs is carried out after the 11 days, based on international standards for ISTA-2006 seed testing. Length of root and shoot were determined separately by a ruler and after that in order to calculate the seed vigour index, these were summed and expressed as seedling length (Mishra et al., 2017).

Seedling vigour index was calculated as the product between percent of germination (G) and the seedling length (SL) in centimetres, with Abdul-Baki and Anderson formula (1973): SVI = G (%) x SL (cm) /100.

As check was used the winter barley variety Simbol due its high germination under low temperature (G=100% tested at 6°C). The obtained results were the subject of ANOVA, correlation analysis and expressed as minimum, mean and maxim values. The objectives of this study were to investigate the behaviour of seed vigour elements using 2 methods in order to detect the influence of temperature level and identify the barley genotypes with a superior resistance to one of the abiotic stress (low temperature during germination) and if the variation of this could help to perform early selection on winter barley lines.

RESULTS AND DISCUSSIONS

Analysis of variance showed a significant influence of temperature level, genotype and their interaction on germination, root and shoot length (Table 1). Also, as source of variation, the temperature level had a significant influence on length of seedling, Seed Index Vigour and on the interaction between these.

Table 1. ANOVA for germination, root length, shoot length, length of seedling and Seed Index Vigour

Source of variation	Germination		Root length		Shoot length		Length of seedling		Seed Index Vigour	
	F	P-value	F	P-value	F	P-value	F	P-value	F	P-value
Temperature	223.59**	8.2E-31	31.06**	1.2E-07	6.23*	0.014	42.11**	1.30E-09	104.05**	1.00E-18
Genotype	6.84**	1.0E-16	5.56**	1.6E-13	10.77**	4.9E-25	8.49**	5.37E-21	10.74**	1.22E-25
T x G	5.99**	1.3E-14	3.15**	1.1E-06	39.82**	2.1E-56	16.04**	1.67E-34	21.68**	7.81E-42

*significant at a probability level of p<0.05; **significant at a probability level of p<0.01.

The registered minimum value of germination (Figure 1) was 85.0% under 25°C while under 6°C, decreased at 33.0%, the percent of decreasing was very high (67%). This means that the temperature influenced negatively this parameter on many winter barley genotypes.

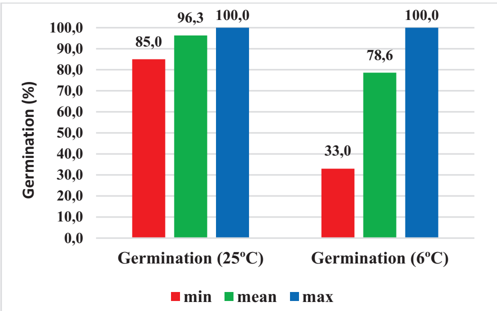

Figure 1. The minimum, mean and maximum value of germination registered under 25°C and 6°C

In Romania, the minimum accepted value for winter barley seed used for sowing, has to be 85.0%. Any variety with germination under this standard is not recommended for sowing and also the price per kg decreased at half (the seeds are used for animal feed).

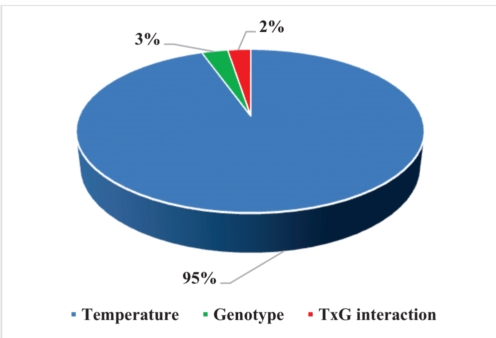

Figure 2. The influence of temperature, genotype and their interaction on germination

The temperature has the biggest influence on the level of germination (95%) while the influence of genotype and the interaction between temperature and genotype is 3% and 2% respectively (Figure 2).

The root length varied between 5.8 and 11.2 cm for first condition and from 7.5 to 10.8 cm in the second condition. We noticed that the minimum value of this parameter had a slightly increasing (1.7 cm), comparing with the registered value under 25°C (Figure 3).

In the case of shoot length, the trend was similar (Figure 3), the minimum value had increased from 4.4 cm (at 25°C) to 9.3 cm (6°C) and the difference between minimum and maximum value was much higher under first condition (14.5 cm) than those between the second condition (8.5 cm).

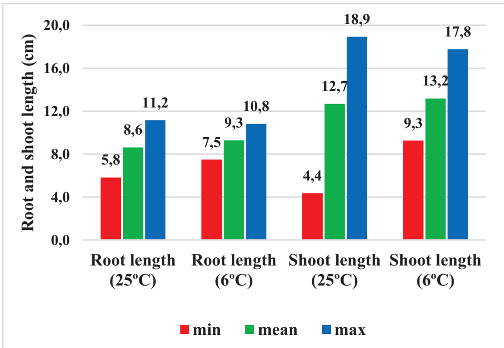

Figure 3. The minimum, mean and maximum value of root and shoot registered under 25°C and 6°C

The temperature, genotype and their interaction were different regarding the root and shoot length (Figures 4 and 5). In the case of root length, the genotype had a higher influence (14%) than the previous parameter (germination) and the influence of temperature still remain important (78%).

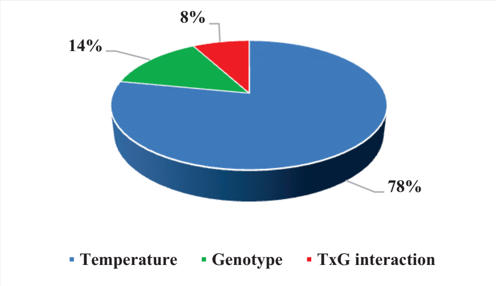

Figure 4. The influence of temperature, genotype and their interaction on root length

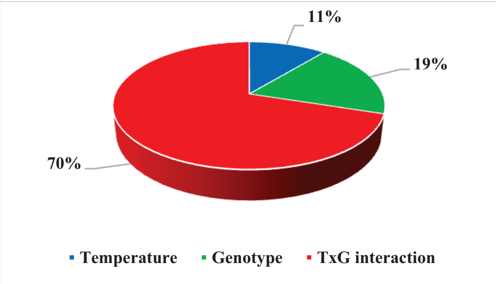

Figure 5. The influence of temperature, genotype and their interaction on shoot length

The shoot length was influenced by the interaction between temperature and genotype (70%) and the contribution of genotype increase at 19%.

The length of seedling oscillated between 13.6 cm and 30.1 cm at first level of tested temperature and from 18.1 cm to 27.8 cm at second level. Also, was noticed a length seedling increasing when the level of tested temperature decreased (Figure 6).

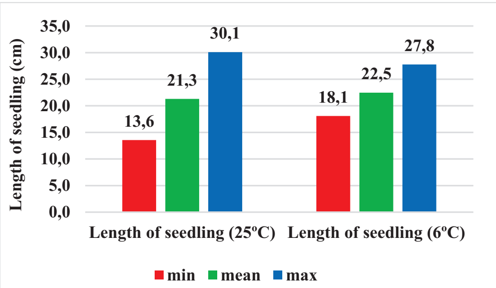

Figure 6. The minimum, mean and maximum value of length of seedling registered under 25°C and 6°C

In Figure 7, it is observed that the level of temperature contribution and studied factors interaction is 63% and 13% respectively.

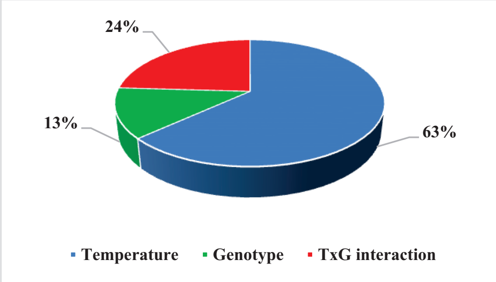

Figure 7. The influence of temperature, genotype and their interaction on length of seedling

The Seed Vigour Index ranged from minimum 13.4 to 29.2 in the first conditions and from 7.2 to 26.1 in the second condition (Figure 8). The analyse of Seed Vigour Index minimum value showed that when the temperature decreased at 6°C, this affected the percent of germination much more for some genotypes than others, resulting a high influence on seed vigour, especially on shoot length (the length is bigger than under the 25°C). According to the level of percent of germination, it is clear that influence of temperature on Seed Index Vigour is high (76%) but the interaction between factors as source of variation, had 16% contribution (Figure 9), higher than the genotype (8%).

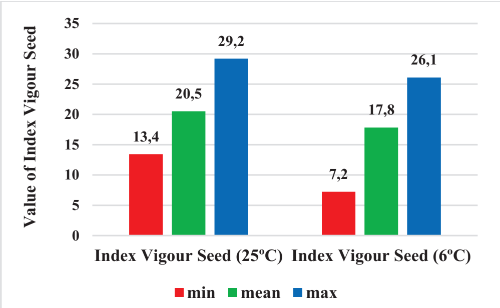

Figure 8. The minimum, mean and maximum value of Index Vigour Seed registered under 25°C and 6°C

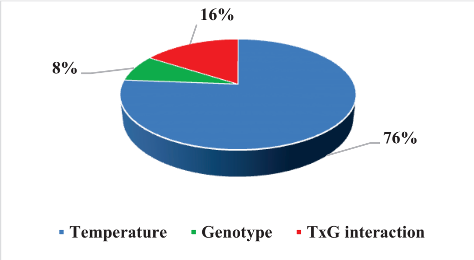

Figure 9. The influence of temperature, genotype and their interaction on Seed Index Vigour

The value of germination (Table 2) under warm condition, ranged from 85 to 100% (Simbol variety had 93%, the mean value was 96.28%) and the line L 383-1 (purple bolded colour), was near germination value limit (85%). Under cold condition ranged from 33 to 97% (Simbol variety had 100% and the mean value was 78.61%) and seventeen genotypes had a germination very low (between 33 and 83%).

Table 2. Experimental data for germination, root and shoot length, length of seedling and Seed Index Vigour

No.	Name of genotype	G (%) 25ºC	G (%) 6ºC	RL (cm) 25ºC	RL (cm) 6ºC	SL (cm) 25ºC	SL (cm) 6ºC	LS (cm) 25ºC	LS (cm) 6ºC	SIV 25ºC	SIV 6ºC
1	**Simbol**	93	100	9.88	9.30	**5.55**	**13.73**	15.43	23.03	**14.5**	**23.0**
2	**L19-10**	97	93	11.14	9.73	11.54	15.19	22.68	24.92	**22.0**	**23.2**
3	**L20-10**	93	96	10.44	9.01	13.37	15.47	23.81	24.48	**22.1**	**23.5**
4	**L9-12**	99	92	9.85	9.73	10.79	15.42	20.64	25.15	**20.4**	**23.1**
5	**L2-12**	95	96	9.32	9.26	12.85	14.97	22.17	24.23	**21.1**	**23.3**
6	**L3-01**	99	97	6.89	9.00	12.63	13.28	19.52	22.28	**19.3**	**21.6**
7	**L1-12**	100	88	7.98	8.73	10.88	13.39	18.86	22.12	**18.9**	**19.5**
8	**L3-12**	91	96	5.82	9.36	11.50	13.70	17.32	23.06	**15.8**	**22.1**
9	**L4-12**	99	97	6.35	10.23	11.70	14.24	18.05	24.47	**17.9**	**23.7**
10	**L5-12**	97	88	6.98	8.36	11.43	12.50	18.41	20.86	**17.9**	**18.4**
11	**L6-12**	97	84	8.52	8.40	11.11	13.81	19.63	22.21	**19.0**	**18.7**
12	**L7-12**	99	93	9.21	8.31	**4.36**	**13.31**	13.57	21.62	**13.4**	**20.1**
13	**L8-12**	100	95	8.36	9.68	11.50	12.75	19.86	22.43	**19.9**	**21.3**
14	**L10-12**	97	89	6.48	9.41	11.62	16.24	18.10	25.65	**17.6**	**22.8**
15	**L11-12**	97	91	8.72	9.34	12.52	14.78	21.24	24.12	**20.6**	**21.9**
16	**L2-13**	97	33	7.04	9.30	12.28	14.11	19.32	23.41	**18.7**	**7.7**
17	**L5-13**	99	41	7.45	8.94	12.06	15.26	19.51	24.20	**19.3**	**9.9**
18	**L6-94**	95	63	9.29	9.32	11.62	15.54	20.91	24.86	**19.9**	**15.7**
19	**L15-15**	99	96	8.85	10.65	12.78	16.52	21.63	27.17	**21.4**	**26.1**
20	**L101-12**	95	96	9.03	9.14	11.24	15.73	20.27	24.87	**19.3**	**23.9**
21	**L333-36**	96	93	6.37	9.99	**8.58**	**17.78**	14.95	27.77	**14.4**	**25.8**
22	**L383-1**	85	64	7.48	9.34	10.43	12.98	17.91	22.32	**15.2**	**14.3**
23	**L201-1**	93	59	7.71	9.15	14.15	9.26	21.86	18.41	**20.3**	**10.9**
24	**L201-2**	93	80	9.29	9.94	17.40	11.51	26.69	21.45	**24.8**	**17.2**
25	**L201-3**	95	36	8.89	9.27	14.69	10.81	23.58	20.08	**22.4**	**7.2**
26	**L201-4**	93	63	10.09	10.13	14.47	10.32	24.56	20.45	**22.8**	**12.9**
27	**L201-5**	97	63	9.96	9.48	14.93	10.81	24.89	20.29	**24.1**	**12.8**
28	**L202-6**	99	72	8.59	9.97	12.18	11.04	20.77	21.01	**20.6**	**15.1**
29	**L600-1**	97	65	8.23	9.27	15.25	12.14	23.48	21.41	**22.8**	**13.9**
30	**L600-4**	99	83	10.46	10.07	16.22	10.26	26.68	20.33	**26.4**	**16.9**
31	**L600-6**	99	80	11.04	9.36	16.28	11.53	27.32	20.89	**27.0**	**16.7**
32	**L600-7**	93	67	9.95	7.87	13.48	11.98	23.43	19.85	**21.8**	**13.3**
33	**L600-9**	100	77	8.25	9.02	16.80	11.34	25.05	20.36	**25.1**	**15.7**
34	**L600-10**	97	64	11.17	10.82	18.93	10.71	30.10	21.53	**29.2**	**13.8**
35	**L600-13**	97	57	8.57	7.49	14.08	10.59	22.65	18.08	**22.0**	**10.3**
36	**L600-14**	95	83	6.89	7.85	15.30	11.47	22.19	19.32	**21.1**	**16.0**
Mean		**96.28**	**78.61**	**8.63**	**9.28**	**12.68**	**13.18**	**21.31**	**22.46**	**20.52**	**17.84**
LSD (5%)		**8.67**		**1.05**		**0.80**		**1.42**		**1.86**	

G (%) – germination; RL (cm) – root length; SL (cm) – shoot length; LS (cm) – length of seedling; SVI – Seed Index Vigour

Table 3. Correlations between germination, root and shoot length, length of seedling and Seed Index Vigour

Parameters	**G 25ºC**	**G 6ºC**	**RL 25ºC**	**RL 6ºC**	**SL 25ºC**	**SL 6ºC**	**LS 25ºC**	**L S6ºC**	**SIV 25ºC**	**SIV 6ºC**
G 25ºC	1									
G 6ºC	0.18	1								
RL 25ºC	0.08	-0.05	1							
RL 6ºC	0.08	0.11	0.21	1						
SL 25ºC	-0.02	**-0.33^{o}**	**0.36***	0.21	1					
SL 6ºC	0.07	**0.44****	-0.24	0.14	**-0.59^{ooo}**	1				
LS 25ºC	0.02	-0.27	**0.70*****	0.25	**0.92*****	**-0.55^{ooo}**	1			
LS 6ºC	0.09	**0.44****	-0.15	**0.44****	**-0.47^{ooo}**	**0.95*****	**-0.42^{oo}**	1		
SIV 25ºC	0.20	-0.23	**0.69*****	0.27	**0.90*****	**-0.53^{ooo}**	**0.98*****	**-0.40^{o}**	1	
SIV 6ºC	0.17	**0.95*****	-0.08	0.25	**-0.42^{oo}**	**0.68*****	**-0.36^{o}**	**0.70*****	-0.32	1

G (%) – germination; RL (cm) – root length; SL (cm) – shoot length; LS (cm) –length of seedling; SIV – Seed Index Vigour

The mean value of the root, shoot and seedling length was higher at 6°C comparing 25°C. We noticed an increase tendency of root, shoot and seedling length with 1.65, 0.50 and 1.35 units respectively. The value of Seed Index Vigour was in average higher under warm condition, difference between the obtained values being 2.68.

It has to be mentioned that some genotypes had a similar behaviour (black bolded colour) under both testing conditions, some of them registered a higher seed index vigour at 6°C than under 25°C (green bolded colour) and several had a lower SIV under the second condition due to a very low germination (red bolded colour).

From the total of tested genotypes, 11 winter barley lines (green bolded colour) had presented a higher seed index vigour under cold condition (similar to check) due to a better shoot developed. Two winter barley lines were remarked due a lowest shoot length value (4.36 and 8.58 cm, blue bolded values) under warm condition, one of them having a higher value (17.78 cm) than check (13.73 cm), when the temperature was low.

The obtained results of correlation coefficient are presented in Table 3, where the germination at 25°C is not correlated with any of seed studied vigour elements, while the germination at 6°C showed distinct significant positive correlation with shoot length, length of seedling and very significant with Seed Index Vigour (0.95***) value at 6°C. A low temperature influenced negatively the shoot length (-0.33°). A higher temperature (25°C) can positively influence the root length in relationship with shoot length, length of seedling and Seed Index Vigour, while at low temperature (6°C), only the root length is positively influenced. The shoot length is differently influenced by the level of temperature, being a strong correlation between this vigour seed element, length of seedling and respectively Seed Index Vigour (0.92*** and 0.90***) at 25°C. Almost the same correlations was at 6°C between these elements (0.98*** and 0.68***), which showed a higher contribution at length of seedling and also to Seed Index Vigour. The length of seedling had a significant role in the Seed Index Vigour final value, considering the strong positively correlation with this.

CONCLUSIONS

Evaluation and analysis of winter barley breeding lines showed that germination (G) and root length (RL) are mainly temperature dependent and shoot length (SL) is highly dependent by the TxG interaction.

Increase seed index vigour (SIV) means an improved seedling response to different temperatures and therefore fast emergence and increased yield. The genotype has a higher influence in developing of root (14%) and shoot (19%), but seed index vigour is influenced by the temperature during the germination (76%).

Based on the described behaviour in the mentioned conditions, it can know that a winter barley variety have a fast emergence under warm or cold conditions.

These results showed that implementation and using these methods in early generation study for seed vigour (especially cold test method) may be very usefully for barley breeder work selection and therefore to breed improved winter barley varieties which performed better under future Romania climate changes.

REFERENCES

Abdul-Baki, A.A., and Anderson, J.D. (1973). Vigour determination in soybean seed by multiple criteria. *Crop Science, 13*, 630–633.

Adebisi, M.A., Okelola, F.S., Alake, C.O., Ayo-Vaughan, M.A., Ajala, M.O. (2010). Interrelationship between seed vigour traits and field performance in new rice for Africa (Nerica) genotypes (*Oryza sativa* L.). *Journal of Agricultural Science and Environment*, *10*(2), 15–24.

International Seed Testing Association. (2006). *ISTA Handbook on Seedling Evaluation*, second ed. International Seed Testing Association, Zurich, Switzerland.

Kim, S.H., Bin, Y.H., Choe, Z.R. (1989). The use of multiple seed vigour indices to predict field emergence and grain yield of naked and malting barley. *Korean Journal Crop Science*, *34*(2), 134–141.

Kim, S.H., Choe, Z.R., Kang, J.H., Copeland, L.O., Elias, S.G. (1994). Multiple seed vigour indices to predict field emergence and performance of barley. *Seed Science and Technology*, *22*, 29–68.

Mishra, A., Kumar, V., Sharma, A.D., Srinivasan, K. (2017). Evaluation of Seed Quality of Barley Varieties through Controlled Deterioration Test. *International Journal of Current Microbiology and Applied Sciences, 6*(6), 3123–3131. ISSN: 2319-7706 .

Perry, D.A. (1973). Interacting effects of seed vigour and environment on seedling establishment. *In: W. Heydecker, Ed., Seed Ecology, Penn. State Univ. Press*, 311–324. University Park,

Stan, O., Petcu, E., Antohe, I., Stan, C. (2008). Caracterizarea vigorii unor hibrizi de porumb prin metoda Cold test 6°C. *Analele I.N.C.D.A. Fundulea*, *LXXVI*, 43–52.

Stan, O., Martura, T., Partal, E., Iordan, H. (2016). Estimarea însușirilor de calitate și vigoare la sămânța noilor genotipuri de porumb, prin metoda coldtest și deteriorare controlată. *Analele I.N.C.D.A. Fundulea*, *LXXXIV*. 141–156.

Wen, D., Hou, H., Meng, A., Meng, J., Xie, L., Zhang, C. (2018). Rapid evaluation of seed vigour by the absolute content of protein in seed within the same crop. *Scientific Reports*, *8*, 55–69.

MISCELLANEOUS

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

PRELIMINARY RESULTS: ASSESSMENT OF NEW SALVIA CHEMOTYPES FOR HERBAL TEA INDUSTRY BY HYBRIDIZATION

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Abstract

Common sage (S. officinalis L.) and Anatolian sage (S. fruticosa Mill.) are the most used sage species in herbal tea industry. Besides that, Antioch sage (S. aramiensis Rech. fill.) naturally presents only in Hatay province of Turkey and used as herbal tea in the region. The risk of sage usage is toxicity from camphor and thujone content. S. officinalis known as its high content of thujone and camphor. In contrast S. fruticosa and S. aramiensis known as their high content of eucalyptol. Both S. officinalis and S. fruticosa has high essential oil content rather than S. aramiensis. The aim was to develop new chemotypes (low camphor and low thujone) by interspecific hybridization for health. Species were reciprocal crossed except for S. officinalis. It was used just as mother for all crosses due to male sterility. Although the chromosome numbers of the species were the same, general crossing ability was very low. Essential oil contents of the hybrids were ranged 0.75-4.21%. Camphor, thujone and eucalyptol contents were evaluated in the crossed population.

Key words*: hybrids, thujone, camphor, sage.*

INTRODUCTION

One of the most important members of the Lamiaceae family, the genus Salvia, with close to 1000 species, is widely distributed in eco-systems and used for different purposes (food, perfumery, ornamental plant, medicine etc.) (Kintzios, 2000; Walker & Sytsma, 2007; Kahraman et al., 2009). In Turkey, the *Salvia* genus has 97 species, 4 subspecies and 8 varieties, of which 55 are endemic (Hedge, 1982; Davis et al., 1988; İpek & Gürbüz, 2010).

S. officinalis L. is the most commonly used herbal tea and spice among the sage species. 1.8 cineole, camphor, thujone, pinen and borneol are the most common components in the essential oil of *Salvia* species used as herbal tea and spices (Lamaison et al., 1991; Cuvelier et al., 1994; Lawrence, 1998). Several studies conducted on *S. officinalis* showed that the main component of the essential oil is thujon and the ratio of 1.8 cineole is lower. In contrast, *S. fruticosa* essential oil has been shown to be rich in 1.8 cineole content and low in thujone content (Bazina et al., 2008; Herraiz-Penalver et al., 2015; Ayanoğlu et al., 2012; Bayram, 1998; Cvetkovikj et al. 2015; Baydar, 2005). *S. aramiensis* Rech. fil., used as herbal tea, is a species with low camphor ratio, little or no thujone. It grows only in Hatay in Turkey. Studies have shown that the camphor rate of *S. aramiensis* essential oil is between 1.0-1.5% and the rate of thujone varies between 0-1.1% (Demirci et al., 2002; Karaman et al., 2007; Aşkun et al., 2010; Kelen & Tepe, 2008; Ayanoğlu et al., 2012; Ertaş et al., 2017).

Camphor and thujone are the most important factors limiting their usage as herbal tea and spices, although each of them has many usages in the field of medicine and cosmetics. Sage can be added to foodstuffs if the thujone content in the final product does not exceed 0.5 mg/kg (Böszörmenyi et al., 2009). In food industry, *Salvia* chemotypes containing low thujone should be preferred (Bielenberg, 2007; Pelkonen et al., 2013). Thujone consumption in *S. officinalis* essential oil should not exceed 5 mg per day and is not recommended for those under 18 years of age (European Medicines Agency, 2010). Both thujone and camphor are known to be very toxic when used for long-term treatment (Millet et al. 1981, De Vincenzi & Mancini, 1997). In particular, even when taken in small amounts, camphor can cause serious or fatal consequences for young children (Liebelt & Shannon, 1993; Theis & Koren, 1995; Shahabi et al., 2012).

Salvia species are divided into various groups according to the similarity of species to each other. *S. officinalis*, *S. fruticosa* and *S. aramiensis* are in the same group (*Salvia*). *S. officinalis* and *S. fruticosa* species have 2n = 14 chromosomes (Hedge, 1982; Doğan et al., 2008). The chromosome number of *S. aramiensis* was determined as 2n = 14 in our study. *S. officinalis*, *S. fruticosa* and *S. aramiensis* are cross pollinated species moreover natural hybridization between *S. officinalis* and *S. fruticosa* has been recorded (Herraiz-Penalver, 2015; Radosavljevic et al., 2019). Artificial interspecific crossing of *Salvia* genotypes also studied but most of them were floriculture except one *S. officinalis* and *S. fruticosa* crossing research (Haque & Goshal, 1981; Putievsky et al., 1990; Tychonievich & Warner, 2011).

A very large variation could obtained in the first generation (F1) in cross pollination between the species. This increases the probability of genotypes having the desired properties (Macukanovic-Jocic et al., 2011; Subaşı & Güvensen, 2011). The results of the studies conducted in which the genetic structure of *Salvia* genotypes associated with essential oil components suggest that genetic factors may be more effective than environmental factors on essential oil components (Skoula et al., 1999; Bazina et al., 2008; Böszörmenyi et al., 2009).

The aim of the study was to reveal the variation among hybrids from three species in the sight of essential oil composition especially eucalyptol, thujone and camphor.

MATERIALS AND METHODS

In the study, parent plants from *S. officinalis*, *S. fruticosa* and *S. aramiensis* were propagated by stem cuttings from Mustafa Kemal University Plant Collection Garden. Species were both used as mother and father plant except for *S. officinalis*. *S. officinalis* flowers were male sterile. *S. fruticosa* plants started flowering followed by *S. officinalis* and the last one was *S. aramiensis*. Pollens of the species were collected until the flowering period of mother species. Crosses were made by emasculation and hand pollination. In the emasculation, process anthers were removed before inflorescence and blooming (opened) flowers were removed. Petals were removed to facilitate the pollination. The flowers were pollinated once and bagged with net covers until harvest for seeds.

After seeds set, approximately 6 weeks after hand pollination, flower branches were harvested further stages. Before sowing seeds soaked in 300 ppm GA3 solution for 24 hour to improve germination. Later seeds were sown in petri dishes. Germination starts in 7 days and continues 4 weeks, after 4 weeks seeds were washed with water and treated replicated. Germinated seeds were planted in plastic viols and plants were placed in green house. The hybrids were placed in pots after extend length almost 20 cm. In summer time pots were moved outside of the green house. Planting material was peat and perlite mix. The hybrids were harvested to obtain their essential oil ratio. For each hybrid, dry leaf samples were weighed and water distilled for 3 hours with Clevenger apparatus. Essential oil ratios are the mean value from dry plant material weight and expressed in g/100 g dry weight percentage.

Essential oils were kept amber vial at +4 °C until analysis. Essential oil components were analysed with GC-MS (Gas Chromatography-Mass Spectrometry) device Thermo Scientific ISQ Single Quadrupole. 5 µl of essential oil was diluted in 2 ml cyclo hexan. Column model was TG-Wax MS (5% Phenyl Polysilphenylene-siloxane, 0.25 mm inner diameter * 60 m lenght, 0.25 µm film thickness). The ionization energy was calibrated as 70 eV, and the mass interval was m/z 1.2- 1200 amu. The scan mode was used as the screening more in data collection. MS transfer line temperature was 250°C, MS ionization temperature was 220°C, and whereas colon temperature was 50°C at the beginning, then it was increased up to 220°C with 3°C/min rate of temperature increase. The structure of each component was defined using mass spectrums (Wiley 9) with Xcalibur software.

RESULTS AND DISCUSSIONS

Sage flowers have 4-lobed superior ovary, could have maximum 4 nutlets from one flower. Seed set of pollinated plants was very low. Germination ratio was varied in different crosses as 4-15%. The essential oil ratios and components were determined from hybrid plants and their parent plants. All the hybrids and their parents cultivated in the same agricultural conditions and harvested at the same time in July. Mean values of essential oil ratio, components, and variation between parental plant species and hybrid genotypes were given in Table 1. The results showed a high variation among the analysed plants. Some hybrid genotypes gave higher essential oil ratio results than their parent species. Highest essential oil ratio in parent species was from *S. fruticosa* as 3%, in the hybrid plant of S. officinalis (mother) x S. aramiensis (father) and *S. officinalis* (mother) x *S. fruticosa* (father) gave the highest essential oil ratio results respectively, 4.21% and 4.08%. Lowest essential oil ratio found in S. aramiensis (mother) x S. fruticosa (father) hybrid plant as 0.75%. Essential oil ratios of hybrids (43 *S. aramiensis* x *S. fruticosa* hybrids, 15 *S. fruticosa* x *S. aramiensis* hybrids, 14 *S.* officinalis x *S. aramiensis* hybrids and 10 *S. officinalis* x *S. fruticosa* hybrids) were given in Table 2. *S. aramiensis* x *S. fruticosa* hybrids showed the lowest mean essential oil ratio (2.16%) value compare to all other hybrid combinations. Highest mean values were obtained from *S. officinalis* x *S. fruticosa* hybrids (3.25%). Different from our results in some studies hybrids of different Salvia species showed intermediate essential oil ratio from their parents (Putievsky et al., 1990; Herraiz-Penalver et al., 2015).

Table 1. Ratio of main components in the essential oil of three *Salvia* species and their hybrids

Parents and hybrids	**Essential Oil Ratio (%)**	**Eucalyptol (%)**	**Thujone (%)**	**Camphor (%)**
S. officinalis	2,0	20-25	25-30	30-35
S. fruticosa	3,0	45-55	N.D.	15-20
S. aramiensis	1,5	55-60	N.D.	5-15
S. officinalis x *S. fruticosa*	2,5-4,08	16,66-71,30	0,04-26,31	0,35-32,15
S. officinalis x *S. aramiensis*	1,71-4,21	12,49-61,91	0,84-21,58	0,25-47,11
S. fruticosa x *S. aramiensis*	1,76-3,84	8,71-60,11	1,19-14,62	8,71-60,11
S. aramiensis x *S. fruticosa*	0,75-3,97	4,02-60,20	0,02-23,77	0,42-57,04

N.D.=Not detected

Table 2. Essential oil ratios of the hybrid plants (%)

Hybrid Name	**Essential Oil Ratio**	**Hybrid Name**	**Essential Oil Ratio**	**Hybrid Name**	**Essential Oil Ratio**
S. ara. x *S. fru.*-1	1,40	*S. ara.* x *S. fru.*-29	2,50	*S. fru. x S. ara.*-14	3,15
S. ara. x *S. fru.*-2	1,46	*S. ara.* x *S. fru.*-30	3,26	*S. fru. x S. ara.*-15	2,90
S. ara. x *S. fru.*-3	3,29	*S. ara.* x *S. fru.*-31	2,97	*S. off.* x *S. ara.*-1	1,79
S. ara. x *S. fru.*-4	2,57	*S. ara.* x *S. fru.*-32	2,57	*S. off.* x *S. ara.*-2	2,26
S. ara. x *S. fru.*-5	1,52	*S. ara.* x *S. fru.*-33	2,79	*S. off.* x *S. ara.*-3	2,69
S. ara. x *S. fru.*-6	2,05	*S. ara.* x *S. fru.*-34	1,79	*S. off.* x *S. ara.*-4	3,00
S. ara. x *S. fru.*-7	1,23	*S. ara.* x *S. fru.*-35	1,14	*S. off.* x *S. ara.*-5	4,21
S. ara. x *S. fru.*-8	2,50	*S. ara.* x *S. fru.*-36	2,00	*S. off.* x *S. ara.*-6	2,28
S. ara. x *S. fru.*-9	3,97	*S. ara.* x *S. fru.*-37	2,79	*S. off.* x *S. ara.*-7	1,85
S. ara. x *S. fru.*-10	3,14	*S. ara.* x *S. fru.*-38	1,57	*S. off.* x *S. ara.*-8	2,36
S. ara. x *S. fru.*-11	1,93	*S. ara.* x *S. fru.*-39	2,50	*S. off.* x *S. ara.*-9	2,11
S. ara. x *S. fru.*-12	2,14	*S. ara.* x *S. fru.*-40	2,61	*S. off.* x *S. ara.*-10	1,85
S. ara. x *S. fru.*-13	0,93	*S. ara.* x *S. fru.*-41	1,43	*S. off.* x *S. ara.*-11	1,81
S. ara. x *S. fru.*-14	2,79	*S. ara.* x *S. fru.*-42	1,14	*S. off.* x *S. ara.*-12	3,37
S. ara. x *S. fru.*-15	2,50	*S. ara.* x *S. fru.*-43	1,00	*S. off.* x *S. ara.*-13	3,57
S. ara. x *S. fru.*-16	1,93	*S. fru. x S. ara.*-1	3,55	*S. off.* x *S. ara.*-14	1,71
S. ara. x *S. fru.*-17	0,75	*S. fru. x S. ara.*-2	2,25	*S. off.* x *S. fru.*-1	3,90
S. ara. x *S. fru.*-18	2,99	*S. fru. x S. ara.*-3	2,50	*S. off.* x *S. fru.*-2	3,79
S. ara. x *S. fru.*-19	2,62	*S. fru. x S. ara.*-4	1,76	*S. off.* x *S. fru.*-3	3,26
S. ara. x *S. fru.*-20	2,18	*S. fru. x S. ara.*-5	2,45	*S. off.* x *S. fru.*-4	4,08
S. ara. x *S. fru.*-21	2,48	*S. fru. x S. ara.*-6	3,15	*S. off.* x *S. fru.*-5	3,62
S. ara. x *S. fru.*-22	2,14	*S. fru. x S. ara.*-7	3,22	*S. off.* x *S. fru.*-6	2,64
S. ara. x *S. fru.*-23	1,14	*S. fru. x S. ara.*-8	3,84	*S. off.* x *S. fru.*-7	3,00
S. ara. x *S. fru.*-24	2,38	*S. fru. x S. ara.*-9	2,68	*S. off.* x *S. fru.*-8	2,50
S. ara. x *S. fru.*-25	1,92	*S. fru. x S. ara.*-10	2,60	*S. off.* x *S. fru.*-9	2,75
S. ara. x *S. fru.*-26	1,96	*S. fru. x S. ara.*-11	2,15	*S. off.* x *S. fru.*-10	3,00
S. ara. x *S. fru.*-27	2,05	*S. fru. x S. ara.*-12	2,60		
S. ara. x *S. fru.*-28	3,07	*S. fru. x S. ara.*-13	3,20		

* Essential Oil: SD=0,78;Max=4,21; Min=0,75; Mean=2,47

Table 3. Chemical composition of the hybrid plants essential oils (%)

Hybrid Name	Eucalyptol	Thujone	Camphor	Hybrid Name	Eucalyptol	Thujone	Camphor
S. ara. x *S. fru.*-1	19,38	0,26	25,71	*S. ara.* x *S. fru.*-42	19,01	0,73	5,83
S. ara. x *S. fru.*-2	16,76	0,54	22,61	*S. ara.* x *S. fru.*-43	31,18	1,25	26,35
S. ara. x *S. fru.*-3	5,21	0,49	19,60	*S. fru. x S. ara.*-1	49,15	14,62	0,42
S. ara. x *S. fru.*-4	20,12	23,77	31,54	*S. fru. x S. ara.*-2	32,93	10,46	18,67
S. ara. x *S. fru.*-5	22,35	1,03	24,42	*S. fru. x S. ara.*-3	25,41	8,45	21,03
S. ara. x *S. fru.*-6	29,35	0,43	21,88	*S. fru. x S. ara.*-4	30,17	9,54	17,31
S. ara. x *S. fru.*-7	21,16	1,02	15,09	*S. fru. x S. ara.*-5	21,75	2,55	24,22
S. ara. x *S. fru.*-8	20,35	0,02	35,05	*S. fru. x S. ara.*-6	59,11	4,71	1,43
S. ara. x *S. fru.*-9	22,44	7,65	18,94	*S. fru. x S. ara.*-7	27,56	7,14	15,63
S. ara. x *S. fru.*-10	26,38	0,40	23,06	*S. fru. x S. ara.*-8	25,64	11,35	17,16
S. ara. x *S. fru.*-11	14,34	1,04	57,04	*S. fru. x S. ara.*-9	8,71	1,19	2,36
S. ara. x *S. fru.*-12	46,55	0,28	5,14	*S. fru. x S. ara.*-10	60,11	3,18	2,97
S. ara. x *S. fru.*-13	55,99	0,34	4,06	*S. fru. x S. ara.*-11	40,93	2,17	22,37
S. ara. x *S. fru.*-14	57,54	0,33	3,86	*S. fru. x S. ara.*-12	42,67	5,39	19,04
S. ara. x *S. fru.*-15	11,39	0,34	36,63	*S. fru. x S. ara.*-13	39,05	2,52	27,19
S. ara. x *S. fru.*-16	12,49	0,35	36,20	*S. fru. x S. ara.*-14	53,53	2,40	9,60
S. ara. x *S. fru.*-17	4,02	0,36	38,79	*S. fru. x S. ara.*-15	34,79	1,27	31,36
S. ara. x *S. fru.*-18	30,03	0,25	24,00	*S. off.* x *S. ara.*-1	12,49	19,07	45,85
S. ara. x *S. fru.*-19	20,11	0,02	30,12	*S. off.* x *S. ara.*-2	42,72	0,84	4,43
S. ara. x *S. fru.*-20	12,29	0,66	21,88	*S. off.* x *S. ara.*-3	17,85	21,58	27,79
S. ara. x *S. fru.*-21	34,25	1,03	27,69	*S. off.* x *S. ara.*-4	17,04	18,65	47,11
S. ara. x *S. fru.*-22	22,67	0,70	25,81	*S. off.* x *S. ara.*-5	61,91	5,65	0,25
S. ara. x *S. fru.*-23	32,27	0,67	18,97	*S. off.* x *S. ara.*-6	35,64	15,37	15,25
S. ara. x *S. fru.*-24	15,93	0,51	23,99	*S. off.* x *S. ara.*-7	34,81	20,76	18,40
S. ara. x *S. fru.*-25	12,92	0,45	44,49	*S. off.* x *S. ara.*-8	43,84	1,09	3,36
S. ara. x *S. fru.*-26	9,67	0,23	38,45	*S. off.* x *S. ara.*-9	48,82	3,24	8,19
S. ara. x *S. fru.*-27	8,43	0,49	22,69	*S. off.* x *S. ara.*-10	37,29	11,22	18,50
S. ara. x *S. fru.*-28	25,98	1,30	21,30	*S. off.* x *S. ara.*-11	32,58	16,31	11,03
S. ara. x *S. fru.*-29	10,43	0,39	37,71	*S. off.* x *S. ara.*-12	41,35	14,72	19,56
S. ara. x *S. fru.*-30	19,73	0,32	24,51	*S. off.* x *S. ara.*-13	38,93	9,67	7,16
S. ara. x *S. fru.*-31	26,91	0,17	23,89	*S. off.* x *S. ara.*-14	35,62	19,42	12,45
S. ara. x *S. fru.*-32	20,64	0,03	35,76	*S. off.* x *S. fru.*-1	47,58	2,30	7,78
S. ara. x *S. fru.*-33	15,00	0,91	28,24	*S. off.* x *S. fru.*-2	39,48	2,76	16,64
S. ara. x *S. fru.*-34	59,21	1,04	1,96	*S. off.* x *S. fru.*-3	71,30	2,66	0,35
S. ara. x *S. fru.*-35	53,78	0,79	0,53	*S. off.* x *S. fru.*-4	55,03	6,03	0,66
S. ara. x *S. fru.*-36	46,54	0,81	10,19	*S. off.* x *S. fru.*-5	23,12	25,77	32,15
S. ara. x *S. fru.*-37	21,84	0,43	19,37	*S. off.* x *S. fru.*-6	16,66	20,28	16,99
S. ara. x *S. fru.*-38	25,57	0,49	29,71	*S. off.* x *S. fru.*-7	22,20	0,04	28,07
S. ara. x *S. fru.*-39	42,25	0,25	16,39	*S. off.* x *S. fru.*-8	21,17	26,31	23,51
S. ara. x *S. fru.*-40	60,20	0,07	1,05	*S. off.* x *S. fru.*-9	25,94	19,65	14,63
S. ara. x *S. fru.*-41	32,71	0,18	37,67	*S. off.* x *S. fru.*-10	28,71	15,95	9,06

**Eucalyptol: SD=15,57; Max=71,30; Min=4,02; Mean=30,72 ***Thujone: SD=7,43; Max=26,31; Min=0,02; Mean=5,35
****Camphor: SD=12,64; Max=47,11; Min=0,25; Mean=19,98

Results of gas chromatography analysis from essential oils were given in Table 3. The aim of the study was to determine whether different chemotypes could obtain by interspecific crossing or not. Especially low camphor and thujone, high eucalyptol levels were important. *S. aramiensis* as a parent has the highest eucalyptol level as 60%, followed by *S. fruticosa*. In all combinations similar or even higher eucalyptol ratios were obtained. Highest eucalyptol ratio observed from the hybrid of *S. officinalis* x *S. fruticosa* crossing as 71.30%. Thujone level was ranged between 0-30% in parents, in hybrids lowest thujone in all combinations observed. Furthermore, low thujone (0,04%) bearing *S. officinalis* x *S. fruticosa* hybrid obtained. Highest thujone level also found in *S. officinalis* x *S. fruticosa* hybrids as 26.31%. The highest camhor bearing parent species was *S. officinalis* with 35% however in hybrids higher camphor ratio were observed as 60.11%. *S. officinalis* parental genotype with low camhor (0.25%) was also observed. Putievsky et al. (1990), conducted that essential oil components of *S. officinalis*, *S. fruticosa* and *S. tomentosa* hybrids were varied and in some case components resembled or between their parents. Study from Herraiz-

Penalver et al., 2015 showed that natural hybrid of *S. officinalis* and *S. lavandulifolia* essential oil components also effected parental species and in some cases both separate main components of parents could occur as 2 main components in their hybrids. Chromatogram sample of the one hybrid from *S. officinalis* x *S. fruticosa* were given in Figure 1.

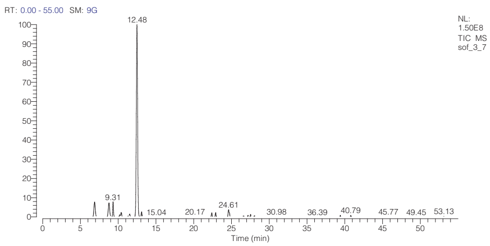

Figure 1. Chromatogram of *S. officinalis* x *S. fruticosa*-3 hybrid

CONCLUSIONS

The study was preliminary results of interspecific hybridisation of *S. officinalis*, *S. fruticosa* and *S. araemiensis*. The hybrids were distinguished from their parent species among essential oil ratios and components. *S. aramiensis* x *S. fruticosa* hybrids 14, 34 and 40; *S. officinalis* x *S. aramiensis* hybrid 5; *S. fruticosa* x *S. aramiensis* hybrid 10 and *S. officinalis* x *S. fruticosa* hybrid 3 were selected regarding to essential oil components. Selected hybrids will cultivate for further studies. In the ongoing study morphological characteristics will also be evaluated.

ACKNOWLEDGEMENTS

This research work was carried out with the support of Coordinatorship of Hatay Mustafa Kemal University project number 18.D.007.

REFERENCES

Aşkun, T., Başer K.H.C, Tümen G., Kürkçüoğlu M. (2010). Characterization of essential oils of some Salvia species and their antibacterial activities. *Turkish Journal of Biology*, *34*. 89–95.

Ayanoğlu, F., Kaya, D.A., Mert, A., Uygur, V. (2012). Hatay yöresinde doğal olarak yetişen adaçayı (*Salvia* spp.) türlerinin kültüre alınması, verim ve kalite özelliklerinin belirlenmesi, Proje Sonuç Raporu (TÜBİTAK-109O161).

Baydar, H. (2005). Tıbbi, Aromatik ve Keyf Bitkileri, Süleyman Demirel Üniversitesi (1. Baskı), Isparta.

Bayram, E. (1998). Batı Anadolu Florasında Yetişen Adaçayı (Salvia fruticosa Mill)'nda Uygun Tiplerin Seleksiyonu Üzerinde Araştırma. Tübitak, TOGTAG-1468.

Bazina, E., Makris, A., Vender, C., Skoula, M. (2008). "Genetic and chemical relations among selected clones of Salvia officinalis", *Journal of Herbs, Spices & Medicinal Plants, 9*(4), 269–273.

Bielenberg, J. (2007). Thujone, Med Monatsschr Pharm. Sep., *30*(9), 322–326.

Böszörményi, A, Héthelyi, E, Farkas, A, Horváth, G, Papp, N, Lemberkovics, E, Szoke, E. (2009). Chemical and genetic relationships among sage (*Salvia officinalis* L.) cultivars and Judean sage (*Salvia judaica* Boiss.). *J. Agric. Food Chem., 57*(11), 4663–4667. doi: 10.1021/jf9005092.

Cuvelier, M.E., Berset, C, Richard, H. (1994). Antioxidant constituents in Sage (*Salvia officinalis*). *J. Agric. Food Chem*., *42*. 665–669.

Cvetkovikj, I., Stefkov, G., Karapandzova, M., Kulevano, S. (2015). Essential oil composition of *Salvia fruticosa* Mil.. populations from Balkan Peninsula. *Macedonian Pharmaceutical Bulletin*, *61*(1), 19–26.

Davis, P.H., Mill R.R., Tan, K. (1988). *Salvia* L. *Flora of Turkey and the East Aegean Islands, Vol.7.,* Edinburgh: Edinburgh Univ. Press.

Demirci, B., Baser, K.H.C., Tumen, G. (2002). Composition of the essential oil of Salvia aramiensis Rech. Fil. growing in Turkey. *J. Flav. Fragr.*, *17*, 23–25.

De Vincenzi, M., and Mancini, E. (1997). Monographs on botanical flavouring substances used in foods. Part VI. *Fitoterapia, 68*(1), 49–61.

Doğan, M., Pehlivan, S., Akaydın, G., Bağcı, E., Uysal, İ., Doğan, H.M. (2008). Türkiye'de yayılış gösteren *Salvia* L. (*Labiatae*) cinsinin taxonomik revizyonu. Tübitak Proje No: 104 T 450

Ertaş, A., Alkan, H., Akdeniz, M., Şahin, H., Alacabey, İ., Odabaşı, M., Acet, Ö., Kolak, U. (2017). The antioxidant activity and chemical composition of essential oils of *Salvia aramiensis* and *Calamintha nepeta*. *The 3rd International Symposium on EuroAsian Biodiversity, s 612. 05-08 July, Minsk – Belarus.*

European Medicines Agency, 2010. Community Herbal monograf on *Salvia officinalis* L. folium. Committee on Herbal Medicinal Products.

Haque, S., Ghoshal, K.K. (1981). Floral biology and breeding system in the genus Salvia L. *Proc. Indian natn. Sci. Acad.*, *47*(5), 716–724.

Hedge, I.C. (1982). Salvia L.: *Flora of Turkey and the East Aegean Islands*. Editör: Davis P.H. Edinburgh: Edinburgh University Press, S. 400-461.

Herraiz-Penalver, D., Elguea-Culebras, G.O., Vioque, R.S., Meridas, S.O. (2015). Identification of a hybrid species of sage (*Salvia officinalis* L. x *S. lavandulifolia*) through the study of the essential oil. *Journal of Essential Oil Research*, *27*(5), 363–372.

Ipek, A., Gürbüz, B. (2010). *Salvia* Species in Flora of Turkey and Their Status in Danger. *Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi, 19*, 30–35.

Kahraman, A., Celep, F., Dogan, M. (2009). Comparative morphology, anatomy and palynology of two *Salvia* L. species (*Labiatae*) and their taxonomic implications. *Bangl. J. Plant Taxon*, *16*, 73–82.

Karaman, S., İlçim A., Çömlekçioğlu, N. (2007). Composition of the essential oils of Salvia aramiensis Rech. fil. and *Salvia cyanescens* Boiss. & Bal. *Pakistan Journal of Botany*, *39*(1), 169–172.

Kelen, M., Tepe, B. (2008). Chemical composition, antioxidant and antimicrobial properties of the essential oils of three Salvia species from Turkish flora. *Bioresource Technology*, *99*, 4096–4104.

Kintzios, S.E. (2000). *Sage: The Genus Salvia*. Amsterdam: Harwood Academic Publishers. ISBN 90-5823-005-8.

Lamaison, J.L., Petitjean-Freytet, C, Carnat, A. (1991). Medicinal Lamiaceae with antioxidant properties, a potential source of rosmarinic acid. *Pharm Acta Helv*, *66*, 185–8.

Lawrence B.M. (1998). Progress in essential oils–sage oil. *Perfum Flavor, 23*, 47–52.

Liebelt, E.L., and Shannon, M.W. (1993). Small doses, big problems: a selected review of highly toxic common medications. *Pediatr. Emerg. Care.*, *9*(5), 292–297.

Mačukanović Jocić, M.P., Stevanovic, Z.D., Mladenović, M., Jocić, G. (2011). Flower morphophysiology of selected Lamiaceae species in relation to pollinator attraction. *Journal of Apicultural Research, 50*(2), 89–101.

Millet, Y., Jouglard, J., Steinmetz, M.D., Tognetti, P., Joanny, P., Arditti, K. (1981). Toxicity of some essential plant oils. Clinical and experimental study. *Clin. Toxicol., 18*(12), 1485–1498.

Pelkonen, O., Abass, K., Wiesener, J. (2013). Thujone and Thujone containing herbal medicinal products: Toxicological assessment. *Regulatory Toxicology and Pharmacology, 65*(1), 100–107.

Putievsky, E., Ravid, U., Diwan-Rinzler, N., Zohary, D., (1990). Genetic affinities and essential oil composition of *Salvia officinalis* L., *S. fruticosa* Mill., *S. tomentosa* and their hybrids. *Flavour Fragr. J.*, *5*, 121–123.

Radosavljević, I., Bogdanović, S., Celep, F., Filipović, M., Satovic, Z., Surina, B., Liber, Z. (2019). Morphological, genetic and epigenetic aspects of homoploid hybridization between *Salvia officinalis* L. and *Salvia fruticosa* Mill. Scientific Reports, 9:3276 | https://doi.org/10.1038/s41598-019-40080-0.

Shahabi, S., Jorsaraei, S.G.A, Moghadamnia, A.A., Zabihi, E., Aghajanpour, S.M., Kani, S.N.M., Pourbagher, R., Hosseini, S.A., Esmaili, M., Yoonesi, A.A., Zarghami, A., Alinezhad, F. (2012). Central effects of Camphor on GnRH and sexual hormones in male rat. *Int. J. Mol. Cell. Med., 1*(4): 191–196.

Skoula, M., Hilali, I.E., Makris, A.M. (1999). Evaluation of the genetic diversity of *Salvia fruticosa* Mill. clones using RAPD markers and comparison with the essential oil profiles. *Biochemical Systematics and Ecology*, *27*, 559–568.

Subası, U., and Guvensen, A. (2011). Breeding systems and reproductive success on *Salvia smyrnaea. Turk J. Bot.*, *35*, 681–687.

Theis, J.G., and Koren, G. (1995). Camphorated oil: still endangering the lives of Canadian children. *CMAJ, 152*(11), 1821–1824.

Tychonievich, J., and Warner, R.M. (2011). Interspecific crossability of selected *Salvia* species and potential use for crop improvement. *J. Am. Society Horticultur. Sci.*, *136*, 41–47.

Walker, J.B., and Sytsma K. J. (2007). Staminal Evolution in the Genus *Salvia* (*Lamiaceae*): Molecular Phylogenetic Evidence for Multiple Origins of the Staminal Lever. *Annals of Botany*, *100* (2), 375–391. http://doi.org/10.1093/aob/mcl176.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

DESCRIBING THE *Ranunculus* GENUS BASED ON THE PLANTS PRESENT IN ALEXANDRU BELDIE HERBARIUM

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Abstract

The present paper presents the morphological and ecological description of species belonging to the Ranunculus genus found in "Alexandru Beldie" Herbarium from "Marin Drăcea" National Institute for Research and Development in Forestry (INCDS), Bucharest. A database was created with information about species, their gathering year, as well as the botanists who have identified and conserved them. The first part of the article shortly describes the herbarium and its specific, together with a presentation of the material and method used for elaborating this paper. As such, the material that was used is represented by the 480 vouchers that contain the specimens of 106 species belonging to the Ranunculus genus. The plants were gathered between 1818 and 1994, with a larger incidence during1880-1890 and 1931-1950. The paper ends with a series of conclusions regarding the analysis of the Ranunculus genus species and specimens present in the herbarium.

Key words: *herbarium, plants, Ranunculus, botanists.*

INTRODUCTION

The "Alexandru Beldie" Herbarium from "Marin Drăcea" National Institute for Research and Development in Forestry, located in Bucharest, contains an impressive collection (40000 vouchers) of certain plants, preponderantly from mountain areas. The plants from the collection are kept in their original folders and are arranged in 600 drawers (Vasile et al., 2017). This collection is enrolled as INDEX HERBARIUM and all the species were gathered by known personalities in the field of systematic botany, one of the Romanian botanists being Alexandru Beldie, who dealt especially with this herbarium. As such, the herbarium is named after Alexandru Beldie, a renowned Romanian botanist, which was very interested in the flora present in Bucegi Mountains (Beldie, 1967; Beldie, 1972).

Besides the *Ranunculus*s pecies presented in this paper, the Herbarium also contains other species such as 32 *Arabis* species (Dincă et al., 2017a), 29 *Alyssum* species (Cântar C. et al., 2018), 40 *Alchemilla* species (Deleanu E. et al., 2018a), 29 *Plantango* species (Deleanu E. et al., 2018), 19 *Centaurea* species (Dincă et al., 2017b), 112 *Hieracium* species (Dincă et al., 2017c), 15 *Ornithogalum* species (Enescu R. et al., 2017) and 19 *Scorzonera* species (Dincă et al., 2017d).

Ranunculus is the largest genus within Ranunculaceae including about 600 species, primarily distributed in temperate to arctic or subantarctic zones, with a few species also present in tropic high montane regions (Tamura, 1995).

The aim of this article is to present the state of this collection, to describe the species and the total number of *Ranunculus* specimens (106 species), together with the date when they were collected, their location, the botanist who collected each exemplar and their conservation.

MATERIALS AND METHODS

The work methods used are the ones characteristic to the research activity. As such, research and bibliographic documentation have played a very important role, especially from a morphologic and ecologic point of view. Together with these methods, analysis and synthesis were used as main method for digitizing and systematizing the data from the herbarium's vouchers. Furthermore, creating the map, preparing the work, results and its conclusions have implied the analysis and synthesis of the initially systematized data.

The vouchers were grouped by species, harvest year, the place where they were harvested and by the specialist who harvested them. An excerpt of the *Ranunculus* genus inventory is rendered in Table 1.

Table 1. The inventory of *Ranunculus* genus from INCDS Bucharest's, "Alexandru Beldie" Herbarium (excerpt)

Plate no.	Drawer no.	Herbarium/ Botanic collection/ Institution	Species	Harvest date	Harvest place	Collected/ Determine d by:	Conser vation degree (1..4)
90	33	Bucharest's Polytechnics School Herbarium/ Botanic Laboratory	*Ranunculus acer* L.	1918.09.07	Bucegi: Colţii lui Barbeş	M. Haret	1
90	3	Herbarul Institutului de Cercetări Silvice/ Ministerul Agriculturii şi Silviculturii	*Ranunculus acris*	1954.08.07 .	Bucegi: Poiana Crucii	Al. Beldie	1
90	50	Forestry Research Institute Herbarium/ Agriculture and Silviculture Ministry	*Ranunculus arvensis* L.	1933.05.01	Păd. Cormana	C. C Georgescu	1
91	22	Bucharest's Polytechnics School Herbarium/ Botanic Laboratory	*Ranunculus illyricus* L.	1939.05.12	Hanul Conachi, jud. Tecuci	C.C Georgescu	2
91	43	Bucharest's Polytechnics School Herbarium/ Botanic Laboratory	*Ranunculus flammula* L.	1937.05.29	Maramureş Vişeul de Jos,alt 457 m	A. Coman	1
91	58	Bucharest's Polytechnics School Herbarium/ Botanic Laboratory	*Ranunculus constantinopolitanus* DC D'Urville	1935.05.01	Ilfov, păd. Cărcioarele	Al. Beldie	1
91	75	Bucharest's Polytechnics School Herbarium, Silviculture Faculty/ Botanic Laboratory	*Ranunculus cassubicus* L.	1943.06.01	Bucegi: Valea Coştilei 1300 m	Al. Beldie	2
92	14	Bucharest's Polytechnics School Herbarium, Silviculture Faculty/ Botanic Laboratory	*Ranunculus hornschuchii* Hoppe	1943.06.01	Bucegi: ValeaAlbă 1600 m	Al. Beldie	1
92	19	Bucharest's Polytechnics School Herbarium, Silviculture Faculty/ Botanic Laboratory	*Ranunculus montanus*	1947.07.17	Bucegi	Al. Beldie	2
92	37	ICEF, Forestry Research and Experimentation Institute	*Ranunculus napolitanus* Ten.	1940.05.12	Hagilar, jud. Comstanţa	I. Lupu	1
93	32	ICEF, Forestry Research and Experimentation Institute	*Ranunculus polyanthemos* L.	1936.06.14	Gurghiu, jud. Mureş	S. Paşcovschi Al. Beldie	1
93	17	Herbarul Şcoalei Politehnicei Bucureşti/ Laboratorul de botanică	*Ranunculus platanifolius* L.	1938.07.01 .	Ţibleş-Fundu Dileţului	I. Morariu	2
94	16	ICEF, Institutul de Cercetări şi Experimentaţie Forestieră	*Ranunculus repens* L.	1937.05.21 .	Pădurea Casa Verde, Timişoara	S. Paşcovschi	1

RESULTS AND DISCUSSIONS

The research material is composed of the 480 *Ralunculus* genre species vouchers present in the maps of "Alexandru Beldie" Herbarium from INCDS "Marin Drăcea". *Ranunculus* belongs to the Ranunculaceae Family, the Ranuculales order having approximately 600 species. Members of the genus include buttercups, spearwort and water crowfoots.

The species belonging to this genus that are present in the above-mentioned collection are the following:

Ranunculus acris is a herbaceous perennial plant that grows up to a height of 30-70 cm, with ungrooved flowing stems bearing glossy yellow flowersabout 25 mm across (Figure 1). Five overlapping petals exist above five green sepals that soon turn yellow as the flower mature. The plant has numerous stamens inserted below the ovary. The leaves are compound, with three lobed leaflets. Unlike *Ranunculus repens*, the terminal leaflet is sessile (https://en.wikipedia.org/wiki/Ranunculus_acris).

Figure 1. *Ranunculus acris*

Ranunculus abortivus produces erect stems ranging from 10 to 60 cm in height. The leaves are variable in shape, while both stems and leaves are hairless. The basal leaves are kidney-shaped to circular and persistent, with scalloped margins, while the stem leaves are alternate and deeply lobed or divided. Those at the bottom have long petioles (stems), while those at the top are shorter-stemmed to stemless, with narrow blades or lobes. Each stem can bear up to 50 flowers. The flower has five petals up to 1.5 to 3.5 mm long, with a ring of stamens around a round cluster of green carpels. The carpels develop into brown, shiny rounded and slightly flattened achenes with a tiny beak (https://en.wikipedia.org/wiki/Ranunculus_abortivus) (Figure2).

Figure 2. *Ranunculus abortivus*

Ranunculus asiaticus, native to the eastern Mediterranean region of south-western Asia, south-eastern Europe (Crete, Karpathos) and north-eastern Africa. It is a herbaceous perennial plant that can reach 45 cm in height, with simple or branched stems. The basal leaves are three-lobed, with leaves higher on the stems more deeply divided; like the stems, they are downy or hairy. The flowers are 3-5 cm in diameter, variably red to pink, yellow, or white, with one to several flowers on each stem (https://en.wikipedia.org/wiki/*Ranunculus_asiaticus*).

Ranunculus bulbosus, is a perennial plant that has attractive yellow flowers, and deeply divided, three-lobed long-petioled basal leaves.

Bulbous buttercup is known to form tufts. The stems are 20-60 cm tall, erect, branching, and slightly hairy flowering. Alternate and sessile leaves are present on the stem. The flower forms at the apex of the stems, and is shiny and yellow with 5-7 petals. The flowers are 1.5-3 cm wide. The plant blooms from April to July (https://en.wikipedia.org/wiki/Ranunculus_bulbosus).

Ranunculus platanifolius. The large white buttercup is an herbaceous plant 30-100 cm tall, with glabrous stem characterized by many branches. The leaves are palmate, each divided into five segments with dentate margins. Flowers are organized into cymes, each flower having a calyx with five sepals, a corolla with five white petals, many stamens with yellow anthers and many styles (Figure 3) (https://en.wikipedia.org/wiki/Ranunculus_planifolius).

Figure 3. *Ranunculus platanifolius*

Ralunculus repens is a herbaceous, stoloniferous perennial plant growing up to 50 cm in height. It has both prostate running stems that produce roots and new plants at the nides, and less erect flowering stems. The basal leaves are compound, grown on a 4-20 cm long petiole and divided into three broad leaflets 1.5-8 cm long, shallowly to deeply lobed, each stalked. The higher leaves from the stems are smaller, with narrower leaflets and may be simple and lanceolate. Both the stems and the leaves are hairy. The flowers are golden yellow, glossy, reaching a diameter of 2-3 cm, usually with five petals, while flower stem is finely grooved. The gloss is caused by the smoth upper surface of the petal that acts like a mirror, the gloss aids in attracting pollinating insect and thermo regulating the flowers reproductive organs. The fruit is a cluster of achenes being 2.5-4 mm long. Creeping buttercup has three- lobed dark green, white-spotter leaves that grow out of the node. It grows in field and pastures pefering wet soils (Figure 4) (https://en.wikipedia.org/wiki/Ranunculus_repens).

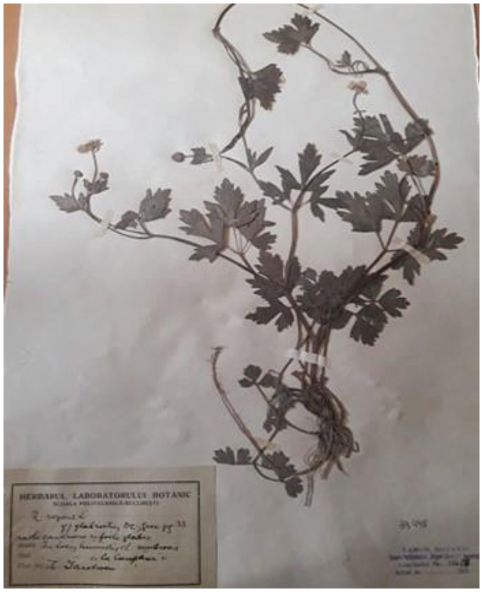

Figure 4. *Ralunculus repens*

The plant's harvest years. The plants were gathered in a time period ranging between 1818 and 1994. The oldest plants of this genre are *Ranunculus amplexicaulis.*, collected in 1818. The periods in which most plants were gathered were 1880-1890 and 1931-1950 (Figure 5).

The plants were gathered from Romanian mountain areas (Bucegi, Retezat, Rotnei) or near cities from our country (Azuga, Constanta, Craiova, Caracal, Brasov, Buzau, Branesti), as well as from some European areas (Austria, Albania, Croatia, Franta). The plants are in a good state.

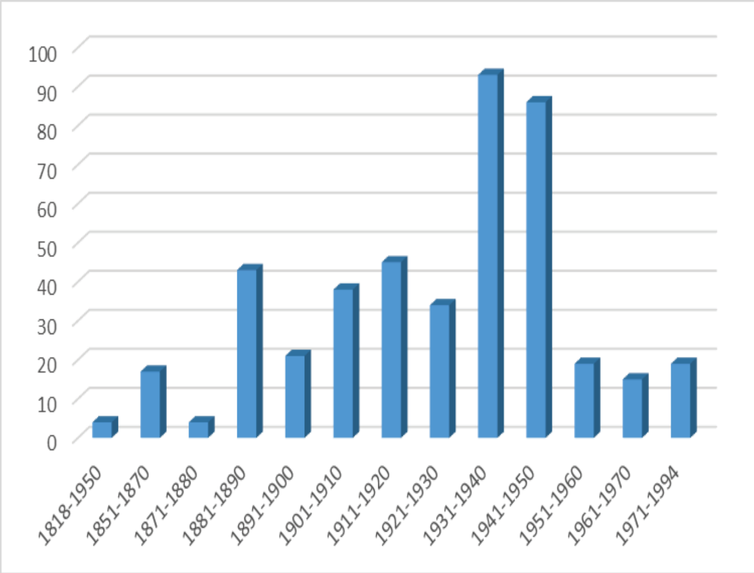

Figure 5. Harvesting periods for *Ralunculus* plants from Al. Beldie

CONCLUSIONS

In "Alexandru Beldie" Herbarium, which contains more than 40,000 vouchers, 480 vouchers belong to the *Ranunculus* genus.

Even if the first fir samples were collected almost 140 years ago, the majority of them are in a good conservation state, meaning that the methods used for preserving the biological materials were adequate.

The plants from this herbarium were gathered between 1818 and 1994, reaching a maximum in the period 1880-1890 and 1931-1950. Furthermore, they were gathered by renowned Romanian and foreign botanists (Al. Beldie, P. Cretzoiu, C. Georgescu, E.I. Nyarady, N. Boscan, G. Bujoreanu).

The plants were gathered from Romanian mountain areas (Bucegi, Retezat, Rotnei) or near cities from our country (Azuga, Constanta, Craiova, Caracal, Brasov, Buzau, Branesti), as well as from some European areas (Austria, Albania, Croatia, Franta). The plants are in a good state.

REFERENCES

Beldie, A. (1967). *Flora andvegetation of Bucegi Mountains*. Bucharest, RO: Academiei R.S.R. Publishing.

Beldie, A. (1972). *Flora andvegetation of Bucegi Mountains*. Bucharest, RO: Academiei R.S.R. Publishing House.

Cântar, I.C., Vechiu, E., Dincă, L. (2018). Plants conserve din "Alexandru Beldie" Herbarium – *Alyssum*genus. *Annals of the University of Craiova Series Biology*, *Horticulture*, *Food produce processing technology, Environmental engineering, 23*(59), 352–358.

Deleanu, E., Ionescu, M., Vechiu, E. (2018). The characteristics of Alchemillagenre plants present in Alexandru Beldie Herbariumfrom INCDS Bucharest. *Annals of University of Craiova - Agriculture, Montanology, Cadastre Series, 48*(2), 76–81.

Deleanu, E., Ionescu, M., Dincă, M. (2018). Plantago genus collection species present in Alexandru Beldie Herbarium. *Annals of the University of Craiova Series:Biology*, *Horticulture*, *Food produce processing technology*, *23*, 374–379.

Dincă, L., Cântar, I.C., Dincă, M. (2017). The characteristics of plant species from *Arabis* type present in Al. Beldie Herbariumfrom INCDS Bucharest. *Annals of West University of Timişoara, ser.* Biology, *20*(2), 115–122.

Dincă, L., Vasile, D., Voiculescu, I. (2017). The characteristics of plants from *Hieracium* genre present in Alexandru Beldie Herbariumfrom INCDS Bucharest. *Lucrări ştiinţifice USAMV Iaşi, Seria Horticultură, 60*(1), 39–48.

Dinca, L., Cantar, I.C. (2017). A short description of *Scorzonera* plant species present in Alexandru Beldie Herbariumfrom INCDS Bucharest, *Annals of University of Craiova - Agriculture, Montanology, cadastre Series, 47*, 118–126.

Dincă, L., Vasile, D., Dincă, M. (2017). Short characterisation of plant species from *Centaurea* genuspresent in Al. Beldie Herbarium from Marin Dracea National Institute for Reseachand Development in Forestry Bucharest. *Annals of University of Craiova - Agriculture, Montanology, Cadastre Series*, 47, 127–135.

Enescu, R., Dincă, L., Cântar, I.C. (2017). Description of plant species of *Ornithoghalum* genus present in *Al. Beldie* Herbarium from "Marin Drăcea" Bucharest NIRDF. *Journal of Horticulture, Forestry and Biotechnology, 21*(3), 89–95.

Tamura, M. (1995). *Ranunculaceae. In: Hiepko, P*. Berlin, DE: Die Natürlichen Pflanzenfamilien, 2nd ed. Vol. 17a (4).

Vasile, D., Dincă, L., Indreica, A., Voiculescu, I. (2016). "Alexandru Beldie" Herbarium - collection of plants and an important database for specialists. *Revista de Silvicultură și Cinegetică*, *21*(39),114–119.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOME OBSERVATIONS ON THE MORPHOLOGY AND GERMINATION OF *Bromus secalinus* L. CARYOPSIS

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Abstract

The morphological features and aspects of the germination process of the caryopsis of a Bromus secalinus population out of a wheat field from Pitesti Research Station were studied. The average value of the lemma - 8.11 mm distributes this population in the lemma long-sized group and the length of the dorsal awn - 4.85 mm on average and their appearance, allocate this population to var. secalinus, f. elongatus. At different moments of the germination process, on the surface of the coleorhiza hairs were noticed. The germination rate of seeds of Bromus secalinus is strongly stimulated by the presence of Triticum durum caryopsis while Brassica napus seeds reduce the number of Bromus germinated caryopsis.

Key words: *Bromus secalinus, caryopsis, coleorhiza hairs, germination process, Pitesti Station population.*

INTRODUCTION

Bromus secalinus has its origin in the Mediterranean region of Eurasia (Ciocârlan, 2009). In Romania, it can be sporadically find in grain fields, meadows, wasteland, field borders, and roadsides from oak woodland zone to beech forest belt. Calcifuge species (Ciocârlan & Chirilă, 1982), *Bromus secalinus* prefer slightly acidic soils with pH ranging from 5.5 to 7.5 (Rzymowska et al., 2010) and a cool climate with rainfall during the year (Anghel et al., 1972). Beeing in regress at some point, in Europe their populations were considered becoming even endangered (Warcholińska in 1981 regarded this species like rare among cereals).

The increasing incidence of the species as a weed in crops is due to the reduced soil tillage (Szymankiewicz et al., 2003) which allows access of the seeds to the light: a large percentage of seeds germination was reported in the case of germination experiments with diaspores placed on the soil surfaces or at the 1.5 cm depth (Cussans et al., 1994; Adamczewski et al., 2015).

The germination ability of the seed decreases with age and it is limited to 1-2 years (Kapeluszny & Haliniarz, 2007).

A plant can produce up to 1400 caryopses (Ciocârlan et al., 2004). These are closely wrapped in paleas; lemma, strongly inrolled at maturity, of 6-9 (11) mm length, minutely bifid at apex is provided with a rudimentary awn or of variable length (4-9 mm) or sometimes it may be missing (Häfliger & Scholz, 1981). Some of caryopsis physical features, namely width, thickness and density of the seeds were used in the separation of *Bromus secalinus* from wheat caryopsis (Hauhouot-O'Hara et al., 2000).

Morphological variability reflects the genetic variance of a species and the ability of their populations to adapt to local environmental conditions (Ionescu et al., 2017; Ionescu et al., 2018). Ecological factors such as soil humidity and trophicity can influence the phenotypic characteristics of *Bromus secalinus* populations through seed storage proteins (Skrajna et al., 2012); also, the crop plants can affect the fertility rate of the *Bromus secalinus* seeds (Rzymowska et al., 2010).

The influence of local environmental conditions on the morphological characteristics and the germination process of the caryopsis of *Bromus secalinus* population from Pitești Station together with some crop plants was targeted in this investigation.

MATERIALS AND METHODS

Morphological observations and those on germination were carried on caryopsis collected in 2016 from a *B. secalinus* populations found in a durum wheat field from Agricultural Research and Development Station - Pitești (Argeș County).

The climate of the area is characterised by an average of the annual precipitation of 700 mm. The soil is of albic luvisols type with a low value of pH (around 5) and a small content of organic substances (total C = 2%); the level of NPK macro-elements is reduced, and aluminium ions are present in the soil solution.

The measurements were made on 100 caryopses collected from the low part of spikelets (usually, the second floret from bottom) (Stace, 2011). A stereomicroscope type S8APO, equipped with a video camera Leica DFC 295 and a SEM FEI Inspect S50 were used to reveal the specific morphological and micromorphological features of the *B. secalinus* population.

In order to observe the behaviour of B. secalinus as well as those of crop plants during the germination process in cases of dense populations of *Bromus secalinus* caryopsis, germination variants were selected based on a 2:1 ratio.

Germination experiments were conducted in the subsequent configurations:

- control variant - 20 caryopsis of *B. secalinus*;
- variant I - 40 caryopsis of *B. secalinus* and 20 of *Triticum durum*;
- variant II - 40 caryopsis of *B. secalinus* and 20 seeds of *Brassica napus* subsp. *napus*.

The variants were experimented with 3 separated repetitions each, at one-week intervals, during 12 to 29 March 2018.

22 hours after the sowing, *B. secalinus* caryopsis have been observed and phases of the germination process were noted. The germination rate was noted during 3 days in all 3 variants.

RESULTS AND DISCUSSIONS

The morphology and micromorphology of the caryopsis of *Bromus secalinus* (Pitești population)

Lemmas of the *B. secalinus* - Pitești population are 7-9 mm long (8.11 mm on average; 0.373 - STD) (Table 1). According to different papers (Häfliger & Scholz, 1981; Ciocârlan et al., 2004; Hauhouot-O'Hara et al., 2000), the length of the lemmas ranges from 6 to 9 (11) mm.

Table 1. The morphological characteristics of the *B. secalinus* - Pitești populations lemma and awn

	Length interval (mm)	Average (mm)	STD
Lemma	7 - 9	8.11	0.37
Awn	2 -7.2	4.85	1.33

The average value distributes this population of *B. secalinus* in the lemma long-sized group, so the caryopsis. It is well known that seed size may determine the viability of the seedlings and the vigour of the adult plants (Massimi, 2018).

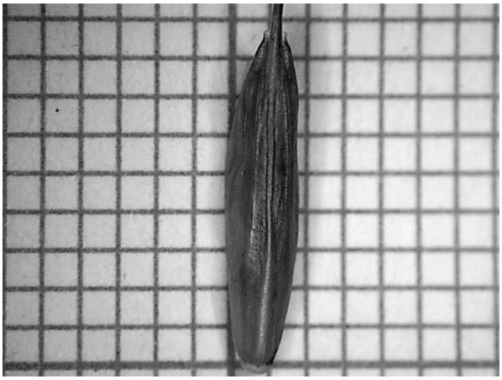

Figure 1. Lemma on dorsal side

The lemma is rounded on dorsal side, 7-veined (Figure 1) and has membranous margins that can be seen from the middle to the upper region (Figure 2).

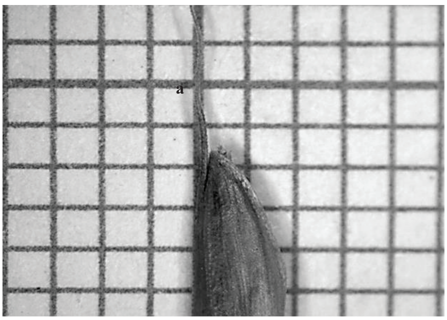

Figure 2. Lemma - margins and apex (a - awn)

At the apex, lemma is minutely bifid and covered with short bristles, similar to those

seen on the awn (Figure 3). The same type of bristle can be discerned at the base of the lemma, above the callus region (Figure 4).
The awn is variable in length, from 2 to 7.2 mm, with 4.85 mm on average (STD = 1.33) (Table 1). It can be straight or slightly flexuous (Figure 2).

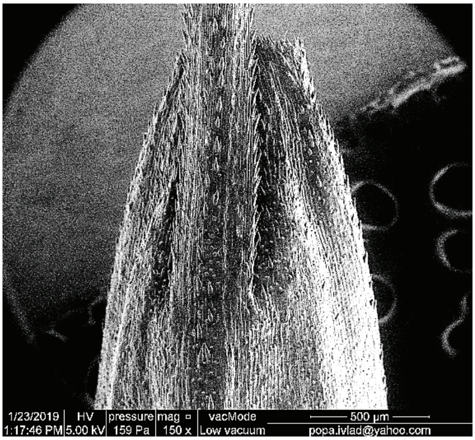

Figure 3. Lemma at the apex

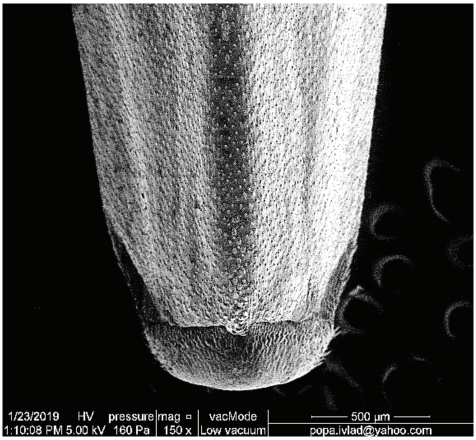

Figure 4. Lemma at the base

According to Flora RSR (Săvulescu, 1972), the length of the dorsal awn and their appearance, allocate this population to var. *secalinus*, f. *elongatus*.
On the ventral side, the caryopsis is protected by a palea almost equalling lemma at the distal end (Figure 5); palea is provided on the edges, from the middle to the base, with stiff bristles (Figure 6).

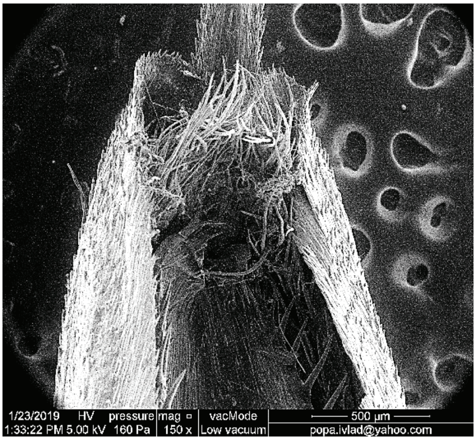

Figure 5. Palea at the distal end

Figure 6. Palea at the base

The germination processes
At 22 hours after sowing, germination was initiated in all three variants. First, the coleorhiza and then the radicle were observed to *B. secalinus* germinated caryopsis.
At different moments of the germination process, on the surface of the coleorhiza hairs were noticed (Figure 7).
The origin and the role of this coleorhiza type, observed in other species, have been discussed in different scientific papers (Nishimura, 1922; Nostrog, 1955; Morita et al., 1990; Debaene-Gill et al., 1994; Chistyakova et al., 2016).
There are no reports on *Bromus* species with this kind of coleorhiza in the reviewed scientific works.

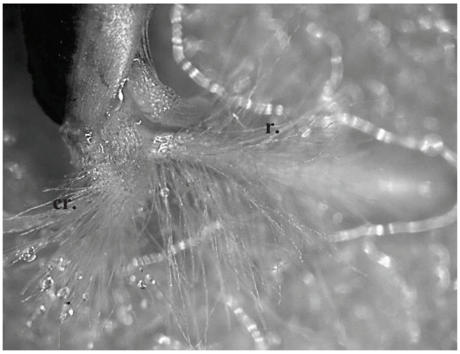

Figure 7. Phase of the germination process;
cr - coleorhiza; r - radicle

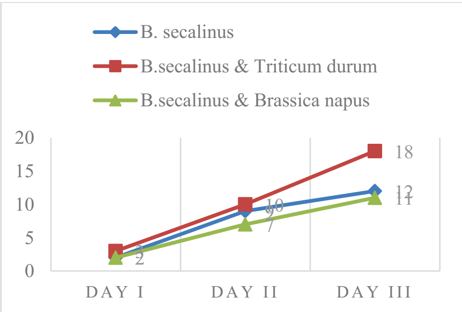

Figure 8. The numbers of *Bromus secalinus* germinated caryopses in three days of experiment (repetition I: 03. 13 – 03. 15. 2018)

The rate of the germination process was different in the three variants, indicating the presence of allelopathy between germinated seeds. Figures 8-10 shows that the germination of seeds of *B. secalinus* is strongly stimulated by the presence of durum wheat caryopsis while *Brassica napus* seeds reduce the number of *B. secalinus* germinated caryopsis. On the other hand, it can be observed, from the explications below, that the rate of germination of the seeds of *Brassica napus* was not influenced by the presence of *B. secalinus* germinated caryopsis whereas the germination of the caryopsis of *Triticum durum* has been influenced.

On the third day of the first repetition had germinate (Figure 8):

- 12 out of 20 caryopsis of *B. secalinus* in the first variant;
- 18 out of 40 caryopsis of *B. secalinus* as well as 8 out of 20 caryopsis of *Triticum durum* in the second variant;
- 11 out of 40 caryopsis of *B. secalinus* as well as 18 out of 20 seeds of *Brassica napus* in the third variant.

On the second repetition, on the third day, the rate of germination was (Figure 9):

- 14 out of 20 caryopsis of *B. secalinus* in the first variant;
- 25 out of 40 caryopsis of *B. secalinus* as well as 14 out of 20 caryopsis of *Triticum durum* in the second variant;
- 14 out of 40 caryopsis of *B. secalinus* as well as 20 out of 20 seeds of *Brassica napus* in the third variant.

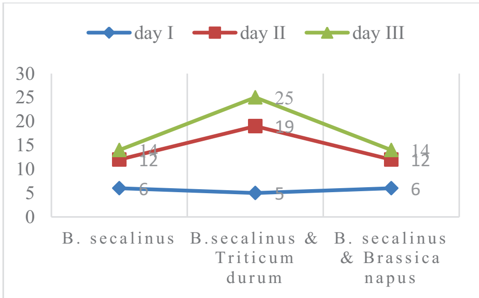

Figure 9. The numbers of *Bromus secalinus* germinated caryopses in three days of experiment (repetition II: 03. 20- 03. 22. 2018)

On the third day of the last repetition had germinate (Figure 10):

- 16 out of 20 caryopsis of *B. secalinus* in the first variant;
- 25 out of 40 caryopsis of *B. secalinus* as well as 15 out of 20 caryopsis of *Triticum durum* in the second variant;
- 25 out of 40 caryopsis of *B. secalinus* as well as 20 out of 20 seeds of *Brassica napus* in the third variant.

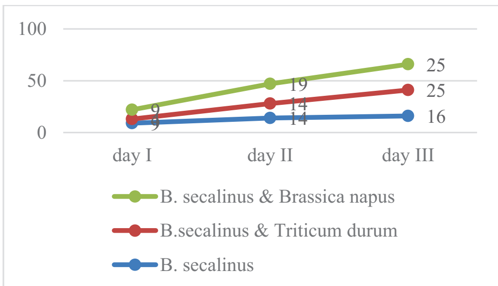

Figure 10. The numbers of *Bromus secalinus* germinated caryopses in three days of experiment (repetition III: 03. 27- 03. 29. 2018)

The role of this type of experiment is to point out the nature of relationships established between plants in an agricultural biocoenosis; also, it can contribute to an appropriate management practice (Penescu & Ionescu, 2013).

CONCLUSIONS

The soil and climate of the Pitesti Station favoured the development of *Bromus secalinus* populations, the size of their caryopsis being close to the upper limit of the length range.

The populations of *B. secalinus* from Pitești are included in the var. *secalinus*, f. *elongatus* and form flexible and slightly divergent awns.

B. secalinus is a species with hairy coleorhiza noticed at the beginning of the germination process.

The germination rate of the *B. secalinus* seeds was influenced by the presence of exudates from the root of the plant species grown together.

ACKNOWLEDGEMENTS

The experiments were carried out in the Research Centre for Studies of Food Quality and Agricultural Products from UASVM Bucharest, Laboratory of microscopy and plant anatomy.

REFERENCES

Adamczewski, K., Kaczmarek, S., Kierzek, R.,Urban, M. (2015). Germination biology and weed thresholds of rye brome (*Bromus secalinus* L.) in wheat (*Triticum aestivum* L.). *Pak. J. Agri. Sci.*, *52*(4), 989–995.

Anghel, Gh., Chirilă, C., Ciocârlan, V., Ulinici, A. (1972). *Buruienile din culturile agricole și combaterea lor. Bucharest*, RO: Ceres Publishing House.

Chistyakova, N.S., Larina, N.P., Rachenko, M.A. (2016). Ecological-biological Aspects of Stipa krylovii Roshev Adaptation at the Initial Stages of Ontogenesis. *Journal of Stress Physiology & Biochemistry*, *12*(3).

Ciocârlan, V., Berca, M., Chirilă, C., Coste, I., Popescu, Gh. (2004). *Flora segetală a României.* Bucharest, RO: Ceres Publishing House.

Ciocârlan, V. (2009). *Flora ilustrată a României*. Bucharest, RO: Ceres Publishing House.

Ciocârlan, V., Chirilă, C. (1982). *Determinatorul buruienilor din culturile agricole*. Bucharest, RO: Ceres Publishing House.

Cussans, G.W., Cooper, F.B., Davies, D.H.K., Thomas, M.R. (1994). A survey of the incidence of the Bromus species as weeds of winter cereals in England, Wales and parts of Scotland. *Weed Res*., *34*, 361–368.

Häfliger, E., Scholz, H. (1981). *Grass Weeds 2*, Basel, Sw: Documenta Ciba-Geigy.

Debaene-Gill, S.B., Allen, P.S., Gardner, J.S. (1994). Morphology of the perennial ryegrass (*Lolium perenne; Poaceae*) coleorhiza and emerging radicle with continuous or discontinuous hydration. *American Journal of Botany*, *81*(6), 739–744.

Hauhouot-O'Hara, M., Criner, B.R., Brusewitz, G.H., Solie, J.B. (2000). Selected Physical Characteristics and Aerodynamic Properties of Cheat Seed for the Separation from Wheat. *Agricultural Engineering International: the CIGR Journal of Scientific Research and Development*, *II*, 1–14.

Ionescu, N., Georgescu, M.I., Săvulescu, E., Penescu, A., Lazăr, A. (2017). Fruits variability of Hibiscus trionum L. weed. *Scientific Papers. Serie A. Agronomy*, *LX*, 479–485.

Ionescu, N., Georgescu, M.I., Penescu, A., Savulescu, E., Voica, M., Lazar, A. (2018). Variation of current morphological characters in winter barley, *Hordeum vulgare* L. *Scientific Papers.Series A. Agronomy*, *LXI* (1), 255–262.

Kapeluszny, J., Haliniarz, M. (2007). Selected elements of germination biology of flaxweed (*Descurainia sophia* Webb. ex Plantl.) and rye brome (*Bromus secalinus* L.). *Annales Universitatis Mariae Curie-Skłodowska. Sectio E, Agricultura*, *62*(2), 226–233.

Massimi, M. (2018). Impact of seed size on seeds viability, vigor and storability of *Hordeum vulgare* (L.). *Agric. Sci. Digest.*, *38*(1), 62–64.

Morita, O., Mitsuishi, S., Goto, M., Fukuda, N. (1990). The germination and establishment of surface-sown grass seeds. 4. Variations in adhering strength of coleorhiza hairs and germination behavior of grass species. *Nippon Sochi Gakkaishi Journal of Japanese Society of Grassland Science*, *36*(1), 1–8.

Nishimura, M. (1922). On the Germination and the Polyembryony of Poa pratensi*s* L. *Shokubutsugaku Zasshi*, 36, 425, en47-en54_1.

Norstog, K.J. (1955). Responses of the oat coleorhiza to various treatments in culture. *The Ohio Journal of Science*, *55*(6), 340–342.

Penescu, A., Ionescu, N. (2013). *Combaterea biologică a buruienilor*. Bucharest, RO: Ceres Publishing House.

Rzymowska, Z., Skrzyczyńska, J., Affek-Starczewska, A. (2010). Occurrence and some morphological features of Bromus secalinus L. in agrocenoses of the Podlaski Przełom Bugu mesoregion. *Fragmenta Agronomica*, *27*(2), 102–110.

Săvulescu T. (coordonator) (1972). *Flora RSR*, vol. XII. Bucharest, RO: Academiei Publishing House.

Skrajna, T., Kubicka, H., Rzymowska, Z. (2012). Phenotypic variation in relation to seed storage

protein polymorphism in *Bromus secalinus* L. (*Gramineae*) populations from north-eastern Poland. *Polish Journal of Ecology*, *60*(1), 41–55.
Stace, C. (2011). *New flora of the British Isles.* Cambridge, UK: University Press Publishing House.
Szymankiewicz, K., Jankowska, D., Deryło, S. (2003). Wpływ płodozmianu i monokultury oraz sposobu uprawy roli na bioróżnorodność flory zachwaszczającej pszenżyto ozime. *Acta Agrophysica*, *1*(4), 746–772.
Warcholińska, A.U. (1981). Flora segetalna Wzniesień Łódzkich. *Acta Universitatis Lodziensis*, *Folia Botanica, 1*. 133–179.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

EFFECT OF SILICON ON THE ACTIVITY OF ANTIOXIDANT ENZYMES AND THE PHOTOSYNTHETIC RATE OF CUCUMBER UNDER MITE INFESTATION

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Abstract

The aim of the study was to evaluate the effect of silicon on the activity of antioxidant enzymes and the photosynthetic rate of cucumber plants (Cucumis sativus L.) cv. Gergana under mite infestation. Four variants consisting of 1 – control, 2 – mite-infested plants, 3 – silicon-treated plants, and 4 – mite-infested plants, treated with silicon, were analyzed. Plants were grown hydroponically and silicon (Si) was applied to the nutrient solution in form of Na_2SiO_3. The plants have been infested by the two-spotted spider mite Tetranychus urticae and 20 days later – analyzed. The results show a decrease in the mite population in the Si-treated plants. The activities of guaiacol peroxidase (GPOD) and syringaldazine peroxidase (SPOD) in the leaves of the mite infested plants increased by 175% and 197% respectively, and the net photosynthetic rate (P_N) in the mite-infested plants was reduced by 51%, compared to the control. It was established that Si supply to a nutrient solution of the mite-infested plants decreases the activities of GPOD and SPOD by 62% and 57% respectively and enhances the net photosynthetic rate by 84%.

Key words*: cucumber, mites, photosynthesis, silicon, stress.*

INTRODUCTION

Silicon (Si) is the second most abundant element on Earth's crust after Oxygen. It has not been considered as an essential element for higher plants, but Si has been proved to be beneficial for the healthy growth and development of many plant species (Epstein, 1999; Ma et al., 2001). The beneficial effects of Si are particularly distinct in plants exposed to abiotic and biotic stresses (Ma, 2004). Over last two decades, more extensive and intensive studies have been performed aiming at better understanding of the possible mechanisms for Si-enhanced resistance and/or tolerance of higher plants to both abiotic and biotic stress (Liang et al., 2003). There are, however, a number of studies of its effects on some phytopathogens – mainly powdery mildew (Shetty et al., 2012). There are also some researches on the effect of Si in insect-induces stress conditions. The increased resistance to herbivore pests caused by silicon treatment has been reported in various sensitive varieties of cereal plants (Keeping & Meyer, 2006; Hou & Han, 2010; Sidhu et al., 2013; Han et al., 2015) and cereal grasses (Massey et al., 2006; Massey & Hartley, 2009).
Higher silicon concentration in the soil or in the nutrient medium causes a decrease in the number of insect and non-insect pests of crop plants (Liang et al., 2005). The positive effect of silicon application has been reported for various pests including *Chilo suppresalis*, *Scirpophaga incertulas*, *Chlorops oryzae*, *Nephotettix bipunctatus cinticeps*, *Nilaparvata lugens*, Spider mites (*Tetranychus* spp.) (Gatarayiha et al., 2010) or mites (Savant et al., 1997; Nikpay et al., 2014).

The two-spotted spider mite *Tetranychus urticae* Koch (Acari: Tetranychidae) is considered one of the most harmful pests in many crops. It is herbivore, with a broad food specialization, developing on over 1200 plant species belonging to more than 140 different families (Van Leeuwen et al., 2010). Small size, rapid development, high reproductive potential, large number of generations, and optional diapause allow the mite to multiply for a short time in high density when the conditions of the environment are favourable. This is accompanied by a rapid decline in the quality of the host plant. During feeding, the mite mechanically damages the leaf epidermis, resulting in a deterioration of the water regime of plants and decrease of the photosynthetic apparatus productivity (changes in CO_2-gas exchange, reduction of chlorophyll content), which can cause a fall of undeveloped blossoms and even death of young plants (Park & Lee, 2002).

Most studies have focused on the effect of silicon on the pathogen or on an enemy and their lifecycle and treatment behaviour. But the information about the effect of Si on the physiological and biochemical changes occurring in the host plant and the involved mechanisms are scarce. This motivated us to conduct this study and gain insight into some of the benefits and effects of Si application.

MATERIALS AND METHODS

Laboratory experiments were performed under controlled environmental conditions in a laboratory at the Department of Entomology, Agricultural University - Plovdiv, Bulgaria: photoperiod 14/10 h (light/dark), photosynthetically active radiation (PhAR) 250 μmol m^{-2} s^{-1}, air temperature 25°C (day)/ 22°C (night) and relative air humidity 50 ± 5%. The seeds were subjected to the process of imbibition for 24 h. They were then placed in inert material (perlite). After the full development of the cotyledons and the appearance of the first true leaf, the young plants were transferred in plastic containers with nutrient solution. Plants of all variants were grown in ½ Hoagland nutrient solution containing all the necessary macro- and micro elements to phase third true leaf and then infested by *Tetranychus urticae*. At the same time started the treatment with Si in form of Na_2SiO_3 to the nutrient solution. The nutrient solution was replaced every week with a fresh one. Experimental plants were grouped in 4 variants: 1 - control plants; 2 - mite-infested plants; 3 - plants + 1.5 mM Si; 4 - mite-infested plants + 1.5 mM Si. Each variant includes 24 test plants. Twenty days after infestation plants were analyzed.

Mite infestation

All experimental plants were infested on the 15th day of their development with 50 moving stages of the two-spotted spider mite. The infestation was carried out with mobile stages of a laboratory grown mite population. For this purpose, leaf cuts of about 5 cm^2 were examined under a stereomicroscope to ensure the presence of the Tetranychid mite only and to remove other mite families (if any). The number of *T. urticae* mobile forms was recorded and controlled and 10 or 20 mites (mostly adult females) were left on each such cut. The examined leaf cuttings were placed on the 2nd and/or 3rd leaf of each cucumber plant (50 mites/plant respectively).

Methods of analysis

Leaf area

The leaf area of the plants was determined by the electronic digital plotter NEO-2 (TU-Sofia, Bulgaria) (Kerin et al., 1997).

Leaf gas exchange parameters

Leaf gas exchange. The index of the leaf gas exchange rate - net photosynthetic rate (P_N) rate, transpiration intensity (E), stomatal conductance (gs) and intracellular concentration of CO_2 (ci) were determined with LCpro+ portable photosynthetic system [Analytical Development Company Ltd., Hoddesdon, England]. Measurements were performed under the following conditions: photosynthetically active radiation (PhAR) of 500 μmol m^{-2} s^{-1}, temperature 25ºC and natural external CO_2 concentration of about 400 vpm.

Photosynthetic pigments

The photosynthetic pigments content was determined spectrophotometrically by the Lichtenthaler method (1987) and was recalculated to a unit of fresh weight (mg g^{-1} FW).

Lipid peroxidation

The lipid peroxidation rate (LP) was determined by the method of Heath and Packer (1968). Specific absorbance at 532 and non-specific at 600 nm were recorded at molar extinction coefficient E = 155 mM^{-1} cm^{-1}. The results are presented as malondialdehyde (MDA) content in a unit of fresh weight (nmol MDA g^{-1} FW).

Enzyme extraction

The fresh plant sample (0.5 g) was ground with 5 ml ice-cold extraction buffer (pH 7.8), quartz sand and 200 mg polyvinylpyrrolidone (PVP).

Samples were centrifuged at 4°C for 10 min at 13500 rpm. The resulting clear supernatant was immediately used for analysis.

Guaiacol Peroxidase (GPOD)

The activity of GPOD was determined spectrophotometrically by the method of Bergmeyer et al. (1974). The mixture of 2.3 ml of KH_2PO_4 buffer (pH 7.0), 300 µl of H_2O_2, 300 µl of 8 mM guaiacol and 100 µl of extract was placed in the cuvette. The absorbance was measured at 436 nm against a blank containing the same components without enzyme extract ($E = 26.6\ mM^{-1}\ cm^{-1}$). The values obtained were expressed as U mg g^{-1} FW.

Syringaldazine peroxidase (SPOD)

The activity of SPOD was determined spectrophotometrically by the method of Imberty et al. (1985). In a cuvette were placed 2.55 ml of 0.1 M Tris-HCl buffer (pH 7.5), 300 µl of 10 mM H_2O_2, 50 µl of 3.5 mM syringaldazine, and 100 µl of the extract. The absorbance was measured at 530 nm against a blank containing the same components without enzyme extract ($E = 27\ mM^{-1}\ cm^{-1}$). The values obtained were expressed as U mg g^{-1}FW.

Statistical data processing

The data are presented as mean ± SD of 3 replicates. The experimental results were statistically processed with the SPSS program using a one-way ANOVA dispersion analysis and Duncan's comparative method, with the validity of the differences determined at a 95% significance level. The different letters (a, b, c, d) after the mean value show statistically significant differences between the variants.

RESULTS AND DISCUSSIONS

At the end of the experiment in order to evaluate the effect of Si application, the population density of the two-spotted mite was estimated. It was found that the number of all the mobile stages of the mite was significantly reduced on the leaves of Si-treated plants compared to those which were infested with mites, but untreated with silicon. In larval stages the decrease was about 60%, and the number of adults was reduced by 24%. The decrease in the number of eggs was the lowest - by about 14% (Table 1). Our results are in line with the ones obtained by Nikpay and Nejadian (2012), who reported a decrease in the number of yellow mite *Oligonychus sacchari* in sugar cane. The authors investigated the impact of various Si-containing fertilizers on the yellow mite in four cane varieties and observed a population density on the 10^{th}, 20^{th}, 30^{th} and 40^{th} days after infection. Although they did not observe significant differences among the variants before the 20^{th} day after the infestation, they suggest that the silicon supply is able to decrease the population density of the mite after that period. They measured a less number of mites at the 30^{th} and the 40^{th} day after the infestation, what they associate with the high resistance of this mite species and the need for a longer period for population breeding, as reported in their previous studies (Nikpay et al., 2011; 2012). Our results show a significant decrease in the number of the pest even earlier - on the 20^{th} day after infestation.

Table 1. Number of eggs and mobile stages of *T. urticae* on leaves of cucumber (*Cucumis sativus* L.), cv. Gergana, 20 days after mite infestation

Parameter/Variant	Plants infested with mite	Plants infested with mite + Si
Eggs	208.75	177.25
Larvae	23	9.12
Adults	38.66	29.37

According to some authors higher silicon concentration in the soil or in the nutrient medium causes a decrease in the number of insect and non-insect pests of crop plants (Liang et al., 2007). Although the mechanisms of silicon`s action are not quite clear, there are several theories. Toledo and Reis (2018) examined the effect of foliar spraying with potassium silicate on coffee plants infested with red mite *Oligonychus ilicis*. The authors report about a reduced population of red mite and suggest that this effect may be due to chemical and physical changes in the plant tissues. They also observed the increased content of lignin and tannins which probably makes plant tissues less attractive to herbivores because of increased hardness and toxic compounds in the plant tissues. About the same effect of the silicon treatment reported Yavas and Unay (2017) in their review article. They suggest that Si could be used to improve plant growth and resistance under stress conditions, as it acts as a mechanical barrier for pests and diseases and prevents of oxidative stress via enzyme activation.

In their research Gatarayiha et al. (2010) did not observe a direct effect of Si on *T. urticae* mortality, but they established that the Si-supply improves the action of the biological agent *Beauveria bassiana*. They suggest that silicon increases the resistance to spider mites by disturbing their feeding and making them more vulnerable to infestation by the fungus *Beauveria bassiana*. The reduced population density is probably due to the increased hardness of the leaf tissues (epidermal cells), as the silicon is deposited in the cell walls (Ma, 2004; Reynolds et al., 2009), what affects the feeding of the mites. He et al. (2015) found that increased levels of silicic acid in plants shortened the stay of leaf-hoppers on the plants and reduces the pest fertility. Kvedaras et al. (2007) suggest that the positive action of silicon enhances in drought conditions. The authors observed that after treating dried sugar cane with silicon, the content of the element in the stem increased and the number of *Eldana saccharina* (Lepidoptera: Pyralidae) and stalk damage decreased. In the case of application on soil or foliar treatment with silica in the form of $CaSiO_3$, the mortality of *Bemisia tabaci* nymphs was increased (Correa et al., 2005). In another study feeding of lice with Silicon reduces their number in the infested plants (Moraes et al., 2004; Gomes et al., 2005). It is reported that pests with piercing-sucking mouth organs and herbivore caterpillars prefer silicon-poor tissues to those with high silicon content. This element reduces feeding, growth, fertility, and the population of *Sogatella frucifera* (Salim & Saxena, 1992). Silicon reduces the reproductive capacity of the phloem-feeding species (*Myzus persicae*) on potatoes and whitefly (*Bemisia tabaci*) in cucumber (Reynolds et al., 2009).

In order to evaluate the stress levels, induced by the mite feeding, the rate of the lipid peroxidation was measured. Malondialdehyde (MDA) is an indicator of the lipid peroxidation rate (LP) and its concentration is related to the degree of the membrane damages (Hsu & Kao, 2007). The analysis of LP shows strong damage of the cell membranes in the leaves of the infested plants (Figure 1).

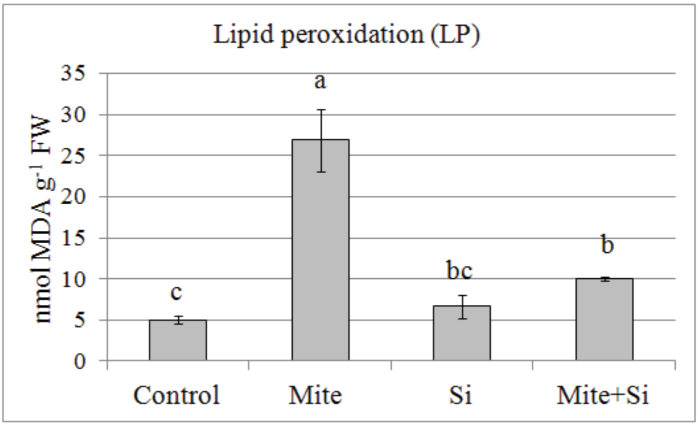

Figure 1. Lipid peroxidation rate (LP) nmol MDA g^{-1} FW in cucumber leaves 20 days after mite infestation

In the stressed plants, MDA content increased more than four times compared to the control. The addition of Si to the nutrient solution of the stressed plants causes a decrease in the MDA content by about 63% compared to the mite-infested plants. Sivritepe et al. (2009) also reported an increase in the LP rate in the leaves of grapevine plants, infested by the two-spotted spider mite. According to Tehri et al. (2014), there is a significant relationship between the resistance of the experimental plants and the LP rate in the leaves. The LP rate increases in parallel with increasing the population of *T. urticae* and is initially higher in the leaves of the resistant variety. According to the authors, a short period of mite feeding on the resistant variety induces subsequent resistance. In another experiment, rice plants were infected with *Pyricularia oryzae* and the authors also reported an increase in MDA content in the leaves of the infected plants (Domiciano et al., 2015). They also observe a decrease in its concentration after treatment of the infected plants with silicon.

The feeding of the two-spotted spider mite significantly increased also the activity of the enzymes of the antioxidant defense system (Figure 2).

The activity of guaiacol peroxidase (GPOD) in the leaves of the infested plants was 175% higher than in the control plants. The Si-supplied plants demonstrate a strong recovery potential. Silicon supply decreases the level of stress and the activity of the guaiacol peroxidase was only 4.31% higher than in the control.

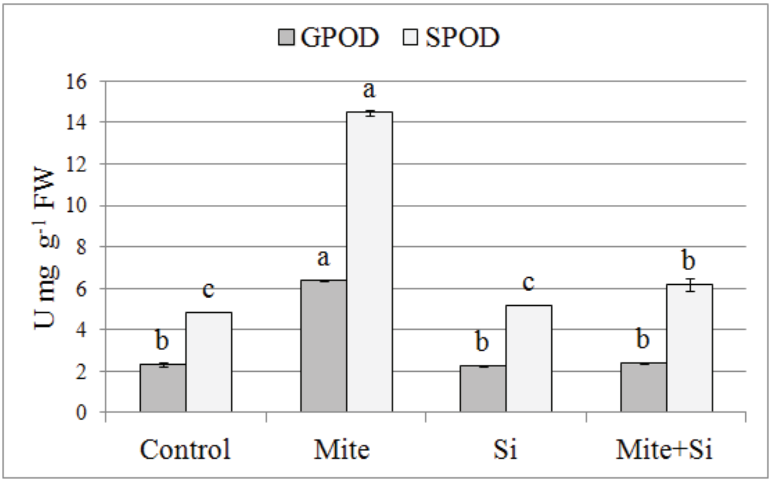

Figure 2. Activity of GPOD and SPOD in cucumber leaves 20 days after mite infestation

Similarly, the activity of syringaldazine peroxidase (SPOD) followed the same tendency. Mite feeding caused a strong increase in SPOD activity by almost 200%. The addition of Si to the nutrient medium of the infested plants leads to a decrease in enzyme activity by 57% compared to the mite-infested, Si-untreated plants.

The mite-induced stress affects negatively the synthetic processes and the biomass accumulation in the infected plants. The data about the effect of the spider mite feeding on the biomass accumulation are presented in Table 2. The mite-infested plants were with reduced growth of all organs. The fresh leaf mass, as well as the fresh mass of the whole plants, were strongly inhibited (by over 60% relative to the control). There was also a decrease in the dry mass of the infected plants, which was more pronounced for the shoots compared to the roots. This effect is explicable as the enemy develops and causes direct damage only to the shoots of the plants, and not to the roots. The leaf area was also significantly reduced in the mite-infested plants compared to the control (by 54%). The Si supply to the nutrient medium of the infested plants resulted in an obvious recovery - the leaf area of these plants was enhanced by 78% in comparison to the infested untreated with silicon plants.

Table 2. Changes in biomass accumulation in cucumber 20 days after infestation with *T. urticae*

Variant	[1]FW Plant (g)	FW Leaves (g)	FW Stems (g)	FW Root (g)	[2]LA (cm^2)	[3]DW Plant (g)	DW Leaves (g)	DW Stems (g)	DW Root (g)
Control	21.072^a	11.411^a	6.848^a	2.813^a	599^a	1.264^a	0.96^a	0.222^a	0.081^a
Mite	8.069^c	4.235^c	2.608^d	1.226^b	274^c	0.679^b	0.525^b	0.102^c	0.051^b
Si	20.701^a	11.619^a	6.104^b	2.976^a	655^a	1.259^a	0.948^a	0.21^a	0.101^a
Mite+Si	15.406^b	8.555^b	2.031^{ab}	2.031^{ab}	488^b	1.105^a	0.859^a	0.166^b	0.078^a

[1]FW = fresh weight; [2]LA = leaf area (cm^2); [3]DWpl = dry weight

The increased photosynthetic area was probably responsible for the enhancement of the net photosynthetic rate. The analysis of the net photosynthetic rate is presented in Figure 3. The feeding of the mites reduced the value of P_N by 51%. Plants of Mite+Si variant showed a strong recovery - an increase of 84%.

This suggests that the use of silicon helps infected plants to recover to some extent from the negative effects of mite feeding and to increase their assimilation ability.

A decrease in the rate of P_N in mite-infested plants has also been reported by Sivritepe et al. (2009) in grapevine. Alatawi et al. (2007) also reported a reduced level of CO_2. They suggest that the rate of the damage is in line with the duration of the feeding period and population density of the mite.

Our results are consistent with the reports of Domiciano et al. (2015) who examined the effect of silicon on rice plants infected with the *Pyricularia oryzae* pathogens.

The authors observed a significant decrease in photosynthesis (P_N) in infected plants.

After the addition of Silicon to the nutrient medium of the infected plants, the authors report about a significantly lower decrease in the value of the indicator compared to the phytopathogen-infected plants, but not treated with silicon.

In order to evaluate if the effect of the mite-induced damage and the Si-treatment is related also to stomatal changes, we measured the rate of the transpiration and the stomatal conductance of the test plants.

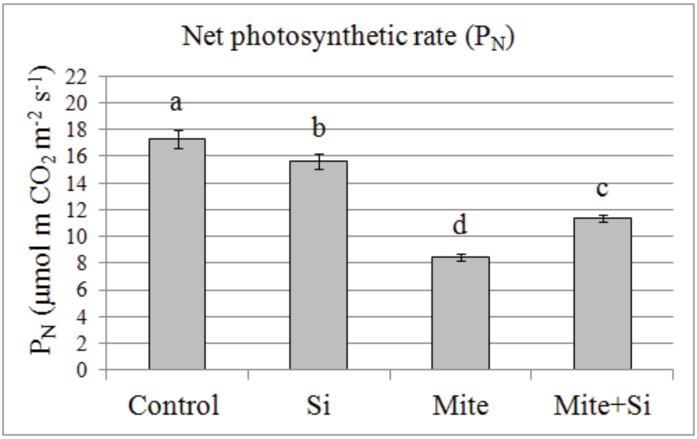

Figure 3. Net photosynthetic rate P_N (µmol CO_2 m^{-2} s^{-1}) in leaves of cucumber (*Cucumis sativus* L.) 18 days after infection with *T. urticae*

Figure 4 presents the data on the transpiration rate. In the plants infested by mites, the process was highly inhibited. The decrease was by 46% relative to the control. After the addition of Si to the nutrient solution of the infested plants, a similar increase (47%) was observed compared to the plants of the mite-infested variant.

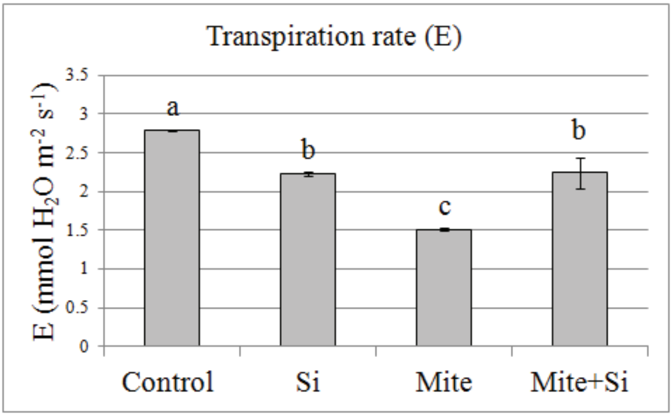

Figure 4. Transpiration rate E (mmol H_2O m^{-2} s^{-1}) in cucumber leaves 20 days after infestation with *T. urticae*

During their feeding, the mites mechanically damage the epidermis including the stomata, and the cells lose their ability to open and to close under control. Depending on the degree of attack and the duration of mite feeding, the intensity of transpiration varies greatly. In the first days after the mite infestation, there is usually an increase in the intensity of transpiration. In a severe and prolonged attack transpiration begins to decrease sharply over time. This is observed during the day with regard to stomatal transpiration. During the night, cuticular transpiration increases due to mechanical damage from the mite feeding (Sivritepe et al., 2009). This causes large losses of water from plant tissues and in case of a strong attack leads to a state of permanent water deficit. The analysis of the stomatal conductance is presented in Figure 5. The stomatal conductance in the leaves of the mite infested plants was 64% lower than in the control. The Si-supplied infested plants had two times higher stomatal conductance than the untreated mite infested plants. It shows that the effect of the Si supply on the photosynthesis is related to stomatal changes.

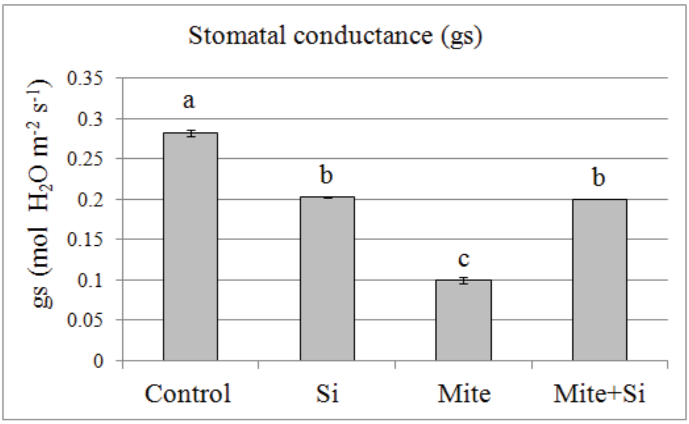

Figure 5. Stomatal conduction gs (mol H_2O m^{-2} s^{-1}) in cucumber leaves 20 days after infestation with *T. urticae*

In order to clarify if the reason for the enhanced net photosynthesis is due only to physiological changes of the leaf anatomy and function, the content of the main photosynthetic pigments was also measured (Table 3). The content of chlorophyll *a* was almost twice reduced in the leaves of the stressed plants. The amounts of chlorophyll *b* and carotenoids were also lower, by 39 and 43%, respectively. In the leaves of the Si-supplied plants, the values of these parameters increased. Chlorophyll *a* content was 45% higher and the increase of the chlorophyll *b* and carotenoids content were by 49 and by 60%, respectively. There is no information about the effect of Si on the pigment content in mite-induced stress conditions, but there are several studies about its positive influence in various abiotic stress conditions like drought (Maghsoudi et al., 2016) or salinity (Rezende et al., 2018). The authors suggest that silicon is able to alleviate the negative effect of low salt concentrations on the photosynthetic pigment content via reducing the production of reactive oxygen species (ROS) or via anatomical changes of the photosynthetic apparatus.

Table. 3. Content of the main photosynthetic pigments (mg g^{-1} FW) in the cucumber leaves 20 days after infestation and treatment with Si

Variant	[1]Chl *a*	Chl *b*	Chl (*a*+*b*)	Carotenoids	Chl *a/b*	Chl (*a*+*b*)/ carotenoids
Control	2.24^{a}	0.77^{a}	3.02^{a}	0.79^{a}	2.91^{b}	3.82^{b}
Mite	1.16^{d}	0.47^{d}	1.63^{d}	0.45^{d}	2.46^{c}	3.63^{c}
Si	1.9^{c}	0.63^{c}	2.53^{c}	0.66^{c}	3.02^{a}	3.83^{b}
Mite+Si	2.13^{b}	0.7^{b}	2.83^{b}	0.72^{b}	3.04^{a}	3.93^{a}

[1]Chl = chlorophyll

CONCLUSIONS

The application of silicon in the form of Na_2SiO_3 in cucumber is able to alleviate the negative effects of spider mite feeding. The results obtained show that Si treatment reduces the population of the spider mite and enhances the net photosynthetic rate. This positive effect is probably due to the increased amount of the main photosynthetic pigments and the protective influence on the cell membranes and the antioxidant defense system. The mechanisms involved are not quite clear. The protective role may be related to the accumulation of silicon in the cell walls of the plants, which makes the tissues harder and less attractive to the pest, as well as to its stimulating effect on the photosynthetic activity of the plants, expressed by improved parameters of the leaf gas exchange, high content of photosynthetic pigments, and an increase in biomass and leaf area of the analyzed plants.

ACKNOWLEDGMENTS

This study was funded by the Research Fund of the Ministry of Education and Science. Project number H16/35 - Agrobiological study on biostimulants and inorganic products for organic control in vegetable crops under stress conditions.

REFERENCES

Alatawi, F., Margolies, D., Nechols, J. (2007). Aesthetic damage thresholds for two spotted spider mites (Acari: Tetranychidae) on impatiens: effect of plant age and level of infestation. *Journal of Economic Entomology*, *100*. 1904–1909.

Bergmeyer, H., Gawehn, K., Grassl, M. (1974). *Enzymes as biochemical reagents.* In: Bergmeier HU (ed) Methods in Enzymatic Analysis. Academic Press, New York. NY.

Correa, R., Moraes, J., Auad e Geraldo, A., Carvalho, A. (2005). Silicon and acibenzolar-s-methyl as resistance inducers in cucumber, against the whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) *Biotype B. Neotropical Entomology*, *34*. 429–433.

Domiciano, G., Cacique, I., Chagas Freitas, C., Filippi, M., DaMatta, do Vale, F., Rodrigues, F. (2015). Alterations in gas exchange and oxidative metabolism in rice leaves infected by *Pyricularia oryzae* are attenuated by silicon. *Phytopathology*, *105*. 738–747.

Epstein, E. (1999). Silicon. *Annual Review in Plant Physiology and Plant Molecular Biology*, *50*. 641–664.

Gatarayiha, M., Laing, M., Miller, M. (2010). Combining applications of potassium silicate and *Beauveria bassiana* to four crops to control two spotted spider mite. *Tetranychus urticae* Koch. *International Journal of Pest Management*, *56*. 291–297.

Gomes, F., de Moraes, J., dos Santos, C., Goussain, M. (2005). Resistance induction in wheat plants by silicon and aphids. *Scientia Agricola*, *62*. 547–551.

Han, Y, Lei, W., Wen, L., Hou, M. (2015). Silicon-mediated resistance in a susceptible rice variety to the rice leaf folder *Cnaphalocrocis medinalis* Guenée (Lepidoptera: Pyralidae). *PLoS One,* 10, 0120557.

Heath, R., Packer, L. (1968). Photoperoxidation in isolated chloroplasts. I. Kinetics and stoichiometry of fatty acid peroxidation. *Archives of Biochemistry and Biophysics*, *125*. 189–198.

Hou, M., Han, Y. (2010). Si-mediated rice plant resistance to the Asiatic rice borer: effects of silicon amendment and rice varietal resistance. *Journal of Economic Entomology*, *103*. 1412–1419.

Hsu, Y., Kao, C. (2007). Cadmium-induced oxidative damage in rice leaves is reduced by polyamines. *Plant and Soil*, *291*. 27–37.

Imberty, A., Goldberg, R., Catesson, A. (1985). Isolation and characterization of *Populus isoperoxidases* involved in the last step of lignification. *Planta*, *164*. 221–226.

Keeping, M., Meyer, J. (2006). Silicon-mediated resistance of sugarcane to *Eldana saccharina* Walker (Lepidoptera: Pyralidae): effects of silicon source and cultivar. *Journal of Applied Entomology, 130*. 410–420.

Kerin, V., Berova, M., Stoeva, N., Vassilev, A., Petkova, H. (2000). *Manual of Laboratory of Plant Physiology*. Academic Publishing of VSI Plovdiv.

Kerin V., Berova, M., Vasilev, A., Zlatev, Z. (1997). *Modern methods of analysis in plant physiology.* Academic Publishing of VSI Plovdiv.

Kvedaras O., Keeping M., Goebel, F., Byrne, M. (2007). Water stress augments silicon mediated resistance of susceptible sugarcane cultivars to the stalk borer *Eldana saccharina* (Lepidoptera: Pyralidae). *Bulletin of Entomological Research*, *97*. 175–183.

Liang, Y., Sun. W., Zhu, Y., Christie, P. (2007). Mechanisms of silicon mediated alleviation of abiotic stress in higher plants: a review. *Environmental Pollution*, *147*(2), 422–428.

Liang, Y., Sun, W., Si, J., Römheld, V. (2005). Effects of foliar- and root-applied silicon on the enhancement of induced resistance to powdery mildew in *Cucumis sativus*. *Plant Pathology*, *54*. 678–685.

Liang, Y., Chen, Q. Liu, Q., W Zhang, W., R Ding, R. (2003). Exogenous silicon (Si) increases antioxidant enzyme activity and reduces lipid peroxidation in roots of salt-stressed barley (*Hordeum vulgare* L.). *Journal of Plant Physiology*, *160*. 1157–1164.

Lichtenthaler H. (1987). Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. Methods in Enzymology, *148*. 350–382.

Ma, J., Miyake, Y., Takahashi, E. (2001). Silicon as a beneficial element for cropplants. In: Datnoff. L., Snyder, G., Korndorfer, G., (Eds.). Silicon in Agriculture. *Studies in Plant Science*, *8*. 17–39.

Ma, C., Li, Q., Gao, Y., Xin, T. (2004). Effect of Silicon Application on drought resistance of cucumber plants. *Soil Science and Plant Nutrition*, *50*(5), 623–632.

Maghsoudi, K., Emam, Y., Pessarakli, M. (2016). Effect of silicon on photosynthetic gas exchange, photosynthetic pigments, cell membrane stability and relative water content of different wheat cultivars under drought stress conditions. *Journal of Plant Nutrition*, *39*. 1001–1015

Moraes, J., Goussain, M., Basagli, M., Carvalho, G., Ecole, C., Sampaio, M. (2004). Silicon influence on the tritrophicinteraction: wheat plants, the green bug *Schizaphis graminum* (Rondani) (Hemiptera: Aphididae), and its natural enemies, *Chrysoperla externa* (Hagen) (Neuroptera: Chrysopidae) and *Aphidius colemani* Viereck (Hymenoptera: Aphidiidae). *Neotropical Entomology*, *33*. 619–624.

Massey, F., Ennos, A., Hartley, S. (2006). Silica in grasses as a defence against insect herbivores: contrasting effects on folivores and a phloem feeder. *Journal of Animal Ecology*, *75*. 95–603.

Massey, F., Hartley, S. (2009). Physical defences wear you down: progressive and irreversible impacts of silica on insect herbivores. *Journal of Animal Ecology*, *78*. 281–291.

Nikpay, A., Nejadian, E. (2014). Field Applications of Silicon-Based Fertilizers Against Sugarcane Yellow Mite *Oligonychus sacchari*. *Sugar Tech.*, *16*(3), 319–324.

Nikpay, A., Arbabi, M., Kahrarian, M. (2011). Evaluation new acaricides in control of sugarcane yellow mite *Oligonychus sacchari* and effect on Stethorus punctillum in south western part of Iran. In Global Conference on Entomology. Chiang Mai: Thailand.

Nikpay, A., Arbabi, M., Kahrarian, M. (2012). Potential application of Neem-Azal against spider mite *Oligonychus sacchari* on two sugarcane commercial cultivars. In 6th World Neem Conference. Nagpur: India.

Park, Y., Lee. J. (2002). Leaf cell and tissue damage of cucumber caused by two spotted spider mite (Acari: Tetranychidae). *Journal of Economic Entomology*, *95*. 952–957.

Rezende, R., Rodrigues, F.A., Soares, J.D.R., Silveira H.R.D.O., Pasqual, M., Dias, G..D.MG. (2018). Salt stress and exogenous silicon influence physiological and anatomical features of in vitro-grown cape gooseberry. *Ciência Rural*, 170–176.

Reynolds, O., Keeping, M., Meyer, J. (2009). Silicon-augmented resistance of plants to herbivorous insects: A review. *Annals in Applied Biology*, 155, 171–186.

Salim, M., Saxena, R. (1992). Iron, silica and aluminium stresses and varietal resistance in rice: effects on white backed planthopper. *Crop Science*, *32*. 212–219.

Savant, N., Snyder, G., Datnoff, L. (1997). Silicon management and sustainable rice production. *Advances in Agronomy*, *58*. 151–199.

Sidhu, J., Stout, M., Blouin, D., Datnoff, L. (2013). Effect of silicon soil amendment on performance of sugar-cane borer, *Diatraea saccharalis* (Lepidoptera: Crambidae) on rice. *Bulletin of Entomological Research*, *103*. 656–664.

Sivritepe, N., Kumral, N., Erturk. U., Yerlikaya. C., Kumral. A. (2009). Responses of grapevines to two spotted spider mite mediated biotic stress. *Journal of Biologycal Sciences*, *9*(4), 311–318.

Shetty, R., Jensen B., Shetty, N., Hansen, M., Hansen, C., Starkey, K., Jorgensen, H. (2012). Silicon induced resistance against powdery mildew of roses caused by *Podosphaera pannosa*. *Plant Pathology*, *61*(1), 120–131.

Tehri, K., Gulati, R., Geroh, M., Madan, S. (2014). Effect of two-spotted spider mite infestation on some biochemical parameters of cucumber leaves. *Annals of Biology, 30*(4), 686–690.

Toledo, M., Reis, P. (2018). Study on potassium silicate spraying in Coffee plants to control *Oligonychus ilicis* (McGregor) (Acari: Tetranychidae). *Advances in Entomology*, *6*. 14–26.

Van Leeuwen, T., Vontas. J., Tsagkaracou. A., Dermauwa. W., Tirry. L. (2010). Acaricide resistance mechanisms in the two-spotted spider mite *Tetranycus urticae* and other important Acari: A review. *Insect Biochemistry and Molecular Biology*, *40*. 563–572.

Yavas, I., Unay, A. (2017). The role of silcon under Biotic and abiotic stress conditions. *Turkish Journal for Agricultural Research*, *4*(2), 204–209.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOME ASPECTS OF MORPHO-ANATOMICAL FEATURES OF THE INVASIVE SPECIES *Eleusine indica* (L.) Gaertn.

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Abstract

Eleusine indica (L.) Gaertn. – goose grass (Poaceae) (syn. Cynosurus indicus L., Eleusine distans Moench, E. gracilis Salisb.) is an invasive species in Romania as well as in several countries in Europe, Asia, Central and South America, the Caribbean and many Pacific islands. In our country this species is found in ruderal locations or alongside town roads. A single plant may produce more than 50,000 small seeds, which can be easily dispersed by wind and water. According to different sources, Eleusine indica (L.) Gaertn. is native to Africa. It is also considered a very extensive plant and often a difficult weed, drought-tolerant and herbicide-resistant. In 2018, Eleusine indica (L.) Gaertn. was studied at the Bucharest University of Agronomic Sciences and Veterinary Medicine. Our study targeted the roots, stems, leaves, inflorescence and caryopsis. Morphological and anatomic analyses were made on the plant material collected from ruderal locations in Bucharest, District 4 (Romania).

Key words: *anatomy, Eleusine indica, morphology, vegetative organs*.

INTRODUCTION

Eleusine indica (L.) Gaertn. - goose grass (Poaceae family), which grows in fields and open grounds and fis ound very often during the rainy season in its native areas. According to different sources, *Eleusine indica* (L.) Gaertn. is originally native to Africa. A single plant may produce more than 50,000 small seeds, which can be easily dispersed and viable seeds persist in the upper soil for only 2-5 years (Standifer, 1984; Rojas-Sandoval & Acevedo-Rodríguez**,** 2014). In our country it is an invasive species developing in ruderal locations. *Eleusine indica* (L.) Gaertn. is a worldwide weed of the tropics and one of the world's serious weedy grasses; at the same time, it has medicinal uses. In the last years there is a continuous enrichment of Romania's flora with new alien plant species (Anastasiu & Negrean, 2006; 2008; Anastasiu et al., 2009; 2011; Oprea & Sârbu, 2010; Sîrbu & Oprea, 2011). According to Anastasiu and Negrean (2005), Romania's alien flora represents some 13% of the country's total flora, which was estimated by Ciocârlan (2009) to 3,335 species; after the year 2000 a number of 47 new alien plant species were recorded in Romania (Sîrbu & Oprea, 2011; Anastasiu & Negrean, 2011). This species was reported in Romania for the first time in Iaşi (Răvăruț & Mititelu, 1960). In our country the presence of this species was also signalled in Crişana, northwestern Romania, (Negrean & Karacsonyi, 1984), Wallachia (Negrean & Constantin, 1999; Oprea et al., 2004), Galați (Sîrbu, 2011), Oltenia (Răduțoiu & Stan, 2013).

Eleusine indica is resistant to herbicides (Valverde et al., 1993; Baird, 1997; Doll, 2000; Jung et al., 2014; Mueller et al., 2011; Vargas et al., 2013; Jalaludin et al., 2010; An Jing et al., 2014; Cha Thye San, 2014; Penescu et al., 2018).

In the native regions *Eleusine indica* is used as a local medicinal plant with antioxidant (Al-Zubairi et al., 2011; Iqbal et al., 2012; Sagnia et al., 2014), antimicrobial (Sagnia et al., 2014) and anticancer (Al-Zubairi et al., 2011; Amarpreet; 2014; Hansakul; 2009) properties. The whole plant, especially the root, is depurative, diuretic, anti inflammatory (De Melo, 2005; Sagnia et al., 2014), febrifuge, laxative and sudorific (Chopra et al. 1956; Nguyen et al., 1989; Nagathein, 1977), antiviral (Iberahim et al., 2016), used for liver disorders and convulsion as well as with antipyretic (Iqbal et al., 2011; Kirtikar & Basu, 1935; Pattanayak et al., 2017) properties. Anatomic studies were carried out by: Babu et al., 2014; Rubina et al., 2007; Hafliger and Schoz, 1981; Bibi et al., 2007; Metcalfe, 1961; Metcalfe and Clifford, 1968; Niteesh Kumar et al., 2017; Saw, 2011. Fahn (1965) studied the stomata of some Poaceae and Cyperaceae members. Numerous reports on foliar anatomy and micro morphology are used for delimiting the different groups or tribes in the Poaceae family (Brown, 1958; Ellis, 1987; Amarasinghe & Watson, 1990; Ahmad et al., 2011).

MATERIALS AND METHODS

The plant and the fresh material, roots, stems, leaves and inflorescence which were used for the macroscopic and micro-morphologic studies was collected in Octomber-November 2017 from District 4 of Bucharest, Romania. For the microscopic studies, free hand sections of the fresh root, culm (internode), leaf and leaf-sheath of *Eleusine indica* (L.) Gaertn. were prepared using a razor blade. The present paper intends to illustrate the morphological features of the plant. Cross-sections performed in the botany laboratory were used to highlight anatomical characteristics. The sections were clarified with chloral hydrate for 24 hours and stained with alaun carmine and iodine green suitable to optical microscopic techniques. Observations and images of the anatomical structures in root, stem and leaves were made with the optical microscope Leica DM1000 LED, a Leica DFC295 video camera and a Leica S8 APO stereo microscope. A Sonny digital camera belonging to the microscopic and plant anatomy laboratory of the same research center was also used. Voucher specimens were deposited in the University of Agronomic Sciences and Veterinary Medicine of Bucharest's Herbarium. The taxonomic status of this plant was checked against different literature sources (Hafliger et al., 198l).

RESULTS AND DISCUSSIONS

Macroscopic characteristics

Morphological features of *Eleusine indica* harvested in Bucharest, District 4 (Romania).

Eleusine indica (L.) Gaertn. (Figure 1) is an annual erect plant, with the culm having 15-40 cm in length, 0.6 mm in diameter, glabrous, branched in the upper part, not just at the base, the nodes 0.51-3.46 mm thick, the stem 3.5-6 mm in diameter with alternate linear leaves, basal leaves 25 cm long and the leaf-sheath 5.5 cm, medium leaves with lamina 20 cm long and the leaf-sheath 5 cm, and the last leaf below the inflorescence has a lamina 14.5 cm long and the leaf-sheath 8.5 cm; the leaf width is 0.7 mm for both basal and upper leaves. Leaf-sheaths are coriaceous on the adaxial side, glabrous on the abaxial side, with short-ciliate along the edges. In the area between the leaf-sheath and the lamina (at the base of the lamina) 3.1 mm long hairs (Figure 2) are observed, the ligula is short 0.5-1 mm and membranous.

Figure 1. Habitus

The stem has branches at the second and third node from the ground surface, both shoots being fertile, with 5.5 cm long inflorescences. In the case of some other specimens, from near the ground develops three shoots 5.9-17 cm in height. The stem shows some shoots from the last nodule under the inflorescence, and the leaf on this stem has a total length of 38 cm (30 cm the lamina and 8 cm the sheath).

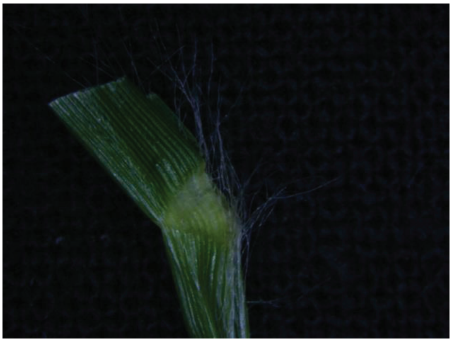

Figure 2. Leaf-sheath with hairs

The plants were harvested in October and November 2018 while some specimens were in the blooming phase and other individuals were in the fructification phase.

The inflorescence consists of 3-8 racemes, each 5-9 cm long, arranged more-or-less digitately, though one raceme may be inserted about 0,8-1 cm below the rest; the spikelets are elliptic, sessile, alternate on the rachis, laterally compressed, have 2-5 flowers and are arranged on two rows. The spikelets are 5.1-5.3 mm long and 2.1-2.6 mm wide (Figure 3).

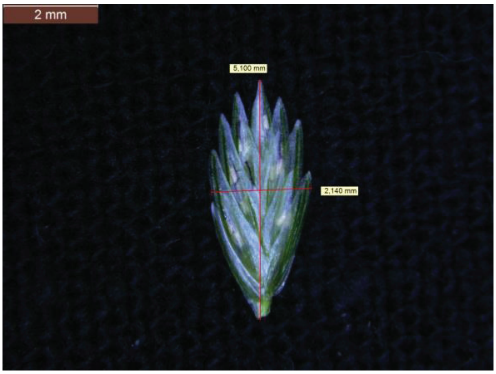

Figure 3. Spikelet

Glumes are shorter than the spikelet, unequal, the lower glum is small (1.1-3.0 mm) and 1-veined: the upper glume is longer than the lower, lanceolate, 2.1-4.1 mm long, 5-7 nerved, lemmas are broadly lanceolate, 2.3-3.1 mm long, membranous, 3-nerved, the palea is broadly lanceolate to narrowly ovate, 1.7-2.4 mm long. The flowers are bisexual with 3 stamens, short anthers, a glabrous ovary, 2 styles, free, with stigmas 0,8-0,9 mm long, plumose, reddish. Caryopses are ellipsoidal, black and 1.04-1.8-2 mm long, conspicuously, embryo basal, nearly circular; the hilum is basal and punctiform. The anatomical analysis of the caryopsis was supplemented by observations under a scanning electron microscope (Figure 4).

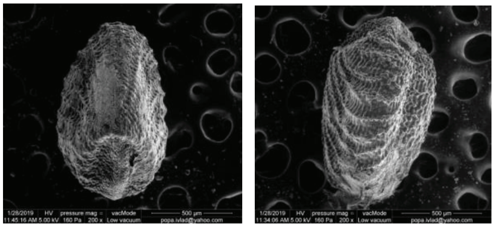

Figure 4. Caryopsis

The microscopic characters were studied as determined by Metcalfe and Chalk, 1950.

The branches of the stem (Figure 5) in the analyzed specimens are atypical because the literature states that branching is at the base of the plant (Nobis et al., 2011).

Figure 5. Stem branched

Microscopical characters

Root anatomy

Cross-section observations: the roots are about 1.5-1.8 mm in diameter; the epiblem with numerous unicellular root hairs; the hypodermis is one layered thick, parenchymatous, and the cells are polygonal in shape; the ground tissue is thin-walled, parenchymatous, consists of cortex, endodermis, pericycle, vascular bundle and pith; the cortex consists of 6-7 layers, polygonal parenchymatous cells, with intercellular spaces; the endodermis is the innermost layer of the cortex and forms a definite 1-layered ring, the cells are parenchymatous; the pericycle layer is ring-like, one-layered; the vascular bundles are of the radial type, polyarch, xylem strands alternate with phloem strands, xylem and phloem are oval to round, the protoxylem occurring near the periphery and the metaxylem inwards; the parenchymatous cells in the centre is the pith.

Culm anatomy (Figure 6)

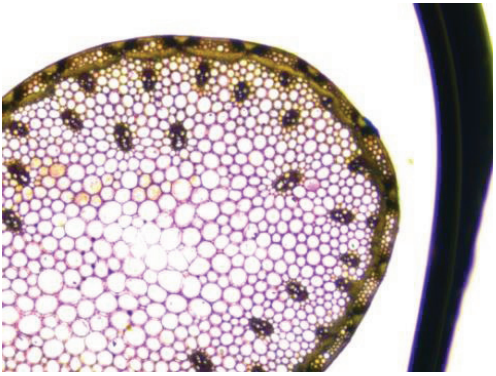

Figure 6. Stem cross-section

Cross-section observations: culms are about 3.5-6 mm in diameter, oval, with the cuticle layer thin, 0.49-0.5 μ thick; the epidermal cells are one-layered, rectangular in shape; the ground tissue consists of two types of cell, sclerenchymatous (4-9 layers, at the periphery) and parenchymatous (oval or rounded, intercellular spaces present); the vascular bundles are scattered in the center of culm, with smaller bundles near the periphery; the outline of the vascular bundles is oval or rounded, the bundle sheath is complete, sclerenchymatous, around each bundle; the vascular bundle presents xylem (vessel elements, fiber and xylem parenchyma) towards the centre and phloem (sieve tube elements, companion cells and phloem parenchyma) towards the epidermis (Figure 7).
The microscopical characters of the culm are: vascular bundle 44.8-75.20 μm, parenchyma cells 11.5-57.5 μm, vessel metaxylem 14.7-13.25 μm.

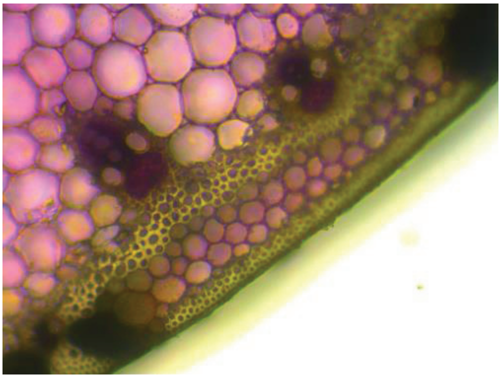

Figure 7. Stem cross-section (detail)

Leaf anatomy - lamina (Figure 8)
Anatomical analyses show that the leaves are amphistomatic with graminaceous type stomata, microhairs, macrohairs, collateral vascular bundles in the mezophill with a Kranz structure.

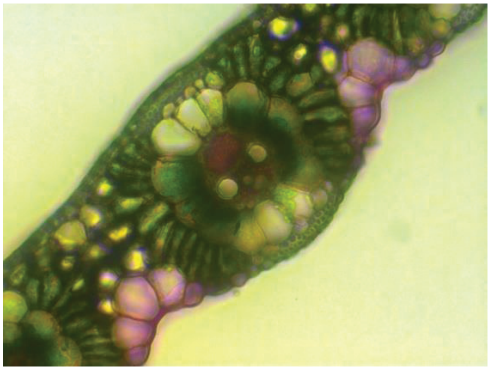

Figure 8. Lamina cross-section

The adaxial side features wide rounded ribs, separated by shallow V-shaped furrows.
The epidermal cells are arranged in parallel rows for both surfaces, with sinuous anticlinal walls; the cells are short and long. The short cells, located intercostally, are filled with silica bodies. The long cells, found between the veins, are elongated. The stomata are of the graminaceous type, present on the upper and lower epidermis. The upper and lower epidermis (Figures 9, 10) are one-layered with rectangular cells, with microhairs and macrohairs.
The bulliform cells (motor cells), with associated colorless cells, are present in the furrows of the upper epidermis; they have an oval or circular shape, are present in narrow

groups penetrating the mesophyll, between two consecutive vascular bundles. The vascular bundles are of the close collateral type. The chlorenchyma cells are radially arranged around the vascular bundles.

The vascular bundles feature sclerenchyma strands, on both the abaxial and adaxial sides. The xylem is found towards the upper side and the phloem towards the lower side. The xylem is composed of vessel elements, fibers and xylem parenchyma. The phloem is made up of sieve tube elements, companion cells and phloem parenchyma.

Adaxial intercostal zone: adaxial intercostal long cells with slightly sinuous walls, 6-9 rows of long cells between two costal zones, 1-2 stomatal rows between two costal zones; microhairs and macrohairs present.

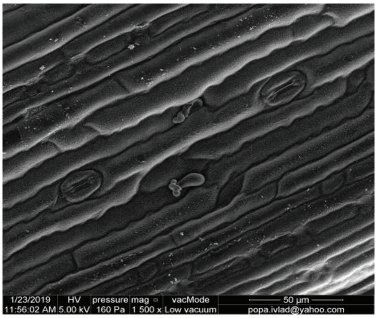

Figure 9. Upper epidemis with stomata and microhairs

Costal zone: saddle-shaped silica bodies, long cells with sinuous walls along with silica bodies. Abaxial intercostal zone: abaxial intercostal long cells with thin to moderately thick sinuous walls, 8-9 rows of long cells between two costal zones, 1-2 stomatal rows between two costal zones; microhairs present.

Costal zone: saddle-shaped silica bodies, long cells with sinuous walls along with silica bodies.

The results of cells' measurement in the leaf's cross-section: 10.65-13.49 µm thick upper epidermis, 0.5-0.6 µm thick cuticle, 135.79-231.7 µm thick mesophyll, 26.38-58.84 µm sclerenchyma sheath; 3.25-51.30 µm parenchyma cells, 10.01-10.35 µm thick lower epidermis, vascular bundles 78.8-80.7 µm. On the lower epidermis there are stomata 24.44-25.49 µm in length, long cells 163.7 µm in length and 15.97 µm in width, the short cells are 21.43-34.57 µm long and 8.10-8.18 µm wide.

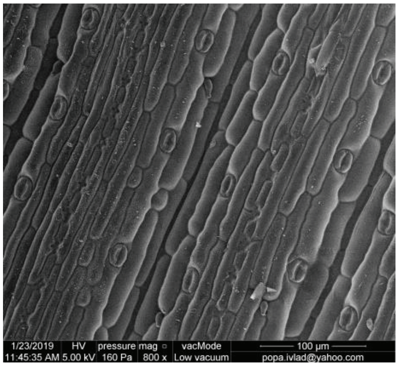

Figure 10. Lower epidermis with stomata and microhairs

The present study revealed that micro-morphological features of the leaf surfaces have a considerable identification.

Friere et al., (2005) reported that *Eleusine indica* is characterized by pear-like microhairs. However, Bibi et al. (2007), Ahmad (2009), Ahmad et al. (2011) did not find microhairs but saddle-shaped silica bodies. The leaf's epidermal features can help elucidate taxonomic problems (Prat, 1936; Metcalfe, 1960; Ellis, 1979; Palmer & Tucker, 1981; Palmer et al., 1985; Davila & Clark 1990; Cai & Wang, 1994; Mejia-Saules & Bisbey 2003; Prat & Vignal, 1968; Sanchez, 1974). These leaf epidermal characters are of great value in grass systematics and characterization of broad groups within the grasses, particularly subfamilies and tribes.

According to Prat (1936), the types of microhairs and silica bodies are highly useful in systematic studies. The anatomical analysis of the leaves was supplemented by observations performed under a scanning electron microscope (Figures 11, 12).

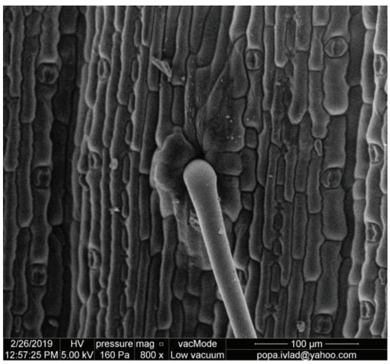

Figure 11. Upper epidemis with stomata and macrohairs

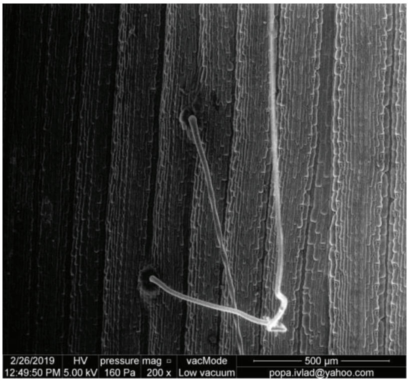

Figure 12. Upper epidemis and macrohairs

Midrib anatomy

The microscopic features of the midribs are as shown in Figure 13.

Cross-section observations: the cuticle is about 0.40 µ thick; the epidermal cells are rectangular in shape; the parenchyma cells have a hexagonal to polygonal shape, 3-5 layers; the vascular bundles are of the closed collateral type, each being circular with outer and inner bundle sheaths.

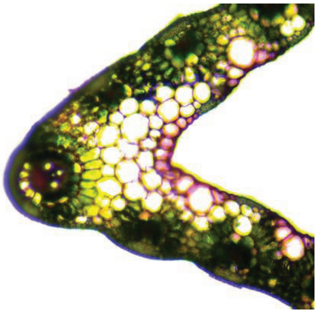

Figure 13. Midrib

The outer sheath is composed of parenchymatous cells, the inner sheath has sclerenchyma phloem lying towards the outer leaf side, the xylem is composed of vessel elements, fiber and xylem parenchyma. The phloem is composed of sieve tube elements, companion cells and phloem parenchyma.

Leaf-sheath anatomy

The cross-section shows that the cuticle is present on the epidermis of both leaf sides. The ground parenchymatous cells are arranged in 4-10 layers, compact, thin walled, hexagonal, variable in size. The vascular bundles are scattered, of the closed collateral type, oval to circular, the bundle sheath is complete. The xylem is composed of vessel elements, fibers and xylem, while the phloem is made up of sieve tube elements, companion cells and phloem parenchyma. The microscopic characteristics of the leaf-sheath are shown in Figures 14 and 15.

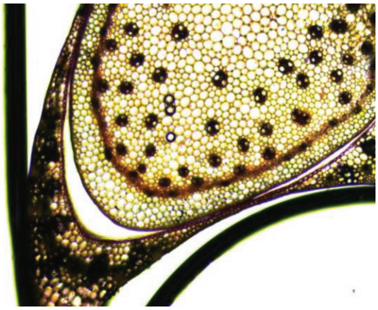

Figure 14. Cross-section in stem and leaf-sheaths

Figure 15. Cross-section of leaf-sheaths

CONCLUSIONS

This study is the first to report a detailed analysis of the morpho-anatomical features of *Eleusine indica* specimens growing in Romania.

The microscopic features (epidermis, stomata, cuticle, ornamentation and roughness) are in use since the beginning of the last century and information on foliar micromorphology can shed more light on structural features and their possible functional attributes.

We are of the opinion that the presently described micromorphological and anatomical characteristics will be quite useful for further studies and can be employed to obtain a key to set apart different species of the genera and different genera of the tribe.

The following characteristics were observed: the epidermis surface, the intercostal cells, the long and short cells, the orientation, the microhairs in both epidermis, the mesophyll, the vascular bundles. The distribution of the short cells' silica bodies along with their shape are bear significance for taxonomic purposes.
It is the first time when macrohairs hair is seen in the leaf in the upper epidermis, as an adaptation to the conditions of our country.

REFERENCES

Ahmad, F. (2009). Taxonomic studies of grasses of salt range of Pakistan, A Thesis Submitted to the Quaid-i-Azam University in Partial Fulfillment of the Requirements for the Degree of Doctor in philosophy - Plant Sciences - (Plant Taxonomy), (pp. 80–81).

Ahmad, F., Khan, M.A., Ahmad, M., Hameed, M, Tareen, R.B., Zafar, M., and Jabeen, A. (2011), Taxonomic application of foliar anatomy in grass of tribe Eragrostideae (*Poaceae*) from salt range of Pakistan. *Pak. J. Bot*., *43*(5), 2277–2284.

Al-Zubairi, A.S., Abdul, A.B., Abdelwahab, S.I., Peng, C.Y., Mohan, S., Elhassan, M.M. (2011). Eleusine indica possesses antioxidant, antibacterial and cytotoxic properties. Evidence - based Complementary and Alternative Medicine, Volume, Article ID 965370.Doi :10.1093/ecam/nep091.

Amarasinghe, V., & Watson, L. (1990). Taxonomic Singnificance of Microhair Morphology in the Genus Eragrostis Beauv. (Poaceae), *Taxon. 39* (1), 59–65.

Amarpreet, K. (2014). Plants Exhibiting Potential for Cancer Treatment. *Int. J. Pharm. Sci. Rev. Res., 27*(2), 23–53. ISSN 0976–044X.

An, Jing, Shen, XueFeng, Ma, QiBin, Yang, CunYi, Liu, SiMin, Chen, Yong. (2014). Transcriptome profiling to discover putative genes associated with paraquat resistance in goosegrass (*Eleusine indica* L.). *PLoS ONE, 9*(6):099940.

Anastasiu, P., & Negrean, G. (2006). *Alien vascular plants in Dobrogea (Romania) and their impact on different types of habitats*. In: Ivanova D. (Ed). Plant, fungal and habitat diversity investigation and conservation. *Proceedings of IV Balkan Botanical Congres. Sofia*, 590–596.

Anastasiu, P., & Negrean, G. (2005). Alien plants in Romania (I). *Analele Şti. Univ*. "*Alexandru Ioan Cuza*" *Iaşi*, *Ser. II, a. Biol. Veg*., *51*, 87–96.

Anastasiu, P., & Negrean, G. (2008). Plante străine noi în România. *Analele Univ. Craiova. Ser. Agricult*., *Montanologie*, *Cadastru*, *38*, 1–10; 47–51.

Anastasiu, P., Negrean, G., Fagaras, M., Samoila, C., Cogalniceanu, D. (2009). Major Gateways and Reservoirs for Alien Plant Species. *Acta Horti Bucuresti*, *36*, 41–60.

Babu, H.R., & Savithramma, N. (2014). Studies on stomata of some selected grass species of Poaceae and Cyperaceae. *World Journal of Pharmacy and Pharmaceutical Sciences*, *3*(7).

Baird, W.M.V., Morejohn, L., Zeng, L., Mysore, K., Kim, H.,1. (996). In: Brown H., Cussans G.W., Devine M.D., Duke S.O., Fernandez-Quintanilla C., Helweg A., Labrada R.E., Landes M., Kudsk P., Streibig J.C., Genetic, molecular and biochemical characterization of dinitroaniline herbicide resistance in goosegrass (*Eleusine indica*). *Proceedings of the Second International Weed Control Congress, Copenhagen, Denmark, 1-4*, 551–557.

Bibi, F., Khan, M.A., Ahmad, M., Zafar, M. (2007). Leaf epidermal anatomy of some grasses. *Pak. J. Sc., 59*(1-2), 47–51.

Breakwell, E. (1915). Anatomical structure of some native xerophytic grasses. *Proc. Linn. Soc. N. S. W., 40*, 42–55.

Brown, W.V. (1958). Leaf anatomy of grass systematics. *Bot. Gaz*., *119*, 170–178.

Cai, L.B. & Wang, S.J. (1994). Studies on the evolutionary trends and mechanism of the constituent cells of the leaf epidermis in Poaceae. *Acta Biologia Plateau Sinica*, *12*, 13–27.

Cha, ThyeSan, Mohamed, Ghazani Najihah, Ismail, Sahid, Chuah, TseSeng. (2014). Molecular basis for resistance to ACCase-inhibiting fluazifop in Eleusine indica from Malaysia. *Pesticide Biochemistry and Physiology*, *111*, 7–13.

Chen, S.L., & Phillips, S.M. (2006). Eleusine Gaertner. In Wu ZY, Raven PH, Hong DY (eds) Flora of China, Vol. 22, Poaceae. Science Press/Missouri Botanical Garden Press, Beijing/St. Louis.

Chopra, R.N., Nayar, S.L., Chopra, I.C. (1986). Glossary of Indian medicinal plants (Including the Supplement). Council Scientific Industrial Research, New Delhi (pp. 330).

Chopra, R.N. (1956). Glossary of Indian medicinal Plants. Council of Scientific and Company, Ltd Conservation. Planning and Management. *Springer, 15,* 595–606.

Ciocârlan, V. (2009). *Flora ilustrată a României*. *Pteridophyta* et *Spermatophyta* (ed. III). RO: Bucureşti: Editura Ceres.

Clayton, W.D., Harman, K.T., Williamson, H. (2006). GrassBase – the online world grass flora. http://www.kew.org/data/grasses-db.html.

Costea, M. (1996). The reccording of some new adventive taxa for Romania in the harbour Constanta. Revue Roumaine de Biologie. *Serie de Biologie Vegetale. Bucharest, 41*(2), 91–96.

Davila, P., and Clark, L.G. (1990). Scanning electron microscopy survey of leaf epidermis of *Sorghastrum* (Poaceae) Andropogoneae. *Am. J. Bot., 77*, 499–511.

De Melo, G.O., Muzitano, M.F., Legora-Machado, A., Almeida, T.A., De Oliveira, D.B., Kaiser, C.R., Koatz, V.L.G., Costa, S.S. (2005). C-glycosylflavones from the aerial parts of Eleusine indica inhibit LPS-induced mouse lung inflammation. *Planta Med., 71*(4), 362–363.

Doll, J. (2000). Glyphosate resistance in another plant. Re*sistant Pest Management, 11*(1), 5–6.

Ellis, R.P. (1987). A procedure for standardizing comparative leaf anatomy in the Poaceae, Te past twenty-five years. In F.R. Sonderstrom et al (Eds.) Proceeding of the International Symposium on Grass Systematics and Evolution, Washington DC.

Ellis, R.P. (1979). A procedure for standardizing comparative leaf anatomy in the Poaceae. The epidermis as seen in surface view. *Bothalia, 12*, 641–679.

Ellis, R.P. (1976). A procedure for standardizing comparative leaf anatomy in Poaceae. The leaf blade as viewed in transverse section. *Bothalia*, *12*(1), 65–109.

Ellis, R.P. (1986). In: *Grass systematics and evolution*. A review of comparative leaf blade anatomy in the Systematics of Poaceae. The past twenty five years. (Eds.): T.R. Soderstorm and K. H. Hilu. Smithsonian Institute,Washington , D.C., 3–10.

Esau, K. (1965). Plant Anatomy**.** U.K: Published by Wiley, New York.

Ettebong, E.O., Nwafor, P.A., Okokon, J.E. (2012). In vivo antiplasmodial activities of ethanolic extract and fractions of Eleucine indica. *Asian Pac. J. Trop. Med., 5*, 673–676.

Ettebong, E.O., Nwafor, P.A., Okokon, J.E. (2012). In vivo antiplasmodial activities of ethanolic extract and fractions of Eleucine indica. *Asian Pac. J. Trop. Med., 5*(9), 673–676.

Freire, S.E., Arambarri, A.M., Bayon, N.D., Sancho, G., Urtubey, E., Monti, C., Novoa, M.C., Colares, M.N. (2005). Epidermal characteristics of toxic plants for cattle from the Salado River basin (Buenos Aires, Argentina). *Bol. Soc. Argent. Bot., 40*(3-4), 1–28.

Grigore, A., Pîrvu, L., Bubuleanu, C., Colceru-Mihul, S., Ioniţă, C., Ioniţă, L. (2016). Medicinal plant crops, Important source of high value-added products. *Scientific Papers. Series A. Agronomy, LIX*, 298–307. ISSN 2285-5785.

Hafliger, E. & Scholz, H. (1981). Monocot Weeds 3 and Grass Weed 1, 2. CIBA.

Hansakul, P., Ngamkitidechakul, C. , Ingkaninan, K., Sireeratawong, S., Panunto, W. (2009). Apoptotic induction activity of *Dactyloctenium aegyptium* (L.) P.B. and *Eleusine indica* (L.) Gaerth. extracts on human lung and cervical cancer cell lines. *Songklanakarin Journal of Science and Technology (SJST), 31*(3), 247–359.

Hilu, K.W., & Randall, J.L. (1984), Convenient method for studying grass leaf epidermis. *Taxon, 33*(3), 413–415.

Hilu, K.W., Dewet, J.M.J., Seigler, D. (1978). Flavonoid patterns and systematics in Eleusine. *Biochem Syst Ecol., 6*, 247–249.

Holm, L.G., Plucknett, D.L., Pancho, J.V., Herberger, J.P. (1977). The world's worst weeds: distribution and biology. East-West Center, University Press of Hawaii, Honolulu, (pp. 609).

Iberahim, R., Adibah, Ahamad Bahtiar, Nazlina Ibrahim, N. (2016). Herpes simplex virus type-1 activity of Eleusine indica methanol extract 2016. *Malaysian Journal of Microbiology, 12*(6).

Myatt, Hla Wai, Kyaw, Naing, Daw, Hla Ngwe, Nu, Nu Aye, Myint, U, Maung, Maung Htay. (2005). Chemical Investigation and Hypotensive Activity Study on Some Myanmar Indigenous Medicinal Plants used in the Treatment of Hypertension. *Jour. Myan. Acad. Arts & Sc., III*(1).

Iberahim, R., Wan Ahmad Yaacob and Ibrahim N. (2015). Phytochemistry, cytotoxicity and antiviral activity of Eleusine indica (sambau), AIP. Conference Proceedings 1678, 030013 doi:http://dx.doi.org/10.1063/1.4931234.

Răduțoiu, D., & Stan, I. (2013). Preliminary data on alien flora from Oltenia. *Acta Horti Bot., 40*. 33–42. DOI: 10.2478/ahbb-2013-0004

Ibrahim, J.A., Muazzam, I., Jegede, I.A., Kunle, O.F., Okogun, J.I. (2007), Ethno-medicinal plants and methods used by Gwandara tribe of Sabo Wuse in Niger state, Nigeria, to treat mental illness. *Afr. J. Tradit. Complement Altern Med., 4*(2), 211–218.

Inamdar, J.A., Mohan, J.S.S., and Subramanian, R.B. (1986). Stomatal classification - a review. *Feddes Report, 97*(3,4), 147–160.

Iqbal, M., & Gnanaraj, C. (2011). Liver Protective Effects of Eleusine indica and Thysanolaena latifolia: Chemopreventive and Antioxidative Effects of Eleusine indica and Thysanolaena latifolia LAP LAMBERT Academic Publishing.

Iqbal, M., Gnanaraj, C. (2012). *Eleusine indica* L. possesses antioxidant activity and precludes carbon tetrachloride (CCl_4)-mediated oxidative hepatic damage in rats. *Environ Health Prev Med., 17*(4): 307–315.

Jalaludin, A., Ngim, J., Bakar, B.H.J., Zazali, Alias. (2010). Preliminary findings of potentially resistant goosegrass (*Eleusine indica*) to glufosinate-ammonium in Malaysia. *Weed Biology and Management, 10*(4), 256–260.

Kirtikar, K.R., and Basu, B.D. (1933). Indian Medicinal Plants. 2nd ed., 49, Leader.

Lee, LimJung, Ngim, J. (2000). A first report of glyphosate-resistant goosegrass (*Eleusine indica* (L) Gaertn) in Malaysia. *Pest Management Science, 56*(4), 336–339.

Meija, T. and Bisbey, F.A. (2003). Silica bodies and hooked papillae in lemmas of *Melica* species (Gramineae: Pooideae). *Bot. J. Linn. Soc., 141*. 447–463.

Metcalfe, C.R., and Clifford, H.F. (1968). *Microhairs on grasses.* Kew Bull., 21:3.

Metcalfe, C.R., and Chalk, L. (1950). *Anatomy of Monocotyledon* Vol. 1. UK **:** Oxford The Clarendon Press.

Metcalfe, C.R. (1961). The anatomical approach to systematics. General introduction with special reference to recent work on monocotyledons. pp. 146-150. In: Recent Advances in Botany, University of Toronto Press, Canada.

Mueller, T.C., Barnett, K.A., Brosnan, J.T., Steckel, L.E. (2011). Glyphosate-resistant goosegrass (Eleusine indica) confirmed in Tennessee. *Weed Science, 59*(4), 562–566.

Nagathein, Ashin (1977). *Pon-pya-say-A-bidon. Vol. 1.* Mingala Press. Yangon.

Negrean, G., & Constantin, N. (1999). Noi plante adventive în flora Bucureştiului. *Acta Horti Bot., 27*. 143–146.

Negrean, G., & Karaksonyi, C. (1984). Panicum dichotomiflorum şi Sida spinosa în flora României. Contrib. Bot. Cluj-Napoca: 33-35. *New Synthesis.* SCOPE Series. Island Press. Washington-Covelo-London. (pp. 368).

Nguyen, V.D., & Doan, T.N. (1989). Medicinal Plants in Vietnam, World Health Organisation 1989 ISBN 92 9061 101 4.

Niteesh, Kumar, Nautiyal, S. (2017). Leaf anatomy of two genera of tribe Eragrostideae (*Poaceae*) from Mandal forest of Kedarnath wildlife sanctuary, Uttarakhand, India. *International Journal of Botany Studies, 2*(5), 50–55.

Nobis, M., Kowalczik, T., Nowak, A. (2011). *Eleusine indica* (Poaceae): A new alien species in the flora Tajikistan. *Polish Botanical Journal, 56*(1), 121–123.

Onaran, A., & Bayan Y. (2016). Antifungal activity of Liquidambar orientalis L., and Myrtus communis L. against some plant pathogenic fungi. *Scientific Papers. Series A. Agronomy, Vol. LIX*, 360–364. ISSN 2285-5785

Palmer, P.G., & Tucker, A.E. (1981). A scanning electron microscope survey of the epidermis of East African Grasses. I. Smithsonian Contributions to Botany, 49: 1–84.

Palmer, P.G., Jones, S.G., Hutchison, S. (1985). A scanning electron microscope survey of the epidermis of east African Grasses 3. Smithsonian contributions to Botany, 55: 1–136.

Pattanayak, S. & Maity, D. (2017). Use of *Eleusine indica* (L.) Gaertn. *(Kechila ghas)* as an antipyretic medicine of herbivores. *Explor Anim Med Res*., *7*(1), 94–96.

Penescu, A., Bran, M., Nichita, M., Ionescu, N., Săndoiu, D.I., Ciontu, C., Şchiopu, T., Gâdea, M., Sonea, C., Bolohan, C. (2018). Comparative study regarding the influence of herbicides on the yield of sunflower crops, the crops being obtained with conventional, clearfield and expres sun technologies in the field conditions of Moara Domneasca. S*cientific Papers. Series A. Agronomy, LXI*(1). ISSN 2285-5785, ISSN CD-ROM 2285-5793, ISSN ONLINE 2285-5807, ISSN-L 2285-5785.

Prat, H. & Vignal, C. (1968). Utilisation des particularités de l'épiderme pour l'identification et la recherche des graminées. *Bol. Soc. Argent. Bot*., *12*. 155–166.

Prat, H. (1936). La Systematique des Graminees. *Annals des Sciences Naturelles, Botanie, 10*(18), 165–258.

Răvăruț, M., and Mititelu, D. (1960). Două specii noi în flora Republicii Populare Române. Stud. Cerc. Acad. R. P. Române, Fil. Cluj, Biol., 11(1): 7–9.

Rojas-Sandoval, J., & Acevedo-Rodríguez, P. (2014). Invasive Species Compendium, *Eleusine indica* (goose grass), Department of Botany-Smithsonian NMNH, Washington DC, USA.

Rubina, A., Sara, S., & Anjum, P. (2007). Stomatal types of Monocots within Flora of Karachi, Pakistan. *Pak. J. Bot., 39*(1), 15–21.

Sagnia, B., Fedeli, D., Casetti, R., Montesano, C., Falcioni, G., Colizzi, V. (2014). Antioxidant and anti-inflammatory activities of extracts from Cassia alata, Eleusine indica, Eremomastax speciosa, Carica papaya and Polyscias fulva medicinal plants collected in Cameroon. *PLoS One, 9*(8), 1–10.

Sanchez, E. (1971). Anatomia foliar de Chlorideae (Gramineae) Argentinas. *Kurtziana*, *6*. 103–218.

Saw, K.K. (2011). Morphological and Microscopical characters of *Eleusine indica* (L.) Gaertn. *Universities Research Journal, 4*(1).

Săndulescu, E.B., Scăeţeanu, Vasile G., Şchiopu, T., Oltenacu, N., Stavrescu-Bedivan, M.M. (2016). Morpho-anatomy and adaptation to some romanian aquatic environments OF Nymphoides peltata (GMEL.) O. KUNTZE (Asterales: Menyanthaceae). *Scientific Papers. Series A. Agronomy, LIX,* 537–542. ISSN 2285-5785.

Scholz, H. (1979). Bottle like microhairs in the genus Panicum (*Gramineae*). *Willdenowia, 8.* 511–515.

Sîrbu, C., Oprea, A., Elias, P. jun., Ferus P. (2011). New contribution to the study of alien flora in Romania. *J. Plant Develop.*

Sîrbu, C., & Oprea, A. (2011). *Plante adventive în flora României*. Iaşi: Edit. Ion Ionescu de la Brad.

Standifer, L.F., Wilson, P.W., Porche-Sorbet, R. (1984). Effects of solarization on soil weed seed populations. *Weed Science, 32*(55), 69–73, ISSN 0043-1745.

Terrel, E., & W.P., Wergin. (1981). Epidermal features and silica deposition in lemmas and awns of *Zizania* Gramineae). *Amer. J. Bot., 68*. 697–707.

Valverde, B.E., Chaves, L, Gonzales, J., Garita, I. (1993). Field evolved imazapyr resistance in Ixophorus unisetus and Eleusine indica in Costa Rica. Brighton crop protection conference, weeds. P*roceedings of an international conference, Brighton, UK, British Crop Protection Council (BCPC), 3*. 1189–1194.

Vargas, L., Ulguim, A.R., Agostinetto, D., Magro, T.D., Thürmer, L. (2013). Low level resistance of goosegrass (*Eleusine indica*) to glyphosate in Rio Grande do Sul-Brazil. *Planta Daninha, 31*(3), 677–686.

Watson, L., Dallwitz, M.J., Johnston, C.R. (1986). Grass Genera of the World: 728 detailed descriptions from an automated database. *Aust. J. Bot. 34*. 223–230 (with 3 microfiches).

IUCN on behalf of the Global Invasive Species Program. Gland. Switzerland. http://www.fws.gov/invasives/.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

SOIL ECOTOXICITY ASSESSMENT AFTER BIODEGRADATION OF SOME POLYMERIC MATERIALS

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Abstract

Plastics are ideal for many applications such as packaging, building materials and different commodities, due to their high durability properties, large availability and lower price. Their widespread use leads to waste disposal problems as plastics degrade in a long period of time, and because of their resistance to microbial degradation, they accumulate in the environment, having a strong negative impact. These facts have led to an increasing interest in the development of biodegradable polymeric materials derived from renewable resources as a method for manufacturing environmentally friendly materials. In order to assess the environmental sustainability, biodegradation studies of PLA/chitosan materials were performed. After the biodegradation process of the tested materials, the soils were analysed in order to evaluate their ecotoxicity using the seed germination bioassay. For this purpose, samples of soil were analysed after 50/100/150 days of biodegradation process and initial soil was used as control. An extraction in water was performed for each sample of soil and the resulted supernatant was diluted to yield 0, 25, 50, 75 and 100% supernatant (extract). The dilutions were then used for seed germination bioassay on radish (Raphanus sativus) and cucumber (Cucumis sativus) seeds. The global germination index (GI), was also calculated. The obtained results showed that the tested soils proved to have a nontoxic effect over the radish and cucumber seeds development, obtaining a GI over 80%.

***Key words**: ecotoxicity, seed germination, biodegradabale, polymeric materials.*

INTRODUCTION

Synthetic plastics are the wonder material of today's world, and life without them is unthinkable. Sadly, these same useful qualities are over shadowed by their balanced contribution to rubbish worldwide and its negative outcome for the environment (Imam et al., 2012). Only in Europe, the total production of plastic materials is over 60.000 t per year, and less than 50% of them end up in the official waste stream (Serrano-Ruíz et al., 2018). Therefore, there is a growing need to develop new polymeric materials that are environmentally friendly and can replace petroleum-based ones. Polymers derived from renewable resources present good properties such as availability, compostability, broad and abundant source range, environmentally-friendly and compatibility with foodstuffs and food application (Nur Hanani et al., 2014). Biopolymers represents a growing sector of the plastics market mostly in packaging and disposable products. Biopolymers are plastics that can be manufactured, at least in part, from renewable materials such as corn and sugar cane (Hottle et al., 2017).

Environmental risks from the use of polymeric materials stand in need for the prediction of the life cycle and side effects fastened to them, so the uses of exposure bioindicators/biological indicators are necessary to an integrated risk assessment (Gonçalves et al., 2016). Another required step is to determine the outcomes of the biodegradation processes in the environment, where the material will repeatedly break down and deliver many and different constituents to the soil. It is important to draw attention to the fact that the molecules released from plastic materials may frequently play a role in plant metabolism, or are allelochemicals themselves that balance interactions between plants (Closas et al., 2014). Therefore, biodegradable polymer wastes introduced into the environment should raise interest. Substances used for their development or compounds formed during the degradation process may either inhibit or

stimulate plant growth. Degree of risk may be different and, as in the case of compost salinity, some of the unfavorable effects of such materials application may be eliminated by dissolving them in soil (Kopec et al., 2013).
Seed germination and plant growth bioassay are the most frequent techniques used to evaluate ecotoxicity. In ecotoxicological tests, limit values of a substance or material can be indirect determined by the effects of different concentrations of the substance on the biological variables of individuals from a certain species or community (Lima et al., 2019). A large variety among bioassays and plant species exist. The seed germination bioassay could be relatively low sensitive to many toxic substances, because many chemicals may not be absorbed by seeds and the embryonic plant draws its nutritional requirements internally from seed stored materials and is effectively isolated from the environment (Mitelut & Popa, 2011).
The purpose of the present study was to determine the ecotoxicity of the soil resulted after the biodegradation of some new polymeric materials, on radish and cucumber seeds.

MATERIALS AND METHODS

Materials
Five polymeric materials were undergone to a biodegradation process in soil according to the standard method SR EN ISO 846/2000, described by Vasile et al. (2018). The materials were composed of PLA (polylactic acid), ATBC (tributyl o-acetyl citrate) or MB (masterbatch) as plasticizers and CH (chitosan). The materials were buried vertically in a glass jar filled with natural soil, characterized by a known capacity to retain water, and with specified water content, which was maintained during the entire experiment. Then, the jars were closed and incubated at a temperature of 25°C for 150 days.
After different periods (50, 100 and 150 days) the resulted soil was tested in order to determine its ecotoxicity on seeds. In Table 1, the soil that was used for analysis was described. The biological material used within these experiments was composed of 2 vegetal species: cucumber seeds (*Cucumis sativus*) and white radish seeds (*Raphanus sativus*).

Table 1. The soil resulted after the biodegradation process of studied biocomposites

No.	Sample of biocomposites undergone to the biodegradation process	Description of analysed soil
1.	-	The control soil, without any samples
2.	**PLA**	The soil in which PLA sample was incubated
3.	**PLA/ATBC**	The soil in which PLA/ATBC sample was incubated
4.	**PLA/MB**	The soil in which PLA/MB sample was incubated
5.	**PLA/ATBC/CH**	The soil in which PLA/ATBC/CH sample was incubated
6.	**PLA/MB/CH**	The soil in which PLA/MB/CH sample was incubated

Working method
For the determination of the ecotoxicity of the soil that resulted after the biodegradation process it was used the method described by Mitelut and Popa (2011).
Thereby, samples of soil were taken after 50, 100 and 150 days of maintaining in soil from the glass recipients, and the soil maintained in the same conditions of temperature (25°C) and humidity without samples buried in it was used as control. For the extraction process, the tested soil was mixed with distilled water, respecting a ratio of 1:2. The soil - water mixture was shaken for 6 hours at 25°C, centrifuged at 8000 rpm for 20 min at 20°C, and then filtered. The resulted supernatant was diluted with distilled water to yield 0, 25, 50, 75 and 100% supernatant (extract).
For seed germination assay, glass Petri dishes (10 cm diameter) were lined with fast speed qualitative filter paper and sterilized. Each dish received 5 ml of extract (10 Petri dishes/sample/concentration/type of seed). In each Petri dish were then distributed 10 seeds of the tested species (cucumber/radish). After incubation at 25°C for 72 hours in the dark, germinated seeds were counted, and the root length was measured.
The germination index (Gi) was calculated according to the formula:

$$\boldsymbol{Gi = \frac{G}{G_0} \times \frac{L}{L_0} \times 100}$$

where G_0 and L_0 represent the germination percentage and rootlet growth of the 100% distilled water control (0% supernatant).
It was also calculated the global germination index (**GI**), which was the Gi averages of 50 and 75% dilution treatments for each sample.

This index represents a very sensitive index which indicates the fact that the soil has no phytotoxic effect when its values are over 80% (Tiquia, 1996).

RESULTS AND DISCUSSIONS

The increase of supernatant concentration inhibits the germination capacity of cucumber seeds (Table 2), when tested after 50 days of incubation. After 150 days of incubation in soil, the germination capacity increased and the highest values were obtained for the soil in which PLA/ATBC/CH sample was incubated. The lowest values of the germination capacity of cucumber seeds was recorded by the control soil, on the entire period of testing.

Table 2. Germination capacity of cucumber seeds (%) (mean values of 100 seeds with standard deviation)

Incubation period (days)	Soil Sample / Supernatant concentration	Control soil	PLA	PLA/ATBC	PLA/MB	PLA/ATBC/CH	PLA/MB/CH
50	25%	88.88 ± 10.54	76.66 ± 20.61	85.55 ± 7.26	93.33 ± 7.07	72.22 ± 13.94	73.33 ± 14.14
	50%	78.88 ± 10.54	85.55 ± 11.30	81.11 ± 16.15	63.33 ± 14.14	75.55 ± 19.43	48.88 ± 14.52
	75%	67.77 ± 25.38	88.88 ± 7.81	77.77 ± 19.22	83.33 ± 7.07	75.55 ± 11.30	54.44 ± 10.13
	100%	72.22 ± 14.81	81.11 ± 15.36	75.55 ± 13.33	67.77 ± 18.55	87.77 ± 6.66	70.00 ± 20.00
100	25%	88.88 ± 10.54	97 ± 6.74	69 ± 21.31	89 ± 11.00	98 ± 4.21	100 ± 0.00
	50%	78.88 ± 10.54	89 ± 14.49	67 ± 20.57	92 ± 7.88	94 ± 4.21	99 ± 3.16
	75%	67.77 ± 25.38	81 ± 21.83	72 ± 28.20	93 ± 8.23	99 ± 3.16	99 ± 3.16
	100%	72.22 ± 14.81	97 ± 6.74	87 ± 14.18	96 ± 6.99	94 ± 8.43	98 ± 4.21
150	25%	88.88 ± 10.54	98 ± 4.21	98 ± 4.21	87 ± 10.59	98 ± 4.21	89 ± 7.37
	50%	78.88 ± 10.54	100 ± 0.00	97 ± 4.83	80 ± 15.63	100 ± 0.00	89 ± 9.94
	75%	67.77 ± 25.38	97 ± 4.83	79 ± 9.94	79 ± 11.00	98 ± 4.21	86 ± 15.05
	100%	72.22 ± 14.81	94 ± 8.43	100 ± 0.00	82 ± 13.16	99 ± 3.16	89 ± 7.37

Regarding the rootlet length of the germinated cucumber seeds (Table 3), it was observed that higher values were obtained for all tested soils compared to the control soil. The values of the rootlet length increased with the increasing period of incubation, meaning that once the polymeric materials start to degrade, the resulted compounds go into the soil and represent good nutrients for plant growth, comparing with the results obtained for control soil (without samples buried it it). These results are in accordance with the results obtained for the germination capacity of cucumber seeds.

Table 3. Rootlet length of cucumber seeds (mm) (mean values of 100 seeds with standard deviation)

Incubation period (days)	Soil Sample / Supernatant concentration	Control soil	PLA	PLA/ATBC	PLA/MB	PLA/ATBC/CH	PLA/MB/CH
50	25%	7.98 ± 6.63	6.69 ± 3.34	12.76 ± 3.16	20.55 ± 4.49	8.56 ± 4.45	15.46 ± 18.79
	50%	3.8 ± 3.26	5.68 ± 3.59	9.1 ± 4.11	13.52 ± 4.54	14.38 ± 7.80	9.17 ± 6.43
	75%	20.58 ± 9.02	6.87 ± 2.19	9.47 ± 4.45	20.43 ± 3.19	16.41 ± 5.77	9.74 ± 3.78
	100%	6.85 ± 4.38	11.86 ± 10.60	9.47 ± 4.45	15.68 ± 5.85	20.22 ± 3.27	8.12 ± 2.24
100	25%	7.98 ± 6.63	39.09 ± 5.02	15.22 ± 7.17	24.73 ± 8.60	41.93 ± 4.86	51.05 ± 2.95
	50%	3.8 ± 3.26	20.53 ± 12.61	10 ± 6.40	25.71 ± 4.47	56.3 ± 7.10	50.51 ± 4.73
	75%	20.58 ± 9.02	39.28 ± 18.79	33.09 ± 20.16	37.69 ± 5.68	51.81 ± 4.52	56.14 ± 5.71
	100%	6.85 ± 4.38	41.92 ± 5.49	24.98 ± 16.22	38.27 ± 9.70	46.84 ± 6.79	55.34 ± 6.40
150	25%	7.98 ± 6.63	58.15 ± 5.75	50.76 ± 4.16	36.52 ± 7.83	47.12 ± 4.87	40.42 ± 3.10
	50%	3.8 ± 3.26	43.45 ± 3.43	54.28 ± 7.92	46.63 ± 6.67	46.55 ± 3.31	33.01 ± 6.19
	75%	20.58 ± 9.02	61.09 ± 5.59	29.59 ± 4.10	47.02 ± 8.21	48.5 ± 2.72	41.63 ± 6.85
	100%	6.85 ± 4.38	58.57 ± 6.80	44.65 ± 5.14	36.11 ± 7.93	45.26 ± 5.09	39.71 ± 5.78

The germination capacity of radish seeds (Table 4) was over 90% for the control soil and for the soil in which PLA/ATBC/CH sample was maintained for all testing periods and concentrations. For the PLA/MB sample for every testing period, for at least one sample the GI was below 80%. However, in general, the values obtained for radish seeds were higher when compared to the germination capacity of cucumber seeds.

Table 4. Germination capacity of radish seeds (%) (mean values of 100 seeds with standard deviation)

Incubation period (days)	Soil Sample / Supernatant concentration	Control soil	PLA	PLA/ATBC	PLA/MB	PLA/ATBC/CH	PLA/MB/CH
50	25%	93 ± 8.23	97 ± 6.47	93 ± 6.74	88 ± 6.32	98 ± 4.21	93 ± 4.83
	50%	93 ± 8.23	97 ± 6.74	91 ± 11.00	90 ± 4.71	97 ± 6.74	92 ± 6.32
	75%	91 ± 7.37	92 ± 7.88	92 ± 6.32	79 ± 8.75	95 ± 7.07	87 ± 8.23
	100%	96 ± 6.99	84 ± 27.16	96 ± 5.16	91 ± 5.67	95 ± 8.49	86 ± 6.99
100	25%	93 ± 8.23	88 ± 9.18	72 ± 12.29	62 ± 35.83	96 ± 5.16	96 ± 6.99
	50%	93 ± 8.23	91 ± 8.75	92 ± 6.32	87 ± 8.23	91 ± 7.37	86 ± 10.74
	75%	91 ± 7.37	90 ± 9.42	83 ± 10.59	69 ± 13.70	96 ± 5.16	95 ± 7.07
	100%	96 ± 6.99	92 ± 7.88	88 ± 10.32	92 ± 9.18	99 ± 3.16	93 ± 8.23
150	25%	93 ± 8.23	90 ± 10.54	96 ± 5.16	81 ± 12.86	93 ± 8.23	86 ± 11.73
	50%	93 ± 8.23	96 ± 5.16	92 ± 7.88	86 ± 9.66	96 ± 6.99	78 ± 17.51
	75%	91 ± 7.37	92 ± 7.88	95 ± 5.27	85 ± 8.49	95 ± 7.07	78 ± 17.51
	100%	96 ± 6.99	94 ± 8.43	88 ± 9.18	75 ± 19.57	93 ± 10.59	85 ± 13.54

The rootlet length of the germinated radish seeds was measured (Table 5). It was observed that the lowest values were obtained for control soil, while the highest values were obtained for the soils in which PLA/ATBC/CH and PLA/MB samples were incubated for 50 days. After 100 days of incubation, a slightly inhibition effect was clearly observed for PLA/MB sample in comparison with the control and the other samples.

Table 5. Rootlet length of radish seeds (mm) (mean values of 100 seeds with standard deviation)

Incubation period (days)	Soil Sample / Supernatant concentration	Control soil	PLA	PLA/ATBC	PLA/MB	PLA/ATBC/CH	PLA/MB/CH
50	25%	28.47 ± 4.12	29.02 ± 5.72	33.78 ± 4.22	66.73 ± 13.81	51.39 ± 10.77	52.7 ± 6.48
	50%	30.04 ± 7.98	30.76 ± 2.88	33.52 ± 7.43	62.92 ± 9.02	50.07 ± 11.62	55.33 ± 7.77
	75%	33.27 ± 5.62	27.58 ± 5.94	29.14 ± 4.24	61.38 ± 11.16	49.29 ± 7.62	61.57 ± 7.06
	100%	31.03 ± 5.71	30.65 ± 4.25	39.8 ± 9.17	70.56 ± 12.79	49.32 ± 8.00	60.88 ± 6.88
100	25%	28.47 ± 4.12	49.7 ± 7.33	30.69 ± 7.63	13.25 ± 11.55	49.26 ± 8.98	40.21 ± 7.22
	50%	30.04 ± 7.98	44.8 ± 8.02	30.5 ± 5.38	41.14 ± 8.42	45.97 ± 6.80	57.12 ± 7.01
	75%	33.27 ± 5.62	53.79 ± 9.55	41.14 ± 10.91	16.94 ± 6.37	41.29 ± 9.04	49.88 ± 10.04
	100%	31.03 ± 5.71	52.28 ± 8.90	37.69 ± 7.78	28.95 ± 6.24	42.55 ± 7.76	51.12 ± 11.53
150	25%	28.47 ± 4.12	38.52 ± 6.85	50.15 ± 9.05	50.06 ± 12.48	57.72 ± 8.79	42.87 ± 8.96
	50%	30.04 ± 7.98	40.39 ± 10.08	44.89 ± 3.63	60.2 ± 9.42	41.22 ± 3.62	39.04 ± 11.06
	75%	33.27 ± 5.62	51.86 ± 8.09	54.86 ± 5.62	56.84 ± 3.98	45.1 ± 8.69	54.79 ± 8.52
	100%	31.03 ± 5.71	41.46 ± 11.05	48.19 ± 7.47	32.94 ± 11.22	54.17 ± 10.64	50.29 ± 10.96

According to GI values (Figures 1 and 2), the tested soils that resulted after the polymeric materials biodegradation tests for 150 days, proved to have a nontoxic effect over the radish and cucumber seeds, obtaining a GI over 80%. The lowest GI was obtained for the control soil for both species of seeds used.

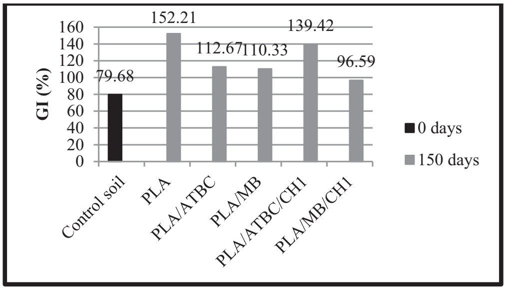

Figure 1. Global germination index (GI) for cucumber seeds

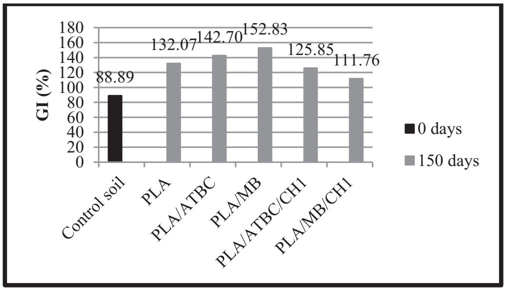

Figure 2. Global germination index (GI) for radish seeds

CONCLUSIONS

The ecotoxic effect of different biodegradable materials based on PLA was studied and the results obtained depend on materials composition, the nature of the biodegradable ingredients and the proportions between them. Depending on their composition, sometimes the materials presented a phytotoxic effect (Souza et al., 2013), and in other situations, depending on the nature of the utilised compounds, the studies showed that the materials did not present an ecotoxic effect (Palsikowski et al., 2018).

In the present study, the tested soils resulted after the polymeric materials biodegradation tests, proved to have, in general, a nontoxic effect over the radish and cucumber seeds, obtaining a GI over 80%. No direct correlation between supernatant concentration and seed germination and root length was found.

ACKNOWLEDGEMENTS

The research leading to these results has received funding from Romanian – European Economic Area (EEA) Research Programme operated by Ministry of National Education (MEN) under the EEA Financial Mechanism 2009 - 2014 and project contract no. 1SEE/2014, ACTIBIOSAFE.

REFERENCES

Closas, L.M., Botet, R., Pelacho, A.M. (2014). An in vitro crop plant ecotoxicity test for agricultural bioplastic constituents. *Polymer Degradation and Stability*, *108*, 250–256.

Gonçalves, S.P.C., Strauss, M., Delite, F.S., Clemente, Z., Castro, V.L., Martinez, D.S.T. (2016). Activated carbon from pyrolysed sugarcane bagasse: Silver nanoparticle modification and ecotoxicity assessment. *Science of the Total Environment*, *565*, 833–840.

Hottle, T.A., Bilec, M.M., Landis, A.E. (2017). Biopolymer production and end of life comparisons using life cycle assessment. *Resources, Conservation and Recycling*, *122*, 295–306.

Imam, S.H., Glenn, G.M., Chiellini, E. (2012). Utilization of biobased polymers in food packaging: assessment of materials, production and commercialization. *Emerging food packaging technologies*, *21*, 435–468. Woodhead Publishing Limited.

Kopec, M., Gondek, K., Baran, A. (2013). Assessment of respiration activity and ecotoxicity of composts containing biopolymers. *Ecotoxicology and Environmental Safety*, *89*, 137–142.

Lima, F.D.R., Martins, G.C., Silva, A.O., Vasques, I.C.F., Engelhardt, M.M., Cândido, G.S., Pereira, P., Reis, R.H.C.L., Carvalho, G.S., Windmöller, C.C., Moreira, F.M.S., Guilherme L.R.G., Marques, J.J. (2019). Critical mercury concentration in tropical soils: Impact on plants and soil biological attributes. *Science of the Total Environment*, *666*, 472–479.

Mitelut, A.C., Popa. M.E. (2011). Seed germination bioassay for toxicity evaluation of different composting biodegradable materials. *Romanian Biotechnological Letters*, *16*(1), 121–129.

Nur Hanani, Z.A., Roos, Y.H., Kerry, J.P. (2014). Use and application of gelatin as potential biodegradable packaging materials for food products. *International Journal of Biological Macromolecules*, *71*, 94–102.

Palsikowski, P.A., Roberto, M.M., Sommaggio, L.R.D., Souza, P.M.S., Morales, A.R.M. (2018). Ecotoxicity Evaluation of the Biodegradable Polymers PLA, PBAT and its Blends Using *Allium cepa* as Test Organism. *Journal of Polymers and the Environment*, *26*(3), 9338–9345.

Serrano-Ruíz, H., Martín-Closas, L., Pelacho, A.M. (2018). Application of an in vitro plant ecotoxicity test to unused biodegradable mulches. *Polymer Degradation and Stability*, *158*, 102–110.

Souza, P.M.S, Morales, A.R., Marin-Morales, M.A., Mei, L.H.I. (2013). PLA and Montmorilonite

Nanocomposites: Properties, Biodegradation and Potential Toxicity. *Journal of Polymers and the Environment*, *21*, 738–759.
Tiquia, S.M. (1996). Assessing toxicity of ‘spent sawdust pig-litter’ using seed germination technique. *Environmental Pollution*, *93*, 249–256.
Vasile, C., Pamfil, D., Râpă, M., Darie-Niţă, R.N., Mitelut, A.C., Popa, E.E., Popescu, P.A., Draghici, M.C., Popa, M.E. (2018). Study of the soil burial degradation of some PLA/CS biocomposites. *Composites Part B*, *142*, 251–262.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

VARIATION OF THE COEFFICIENT OF EXPANSION IN THE TURKEY OAK STANDS OF OLTENIA PLAIN FROM ROMANIA

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Abstract

The paper presents the results of the investigations carried out in the Turkey oak stands in the Oltenia Plain, whereby a statistically fundamental relation was established between the coefficient of expansion (F) of the stands and its main dendrometric characteristics (diameter - D and height - H), expressed by an equation of the form logF = a0 + a1logD + a2logH, whose coefficients are statistically significant. By demonstrating the joint influence of both dimensional characteristics (D and H) on the coefficient of expansion (F), the influence of the structure of the stands on their production was revealed, thus eliminating the error-generating hypothesis expressed by the identity FD = FH. Finally, for practical purposes, for the area of the study case, tables were developed to determine the coefficient of expansion (F) according to the two characteristics of the tree (D and H). By this way of determining the form factor (F) of the stands, substantial improvements are made to the precision of their volume determination by means of the general formula V = G x H x F.

Key words: *coefficient of expansion, Turkey oak, Oltenia Plain, Romania.*

INTRODUCTION

The Turkey oak is one of the four European representatives of the section *Cerris* in the subgenus *Quercus*, which also includes *Q. suber* L., *Q. trojana* Webb. and *Q. macrolepis* Kotschy (Tutin et al., 1964; Menitski, 1984; Boratyński et al., 2006; Danielewicz et al., 2014). In optimum conditions this tree attains a height of up to 35-40 m and a breast-height diameter of up to 2 m (Danielewicz et al., 2014). *Quercus cerris* is a sub-Mediterranean species with a geographical range embracing a vast area extending from south-western France through Italy, Switzerland, Austria and the Balkan Peninsula to Asia Minor and Lebanon (Menitski, 1984; Danielewicz et al., 2014; Meusel et al., 1965; Jalas et al., 1976; Browicz, 1953). Its northern limit in central Europe goes across central Slovakia (Danielewicz et al., 2014; Požgaj, et al., 1986; Pagan, 1992) and southern Moravia (Danielewicz et al., 2014; Kaplan, 2012). In the European part of its range, the localities of the Turkey oak extend from the sea level to a height of about 1,500 m a.s.l., in Anatolia usually between 500 and 1,500 m a.s.l., and in the Lebanon mountains, from 1,300 to 2,200 m a.s.l. (Boratyńskiet al., 2006; Danielewicz et al., 2014; Jalas et al., 1976). The climate in the areas where this species occurs ranges between warm temperate (maritime and transitional) and subtropical (from maritime to the continental and highland types) (Danielewicz et al., 2014). Sustainable forest management (SFM) is very important for study the biodiversity of the habitat forests and based on the concept of sustainable development and integrates three equally important pillars: environmental soundness, social justice, and economic viability (Paluš et al., 2018).

1.1. Geobotany characteristics for the analyzed *Quercus cerris* forest

The *Quercus cerris* forests in the studied territory are well structured and from the consistency point of view they are solid stands and very rare they are clearing stands) (Niculescu et al., 2009). The clearing of the forest appears as a result of the anthropic factors and less because of unfavourable stationary conditions (Niculescu et al., 2009). From the ecological point of view, taking into consideration the ecological indicators, it is found out that the *Quercus cerris* forests are xero-mesophyll, moderate-thermophyll and acid-neutrophyll (Niculescu et al., 2009; Borza, 1931; Mucina et al., 2016). From the phytocoenotaxonomical point of view the analyzed the *Quercus cerris* forests are formed of 9 species which are specific to the *Quercetalia pubescentis* et *Quercion petraeae*, 16 species of the *Queretea pubescenti-petraeae*, 4 species for the *Quercion frainetto*, 52 species belong to the *Querco-Fagetea* et *Fagetalia* and the rest are companions. From the floristically physiognomy and composition point of view the analysed *Quercus cerris* forests present at the arborescent stratum together with the dominant and enlightening species the following species: *Acer tataricum, A. campestre, Carpinus betulus, Pyrus pyraster, Malus domestica* etc. (Niculescu et al., 2009). The shrub and subshrub strata are well developed being represented constantly by the following species: *Crataegus monogyna, Prunus spinosa, Cornus sanguinea, C. mas, Euonymus europea, Rosa canina, Ligustrum vulgare* (Niculescu et al., 2009).

1.2. General information of the coefficient of expansion used in the analysed *Quercus cerris* forest

The coefficient of expansion (F) of the stand, defined as the average value of the component tree coefficients, results from the overall relation of the volume (*V = G x H x F)*:

$$F = \frac{V}{G\,x\,H} \qquad (1)$$

where:

V is the real volume of the tree;

G - base area of the stand;

H - average height of the stand.

The coefficient of expansion (F) of a stand according to the relation (1) can be obtained only if the volume of the tree is determined and known. This is often unknown, and F is determined precisely in order to measure the stand.

In our country, at the equian stands are used the usual coefficients of expansion, the value of which can be determined expediently using the production tables (Giurgiu, 1972; Giurgiu et al., 2004; Giurgiu & Drăghiciu, 2004)according to species, production class, age and average height.

Taking into account the mathematical-auxological model which formed the basis of the general production tables, the coefficient of expansion varies only with the average height (H) of the stands, based on the identity $F_{(D)} = F_{(H)}$, where $F_{(D)}$ is the joint coefficient obtained from the production tables according to the average diameter (D) of the base surface of the stand, and $F_{(H)}$is the coefficient of expansion of the stand, extracted from the production tables according to the average height H of the stand.

This method of determining the coefficient of expansion, without taking into account the influence of the stand structure on their production, is a simplifying and error-generating method (Leahu, 1994). Knowing that the structure bearer is the diameter, it is absolutely necessary that both the height (H) and the diameter (D) be taken into account when determining the coefficient of expansion.

In 2004, Leahushows that with *the aging of the stand, the coefficient of expansion decreases,* and according to Giurgiu (1979), *at the same height of the trees, the coefficient of expansion decreases as their diameter increases and vice versa*. This means that there are recorded the differences between the average coefficient of expansion of the real stand (F_r) and the normal stand (F_n) given in the production tables.

MATERIALS AND METHODS

2.1. Study area

The research was carried out in the pure Turkey oak stands of the Oltenia Plain from Romania, which occupy an area of 6451.2 ha (Cojoacă, 2010). The choice of the stands for the study of the variation of the coefficient of expansion was used deliberately. The research material consists of 16 sample areas of 2000 m^2 installed on the entire area of the Oltenia Plain of Romania (Amaradia Forestry District, Caracal Forestry District, Craiova Forestry District, Perisor Forestry District, Segarcea Forestry District and Vanju Mare Forestry District).

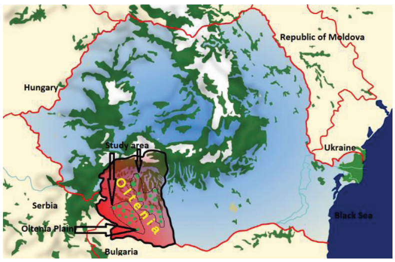

Figure 1. Map of Romania with the study area (Source:https://www.google.ro/search?q=harta+campia+olteniei&tbm - modified)

2.2. Field data and Methods

The sample surfaces were placed in landscape planner or practically pure units, of full-consistency, with ages ranging from 30 to 105 years old.

As a result, the distribution of the sampling areas would ensure the study of the stands in the various conditions: stationary, age, productivity, etc.

At the setup of the sample surfaces, account was taken of the weight of the Turkey oak, the structure by age classes, production classes and the consistency of the stands, as well as by the stationary and typological considerations, following the consultation of numerous parcel descriptions in forestry arrangements.

The sample markets, within the studied stands were placed according to representativeness criteria and their number was determined using known formulas from mathematical statistics (Giurgiu, 1972; Giurgiu et al., 2004).

Within each trial area, fieldwork consisted of:

- inventories of diameter category of 2 by 2 cm ranges by measuring the diameter at 1.30 m of the soil of all the trees in its range;
- the measurement of heights for all the trees of the surface;

The calculation of the tree volume in the sample areas was performed using the bifactorial regression equation (Niculescuet al., 2009; Pagan, 1992):

$$\log v = a_0 + a_1 \log d + a_2 \log^2 d + a_3 \log h + a_4 \log^2 h \quad (2)$$

where:

v represents the unitary volume of the trees by categories of diameters d, in m^3;

d - the base diameter of the tree in cm;

h -the tree's height;

a_0, a_1, a_2, a_3, a_4 - regression coefficients, established by species.

RESULTS AND DISCUSSIONS

Based on the field data and using the relation (1) we calculated the coefficients of expansions for each studied stand (Table 1).

Further on, starting from the research material made available, consisting of the 16 sample areas located in the Turkey oak stand in the Oltenia Plain, we studied the variation of the coefficient of expansion (F) in relation to the average height (H) as well as the average diameter (D).

For the analytical expression of the relationship between the coefficient of expansion (F) of the stands and their main dendrometric characteristics (D and H), the following regression equation was chosen:

$$\log F = a_0 + a_1 \log D + a_2 \log H \quad \text{.................. (3)}$$

where:

F represents the co-coefficient of expansion;

D- the average diameter of the stalk, in cm;

H - the average height of the stand, in m; a_0, a_1, a_2-regression coefficients.

Using the facilities of the SPSS statistics program (Statistical Package for the Social Sciences), the coefficients a_0, a_1, $a_2$of the multiple regression equation were determined.

From the analysis of the general statistics of the regression equation (Table 2), one can see how the multiple coefficient of correlation (*R*), the coefficient of determination (*R Square*) and the adjusted value of the coefficient of determination (*Adjusted R Square*) have very high values (close to 1), which means we have chosen the model correctly.

Thus, taking into account that the value of coefficient of determination (R^2) is 0.919, it results that 91.9% of the dependent variable (the coefficient of expansion F) is explained by the variation of the variables included in the model (D and H).

So the chosen linear regression model explains the dependence of the variables.

Also, the standard error (Standard error) was calculated, which is equal to 0.00447 meaning very close to zero, which means that the observed points are on the straight line of regression.

Testing of the chosen model was performed by the variance analysis method (ANOVA test) (Table 3).

It has thus been verified whether the model is relevant, meaning if the regression equation parameters differ significantly from 0.

As the unilateral critical probability (Sig.) is less than 0.05% (the assumed risk threshold or the fixed significance threshold) means that the model is statistically relevant (rejecting the hypothesis of the lack of significance of the independent variables).

Table 1. Base area, volume and coefficient of expansion in experimental areas (Cojoacă, 2010)

Test market symbol	Age, years	Production class	The average diameter of the base area, d_g	The average height, h_g	The base area, G		Volume, V		Coefficient of expansion F
					m^2/survey	m^2/ha	m^3/survey	m^3/ha	
Se_1	45	III	18.1	16.6	5.50	27.50	46,524	232,620	0.510
Se_4	40	III	17.9	15.9	6.89	34.45	54,460	272,300	0.497
Se_5	35	III	12.6	12.8	5.11	25.55	33,867	169,335	0.518
Se_7	30	III	13.4	12.7	4.78	23.90	30,979	154,895	0.510
Se_9	50	III	19.7	17.6	5.92	29.60	51,615	258,075	0.495
Se_{10}	50	III	19.7	18.2	5.97	29.85	53,522	267,610	0.493
Se_{11}	35	III	16.4	15.1	4.80	24.00	36,620	183,100	0.505
Se_{13}	55	III	22.6	18.7	6.26	31.30	57,425	287,125	0491
Se_{15}	105	III	33.5	23.6	7.39	36.95	81,624	408,120	0.468
Se_{17}	65	III	24.7	20.5	6.37	31.85	62,283	311,415	0.477
Se_{19}	55	III	21.1	16.8	5.67	28.35	46,234	231,170	0.485
Se_{20}	95	III	28.4	22.2	6.65	33.25	69,097	345,485	0.468
Se_{25}	75	III	27.0	21.3	733	36.65	73,880	369,400	0.473
Se_{26}	55	II	26.6	22.0	6.03	30.15	64,150	320,750	0.484
Se_{27}	70	II	30.8	24.8	729	36.45	86,231	431,155	0.477
Se_{28}	55	II	28.7	21.2	6.54	32.70	64,661	323,305	0.466

Table 2. Model Summary[b]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
					R Square Change	F Change	df1	df2	Sig. F Change
1	0.959[a]	0.919	0.907	0.00447	0.919	73.932	2	13	0.000
a. Predictors: (Constant), logH, logD									
b. Dependent Variable: logF									

Table 3. ANOVA[a]Test

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	.003	2	.001	73.932	.000[b]
	Residual	.000	13	.000		
	Total	.003	15			
a. Dependent Variable: logF						
b. Predictors: (Constant), logH, logD						

Estimated values for model coefficients, as well as the statistics needed to verify common assumptions about coefficients, are shown in Table 4. It can be seen (Table 4) that the significance level (*Sig.*) for both the constant and the independent variables (log D and log H) is less than 0.05 and the confidence intervals do not contain 0, where it follows that all the coefficients are statistically significant and the model is relevant.

This means that the size of the coefficients of expansions (F) in the studied area depend on both the diameter D and the height H of the stands. Thus, the estimated model is as follows:

$$\log F = -0.217 - 0.215 \log D + 0.153 \log H \quad (4)$$

By tabling the (4) relation which expresses the relation between the coefficient of expansion (F) and the diameter (D) and the height (H) for the Turkey oak stands in the Oltenia Plain it was possible to obtain the coefficient of was possible to obtain the coefficient of expansion of the stand depending on diameter and height (Table 5). The graph in Figure 2 and 3 suggests the variation of the coefficient of expansion in relation to both the diameter and the height. It can be noticed that at the same average diameter of the stand, the average coefficients of expansion increase as the height increases, while maintaining the constant height, the average values of the coefficient of expansion decrease with the increase of the average diameter of the stand.

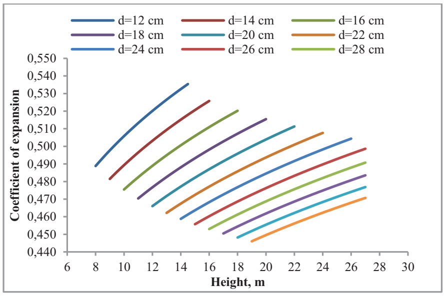

Figure 2. The variation of the coefficient of expansion in relation to both the height

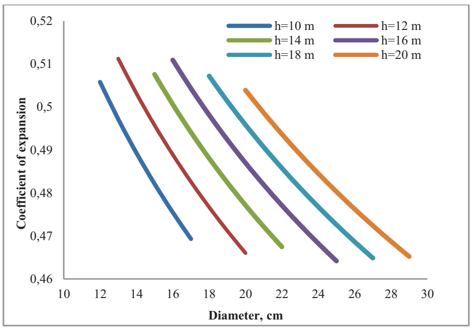

Figure 3. The variation of the coefficient of expansion in relation to both the diameter

Table 4. Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations		
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part
1	(Constant)	-.217	.027		-8.023	.000	-.276	-.159			
	logD	-.215	.049	-1.851	-4.356	.001	-.321	-.108	-.943	-.770	-.343
	logH	.153	.070	.924	2.174	.049	.001	.305	-.895	.516	.171
a. Dependent Variable: logF											

Table 5. The average coefficients of expansion (F) of the Turkey oak in the Oltenia Plain
logF = -0.217 - 0.215logD + 0.153logH

Average heights, H - m	Average diameters, D - cm											
	12	14	16	18	20	22	24	26	28	30	32	34
8	0.489	-	-	-	-	-	-	-	-	-	-	-
8.5	0.493	-	-	-	-	-	-	-	-	-	-	-
9	0.498	0.481	-	-	-	-	-	-	-	-	-	-
9.5	0.502	0.485	-	-	-	-	-	-	-	-	-	-
10	0.506	0.489	0.475	-	-	-	-	-	-	-	-	-
10.5	0.510	0.493	0.479	-	-	-	-	-	-	-	-	-
11	0.513	0.496	0.482	0.470	-	-	-	-	-	-	-	-
11.5	0.517	0.500	0.486	0.474	-	-	-	-	-	-	-	-
12	0.520	0.503	0.489	0.477	0.466	-	-	-	-	-	-	-
12.5	0.523	0.506	0.492	0.480	0.469	-	-	-	-	-	-	-
13	0.527	0.509	0.495	0.483	0.472	0.462	-	-	-	-	-	-
13.5	0.530	0.512	0.498	0.485	0.474	0.465	-	-	-	-	-	-
14	0.533	0.515	0.501	0.488	0.477	0.467	0.459	-	-	-	-	-
14.5	0.535	0.518	0.503	0.491	0.480	0.470	0.461	-	-	-	-	-
15	-	0.521	0.506	0.493	0.482	0.472	0.464	0.456	-	-	-	-
15.5	-	0.523	0.508	0.496	0.485	0.475	0.466	0.458	-	-	-	-
16	-	0.526	0.511	0.498	0.487	0.477	0.468	0.460	0.453	-	-	-
16.5	-	-	0.513	0.500	0.489	0.479	0.470	0.462	0.455	-	-	-
17	-	-	0.516	0.503	0.492	0.482	0.473	0.465	0.457	0.450	-	-
17.5	-	-	0.518	0.505	0.494	0.484	0.475	0.467	0.459	0.452	-	-
18	-	-	0.520	0.507	0.496	0.486	0.477	0.469	0.461	0.454	0.448	-
18.5	-	-	-	0.509	0.498	0.488	0.479	0.471	0.463	0.456	0.450	-
19.0	-	-	-	0.511	0.500	0.490	0.481	0.473	0.465	0.458	0.452	0.446
19.5	-	-	-	0.513	0.502	0.492	0.483	0.474	0.467	0.460	0.454	0.448
20	-	-	-	0.515	0.504	0.494	0.485	0.476	0.469	0.462	0.455	0.450
20.5	-	-	-	-	0.506	0.496	0.486	0.478	0.470	0.464	0.457	0.451
21	-	-	-	-	0.508	0.497	0.488	0.480	0.472	0.465	0.459	0.453
21.5	-	-	-	-	0.509	0.499	0.490	0.482	0.474	0.467	0.461	0.455
22	-	-	-	-	0.511	0.501	0.492	0.483	0.476	0.469	0.462	0.456
22.5	-	-	-	-	-	0.503	0.493	0.485	0.477	0.470	0.464	0.458
23	-	-	-	-	-	0.504	0.495	0.487	0.479	0.472	0.465	0.459
23.5	-	-	-	-	-	0.506	0.497	0.488	0.480	0.473	0.467	0.461
24	-	-	-	-	-	0.508	0.498	0.490	0.482	0.475	0.468	0.462
24.5	-	-	-	-	-	-	0.500	0.491	0.484	0.476	0.470	0.464
25	-	-	-	-	-	-	0.501	0.493	0.485	0.478	0.471	0.465
25.5	-	-	-	-	-	-	0.503	0.494	0.486	0.479	0.473	0.467
26	-	-	-	-	-	-	0.504	0.496	0.488	0.481	0.474	0.468
26.5	-	-	-	-	-	-	-	0.497	0.489	0.482	0.475	0.469
27	-	-	-	-	-	-	-	0.499	0.491	0.484	0.477	0.471

The research carried out in the Turkey oak stands in the Oltenia Plain on the variation of the coefficient of expansion led to the following results:

- based on the experimental material available, a link between the coefficient of expansion (F) of the stands and its main dendrometric characteristics (D and H) was established by means of the multiple linear regression, expressed by the following equation: logF = -0.217 - 0.215 logD + 0.153logH, whose coefficients are statistically significant;
- the determination coefficient R^2 = 0.919 shows that approximately 92% of the variation of the coefficient of expansion F is explained by the variation of the diameter D and the height H of the stand;
- with the help of the ANOVA test (Table 3) it was demonstrated that the model adopted was statistically relevant (the hypothesis of the lack

of significance of the independent variables was rejected - Sig. <0.05%);

- by demonstrating the joint influence of both dimensional characteristics (D and H) on the coefficient of expansion (F), the influence of the structure of the stands on their production was revealed, thus eliminating the error-generating hypothesis expressed by the identity $F_D = F_H$;

- given the fact that in establishing the coefficient of expansion (F) of the stand its is taken into account its variability in relation to both the height (H) and the diameter (D), it is possible to make substantial improvements to the determination of the volume of the stands by means of the general formula V = G x H x F. Thus, assuming that the surface would be determined by integral inventories and the height by exact procedures, it is assured a precision as high as in the average tree of the stand method, provided these tables be applied only to the stand category for which they were developed (Leahu, 1994);

- for practical purposes, by tabulation of the relation (4), for the area under study and for the types of stands where the research was carried out, it was possible to prepare provisional tables for determining the coefficient of expansion according to the two features of the stand (D and H) (Table 5). Knowing that variation in the coefficient of expansion of a stand differs from the composition, consistency, age, and production class of the stand (Giurgiu, 1979) for extending the application of these tables to other stand species is necessary complex biometric research.

The research carried out on small spaces, which established the common influence of the diameter D and the average height H on the average coefficient of expansion F of the stand, are in accordance with the recommendations of the specialty literature. Thus, Giurgiu et al. (1979) and Leahu (1994) recommends “the continuation and deepening of the research regarding the auxological dynamics of the stands in relation to the age, structure and household interventions in order to adapt the mathematical-auxological models and the production tables to the regional particularities of the forest ecosystems, passing from general to regional ones on ecological bases.”

CONCLUSIONS

By demonstrating the joint influence of both dimensional characteristics (D and H) on the coefficient of expansion (F), the influence of the structure of the stands on their production was revealed, thus eliminating the error-generating hypothesis expressed by the identity $F_D = F_H$.

In conclusion, for practical purposes, for the area of the study case, tables were developed to determine the coefficient of expansion (F) according to the two characteristics of the tree (D and H).

By this way of determining the form factor (F) of the stands, substantial improvements are made to the precision of their volume determination. We can be noticed, after this analysis in the research area in the pure Turkey oak stands of the Oltenia Plain, that at the same average diameter of the stand, the average coefficients of expansion increase as the height increases, while maintaining the constant height, the average values of the coefficient of expansion decrease with the increase of the average diameter of the stand. The research carried out on small spaces, which established the common influence of the diameter D and the average height H on the average coefficient of expansion F of the stand, are in accordance with the recommendations of the specialty literature.

REFERENCES

Boratyński, A., Boratyńska, K., Filipiak, M. (2006). Systematics and geographical distribution. In: Dęby. Ed. W. Bugała. NaszeDrzewaLeśne. *Monografie Popular non aukowe, 11,* 63–114. Bogucki Wyd. Nauk., Poznań.

Borza, A. (1931). Phytosociological studies in Retezat Mountains. *Bul. Gr. Bot. Cluj*, *14*, 1–84.

Browicz, K. (1953). The oaks cultivated in Poland. *Rocz. Dendr*., *9*. 71–122.

Cojoacă, F.D. (2010). Research on the structure, growth and production of the *Quercus cerris* and *Quercus frainetto* in Oltenia Plain. PhD thesis, University Transilvania, Braşov.

Danielewicz, W., Kiciński, P., Antosz, L. (2014). Turkey oak (*Quercus cerris* L.). In Polish forests, Acta Sci. Pol. Silv. Colendar. *Rat. Ind. Lignar*., *3*(2), 5–22.

Gerorgescu, C.C. (1941). Turkey oak as a type of forest I. *Rev. Pad., 53*(8-9), 444–457.

Giurgiu, V. (1972). *Methods of mathematical statistics applied in forestry*. Editura Ceres, Bucureşti.

Giurgiu, V. (1972). *Biometrics trees and stands in Romania*. Editura Ceres, Bucureşti.
Giurgiu, V., Decei, I., Drăghiciu, D. (2004). *Methods and dendrometrical tables*. Editura Ceres, Bucureşti.
Giurgiu, V., Drăghiciu, D. (2004). *Mathematical-auxologica models and tables production stands*. Editura Ceres, Bucureşti.
Giurgiu, V. (1979). *Dendrometrical and forestry auxological*. Editura Ceres, Bucureşti.
Jalas, J., Suominen, J. (1976). Eds. Atlas Florae Europaeae. Distribution of Vascular Plants in Europe. Vol. 3, Salicaceae to Balanophoraceae, The Committee for Mapping the Flora of Europe and Societas Biologica Fennica Vanamo Helsinki.
Kaplan, Z. (2012). Flora and phytogeography of the Czech Republic. *Preslia, 84*, 505–573.
Leahu, I. (1994). *Dendrometry*. Editura Didactică şi Pedagogică. R.A., Bucureşti.
Menitski, Yu. L. (1984). Asia's oaks, Nauka Leningrad.
Meusel, H., Jäger, E., Weinert, E. (1965). Comparative chorology of the Central European Flora. Gustav Fischer Jena.
Mucina, L., Bültmann, H., Dierßen, K., Theurillat, J.P., Raus, T., ČarniTichý, L. (2016). Applied Vegetation.
Niculescu, M., Bercea, I., Matei, G., Nuta, I.S., Iovu, I., Ciupitu, Ş.T., Sălceanu, C. (2009). Researches about *Quercus cerris* forests situated in the North-East of Dolj County. *Annals of the University of Craiova, Agriculture-Montanology-Cadastre Series, Craiova*, *39*, 291–296.
Pagan, J. (1992). Forest dendrology. Technická Univ. Zvolen.
Paluš, H., Parobek, J., Šulek, R., Lichý, J., Šálka, J. (2018). Understanding Sustainable Forest Management Certification in Slovakia: Forest Owners' Perception of Expectations, Benefits and Problems. *Sustainability*, *10*(7), 2470.
Požgaj, J.,Horváthová, J. (1986). A contribution towards the understanding of the variability and ecology of selected species of the genus *Quercus* L. in Slovakia. Acta Dendrobiol.
Tutin, T.G., Heywood, V.H., Burges, N.A., Velentine, D.H., Walters, S.M., Webb, D.A. (1964). *Quercus* L. In: Flora Europaea. Vol. 1, Cambridge Univ. Cambridge, 61–64.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

COMPARISON OF ENERGY CONSUMPTION IN CULTIVATION OPERATIONS FOR WHEAT PRODUCTION IN TURKEY

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Abstract

The objective of this study was to compare energy consumptions under traditional cultivation system for wheat production in Turkey. The data used in this study were collected through a questionnaire by face to face interviews. The amount of energy that consumed in tillage, planting, fertilizing, agricultural spraying, harvesting and transport stages were calculated for wheat cultivation. The energy inputs such as human labor, tractors, tools/machineries, fuel/oil, fertilizers, pesticides, irrigation and seed obtainment processes were taken into consideration to determine the amount of energy that used in wheat cultivation. The total energy used in various farm inputs for wheat cultivation was 17159.5 MJ/ha. The energy output/input ratio, specific energy, energy productivity and net energy efficiency were found to be 2.21, 7.18 MJ/kg, 0.14 kg/MJ and 20746.5 MJ/ha, respectively.

***Key words**: wheat, energy consumption, Turkey.*

INTRODUCTION

Energy consumption per unit area in agriculture is directly related with the development of technological level and cultivation. The inputs such as fuel, electricity, machinery, seed, fertilizer and chemical take significant share of the energy supplies to the cultivation system in modern agriculture. The use of intensive inputs in agriculture and access to plentiful fossil energy has provided an increase for standards of living and food production. However, some problems in agricultural production have been faced due to mainly high level dependency on fossil energy. The problems with the use of fossil energy have become into focus by oil embargo in 1973 and increase in energy prices. Nowadays environmental issues such as global warming are the major concerns related to the use of fossil energy. Furthermore considering that fossil energy is a limited resource, it has to be conserved for future generations by using efficiently in a sustainable manner. Energy is considered to be a key player in the generation of wealth and also a significant component in economic development. This makes energy resources extremely significant for every country in the world. Energy use has been a matter of policy concern since the 1970s. After the oil crises in 1973 and 1979, governments intensively promoted energy conservation measures. Then in 1980s, the primary focus shifted to air pollution caused by combustion of fossil fuels. In the recent years, energy use and associated greenhouse gas emissions and their potential impacts on the global climate change have been the worldwide concern. Improving the end-use energy efficiency is one of the most effective ways to reduce energy consumption in the industrial, commercial, transportation, utility, residential and agricultural sectors and their associated pollutant emissions (Dincer et al., 2004).

Efficient use of the energy resources is vital in terms of increasing production, productivity, competitiveness of agriculture as well as sustainability to rural living. Energy auditing is one of the most common approaches to examine energy efficiency and environmental impact of the cultivation system. It enables researchers to calculate input-output ratio, other relevant indicators and energy use pattern in an agricultural activity. Moreover, the energy audit provides sufficient data to establish functional forms to investigate the relationship between energy inputs and outputs. Estimating these functional forms is very useful in terms of determining elasticities of inputs on yield and production. Energy use pattern and contribution of energy inputs vary depending on farming systems, crop season and farming conditions. Considerable work has been conducted on the use of energy in agriculture with respect to efficient and economic use of energy for sustainable production. Energy input–output analysis is usually used to evaluate the efficiency and environmental impacts of production systems. This analysis is important to perform necessary improvements that will lead to a more efficient and environment–friendly production system (Ozkan et al., 2003; 2004). Comprehensive studies have been performed on energy use in different agricultural products. Optimization of energy use in agriculture is reflected in two ways, i.e. an increase in productivity at the existing level of energy inputs or conserving the energy without affecting the productivity (Singh et al., 2004).

For that reason, energy use in wheat cultivation was examined in the present study. The main objective of this study is to evaluate the energy input and output for wheat cultivation in of Turkey. Data for the cultivation of wheat were collected from farms by using a face to face questionnaire method. This study seeks to analyze the effect of indirect and direct energy on yield using functional form. The energy ratio, specific energy, energy productivity and net energy efficiency as indicators for the determination of energy efficiency were calculated in the wheat cultivation.

MATERIALS AND METHODS

The basic characteristics of surveyed farms

The data included energy inputs from different sources, data input to various farm operations and yield. Taking actual farm size as the variable, the total 132 farms was randomly selected by using stratified random sampling. The permissible error was defined to be 5% for 95% reliability (Yamane, 1967).

$$n = \frac{\left(\sum N_h S_h\right)}{N^2 D^2 + \sum N_h {S_h}^2} \quad \text{……………………..(1)}$$

Where: n is the required sample size; N is the number of holdings in target population; N_h is the number of population in h the stratified; S_h is the variance of h the stratified; D is the precision where (x-X).

The total agricultural land of the investigated 132 farms is 2413.7 hectares. Almost 30% of the farm land less than 5 hectares, and 50% less than 10 hectares. In spite of large number of small farms, less than 5 hectares, cultivate only 5% of total land, and 14% cultivates by the farms less than 10 hectares, cumulatively. Although, farms over 50 hectares which are only 6% of the total number of investigated farms, cultivate 28% of total land. The average size of farms is 18.3 hectares, and 37.2% of farm land is under irrigation. This proportion is relatively high at small farm groups.

In general, farm lands have been divided into many parcels. The average number of parcel is three. 80.3% of average operated land by farms is owned, 13.5% of it rented and rest (6.2%) is share cropped. Tenancy is more common at small farm groups. The share of rented land at small farm group (less than 5 hectares) increases up to 25%. Share cropping is not common in the region. Its share is only around 6%. This proportion is lower at small farm groups.

Analysis of energy use in wheat cultivation

A proforma questionnaire was prepared in order to collect the required information related to the land possessed by the farmers and the utilization pattern, crop yield, operation time, fuel consumptions, fertilizer, chemical and seed inputs. The energy inputs as direct and indirect inputs were studied in two groups for the wheat cultivation in the Southeast Anatolia region of Turkey.

Direct energy inputs

Direct energy inputs, which were calculated according to inputs that used directly and have high energy values in the cultivation of wheat. In this sense, during the cultivation process, fuel and oil energies that consumed by agricultural tools/machineries was evaluated as direct energy inputs. Thus,

$$EI_{direct} = E_{fuel} + E_{oil} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(2)$$

where:

EI_{direct} = Direct energy input (MJ/ha);
E_{fuel} = Fuel energy consumption (MJ/ha);
E_{oil} = Oil energy consumption per area (MJ/ha).

Energy input of fuel and oil consumption

The amount of consumed fuel energy for unit cultivation area in the wheat cultivation (ha) is calculated depending on the amount of fuel that consumed by the tractor and the calorific value of diesel fuel, as it can be seen down below:

$$E_{fuel} = M_{fuel} \times FHV_{fuel} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(3)$$

where:

E_{fuel} = Fuel energy consumption per area (MJ/ha);
M_{fuel} = Fuel consumption of the tractor (L/ha);
FHV_{fuel} = The lower calorific value of fuel (MJ/L).

The oil energy input which occurs due to engine oil consumption in wheat cultivation was determined by considering of the oil consumption of the farm tractor per hour, during the process of cultivation. The oil consumption of the farm tractor per hour was determined by depending on the highest PTO (power take off) shaft power of the tractor, which can be formulated as follows (Gungor & Ozturk, 2019):

$$OCT = 0{,}00059 \times HSP_{max} + 0{,}02169 \ldots\ldots\ldots(4)$$

where:

OCT = The oil consumption of tractor (L/h);
HSP_{max} = PTO shaft power of the tractor (kW).

The maximum PTO shaft power (HSP_{max}) of the agricultural tractor that was used for the cultivation process of wheat was considered as 88% of its rated power (*RPT*, kW) and determined as follows (Ozturk, 2018):

$$HSP_{max} = 0{,}88 \times RPT \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(5)$$

The oil energy amount that consumed per unit area in wheat cultivation process was calculated depending on the amount of oil that consumed by the tractor per hour during the manufacturing process, the lower calorific value of consumed oil and work efficiency of the tractor per area, which is defined as:

$$E_{oil} = OCT \times OHV_{oil} \times WCT \ldots\ldots\ldots\ldots\ldots\ldots(6)$$

Where:

E_{oil} = Oil energy consumption per area (MJ/ha);
OCT = Oil consumption of tractor per hour (L/h);
OHV_{oil} = The lower calorific value of oil (MJ/L);
WCT = Working capacity of the tractor (h/ha).

During cultivation operations in the field, the lower calorific value of engine oil (SAE 40) was taken into account as 38.2 MJ/kg.

Indirect energy inputs

In the cultivation of wheat, the amount of energy that consumed for human labor, agricultural tools/machineries, chemical fertilizers and the seed production were considered as indirect energy inputs. Therefore,

$$EI_{indirect} = HE + ME + EF + SE \ldots\ldots\ldots\ldots\ldots(7)$$

where:

$EI_{indirect}$ = Indirect energy input (MJ/ha);
HE = Human labor energy (MJ/ha);
ME = Energy consumption for machines (MJ/ha);
EF = Energy input of fertilizer (MJ/ha);
SE = Seed energy per unit area (MJ/ha).

Energy input of human labor

Relating to these applications the human labor energy is determined as indirect energy consumption (Ozturk, 2016):

$$HE = \frac{NL \times WH}{CA} \times EEL \ldots\ldots\ldots\ldots\ldots\ldots\ldots(8)$$

where:

HE = Human labor energy (MJ/ha);
NL = Number of laborer (person);
WH = Working hours (h);
CA = Cultivated area (ha);
EEL = Energy equivalent of labor (MJ/h).

Indirect energy input for tools/machines

The energy amounts that related to indirect energy consumption of agricultural tools/machineries usage are taken into account, as follows (Ozturk, 2011):

- The amount of energy that consumed for the manufacturing of tools/machineries, including to their disinterment of the raw materials, transportation and forge,
- The amount of energy that used for the materials from raw condition in factory/workshop to design of tools/machineries and their manufacturing processes,
- The amount of energy that used for the process of mending/maintenance of tools/machineries and
- The amount of energy that used for transportation and distribution of the tools/machineries

During the cultivation of wheat, the indirect energy consumption related to farming tools/machineries for each field application was determined as (Ozturk, 2016),

$$IDEM = \frac{PEM + MME + TDE}{EL \times CEW} \times NT \quad \ldots\ldots\ldots\ldots..(9)$$

where:

$IDEM$ = The indirect energy consumption related to farming tools/machineries per area (MJ/ha);
PEM = Manufacturing energy of machineries (MJ);
MME = Maintenance energy of machineries (MJ);
TDE = The amount of energy that used for transportation and distribution of the tools/machineries (MJ);
EL = Economic life of the tools/machineries (h);
CEW = The capacity of effective work (ha/h);
NT = The number transactions (quantity).

Indirect energy input for chemical fertilizer

Relating to these applications, the indirect energy consumption of chemical fertilizer usage can be expressed as:

$$EF = \sum_{l=1}^{l=s} \left[\sum_{n=1}^{n=u} \frac{\lfloor N \times N_{eq} \rfloor_n}{FA} + \sum_{n=1}^{n=u} \frac{\lfloor P_2O_5 \times P_{eq} \rfloor_n}{FA} + \sum_{n=1}^{n=u} \frac{\lfloor K_2O \times K_{eq} \rfloor_n}{FA} \right]_l \quad ..(10)$$

where:

EF = The total energy input of fertilizer (MJ/ha);
N = The amount of nitrogen fertilizer (kg);
N_{eq} = Energy consumed for N fertilizers (MJ/kg);
P_2O_5 = The amount of phosphorus fertilizer (kg);
P_{eq} = Energy consumed for P fertilizers (MJ/kg);
K_2O = The amount of potassium fertilizer (kg);
K_{eq} = Energy consumed for K fertilizers (MJ/kg);
FA = The fertilized area (ha);
n = The number of fertilizer application.

The production energy values of chemical fertilizers that used in wheat cultivation are given in Table 1.

Table 1. Energy Consumption Values for the Production of Pure Substance in Chemical Fertilizers (Ramirez & Worrel, 2006)

Chemical fertilizers	Energy consumption (MJ/kg)
Nitrogen (N)	45
Phosphorus (P_2O_5)	8
Potassium (K_2O)	5

Indirect energy input for seed

Relating to these applications, the indirect energy consumption of the seed usage can be determined from

$$SE = S\,(SPE+EPT) \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(11)$$

where:

SE = The seed energy per unit area (MJ/ha);
S = Sowing ratio (kg/ha);
SPE = Energy consump. for seed product. (MJ/kg);
EPT = Energy consumption of packaging and transportation (MJ/kg).

The energy consumption for seed production and packaging/transportation equivalent was taken into account as 25 MJ/kg for determination of the seed energy per unit area (Oren & Ozturk, 2006).

Total energy input for wheat cultivation

In wheat cultivation, the direct and indirect energy inputs were taken into account as total energy input. Thus,

$$TEI = EI_{direct} + EI_{indirect} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots(12)$$

where:

TEI = Total energy input(MJ/ha);
EI_{direct} = Direct energy input (MJ/ha);
$EI_{indirect}$ = Indirect energy input (MJ/ha).

Energy output for wheat cultivation

Two major outcomes were obtained, the wheat seed (grain) as a main product and the stem part of plant as a subsidiary product. The total amount of energy that obtained from at the end of wheat cultivation, including main and subsidiary products can be calculated as follows (Ozturk, 2011):

$$TEI = (EMP \times E_{mp}) + (ESP \times E_{sp}) \quad \ldots\ldots\ldots\ldots\ldots.(13)$$

where:

TEI = Total energy input (MJ/ha);
EMP = Amount of the main product (kg/ha);
E_{mp} = Energy of the main products (MJ/kg);
ESP = Amount of the subsidiary product (kg/ha);
E_{sp} = Energy of the subsidiary product (MJ/kg).

For the determination of the energy outputs in wheat cultivation, the energy equivalent were taken into account as 14.7 MJ/kg and for 12.5 MJ/kg for grain and wheat straw, respectively (Ozturk, 2016).

Energy efficiency for wheat cultivation

The indicators that were used for the determination of energy efficiency in the cultivation of wheat with the applications of the different mechanization techniques can be listed in Table 2.

RESULTS AND DISCUSSIONS

The energy equivalents of the input and output and indicators of energy efficiency in wheat cultivation are illustrated in Figures 1, 2 and 3, and Table 2. As can be seen from Figure 1, the total energy used in various farm inputs was 17159.5 MJ/ha for wheat cultivation. The results revealed that 7.6 h of machinery power per hectare were consumed to cultivate wheat in the research area. The human labor was 23.92 MJ/ha in wheat cultivation. The machine energy input was found to be of the order of 490.6 MJ/ha, while the man power 38.18 MJ/ha in wheat cultivation. Out of all the farm operations, seedbed preparation consumed the maximum energy followed by sowing and cultural practices, and harvesting.

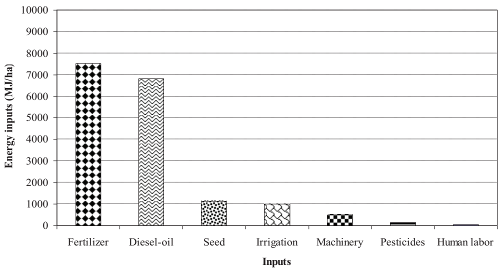

Figure 1. The Energy Inputs for Wheat Cultivation

In wheat cultivation, out of all the inputs, fertilizers have the biggest share in the total energy with a 43.8%. Fertilizer energy input is followed by the diesel-oil energy. According to Muhadar and Hignet (1982), energy used in the production of fertilizers accounts for about 40% of total energy used in agricultural production in developed countries. Most of this energy was consumed in the production of nitrogen, phosphorus and potassium fertilizers. In this study, nitrogen, phosphorus and potassium were considered as chemical fertilizer inputs. Results show that nitrogen is the most important energy source with a value of 6714.5 MJ/ha, whereas phosphorus accounted for 801.42 MJ/ha. The diesel-oil energy was mainly used for operating tractor for performing the various farm operations. The results showed that the average yield in wheat production was 2612.1 kg/ha and wheat cultivation consumed a total of 17159.5 MJ/ha input energy (Figure 2). Therefore, in wheat cultivation the energy output/input ratio, specific energy, energy productivity and net energy efficiency were 2.21, 7.18 MJ/kg, 0.14 kg/MJ and 20746.5 MJ/ha, respectively (Table 2).

However, Canakci et al. (2004) determined that the energy output/input ratio and specific energy for cultivating wheat were 2.8 and 5.24 MJ/kg in Antalya region of Turkey. Wheat is an energy frugal crop, compared to most other food crops. The energy inputs for wheat production in various regions of the U.S. calculated by Krummel and Chick of Cornell University. They found that the energy output/input ratios were 0.43, 1.66, 2.21, 3.36 and 3.75 for wheat production in Texas, New Mexico, North Dakota, Ohio and Nebraska, respectively (Briggle, 1980).

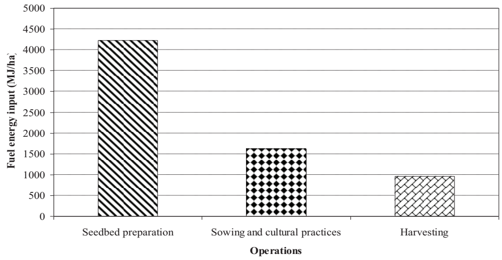

Figure 2. Fuel Energy Inputs for Wheat Cultivation

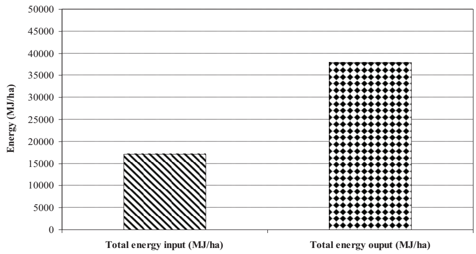

Figure 3. Energy Input and Output for Wheat Cultivation

Table 2. Energy Efficiency Indicators for Wheat Cultivation

Indicators	Definition	Unit
Energy ratio	Total achieved energy amount/Total consumed energy amount	2.21
Specific energy	The amount of total consumed energy/The total amount of harvested product	7.18 MJ/kg
Energy productivity	The total amount of harvested product/ The amount of total consumed energy	0.14 kg/MJ
Net energy efficiency	Total achieved energy amount/Total consumed energy amount	20746.5 MJ/ha

CONCLUSIONS

The total mean energy inputs as direct and indirect forms were examined in the wheat cultivation. Considerable savings could be obtained in machinery energy inputs by adopting a reduced tillage method. The average values of estimated energy output/input ratio was 2.21 for cultivation of wheat. The low level of energy output/input ratio indicated that all the farmers were not fully aware of the right production techniques or did not apply them at the proper time in the right quantity. The diesel-oil and the level of fertilizer input particularly nitrogen, were two of the most significant determinants of the total energy input for wheat cultivation. These results indicate that wheat cultivation in Turkey heavily depends on fossil fuels which in turn lead to many environmental problems. Therefore, policies should emphasize development of new technologies and alternative energy resources aiming efficient use of energy. In addition to these, the results imply that Turkish field crop production might be considered as sensitive to changes in prices and availability of fossil fuels due to mainly its significant share in total consumed energy in field crop production. The results of this study can be used by policy makers and other relevant agencies for recommendations to farmers in order to use energy more efficiently. Proper management of resources and their application at the right time can improve the efficiency of the farmers in the use of farm inputs.

REFERENCES

Briggle, L.W. (1980). *Introduction to energy use in wheat production*. Handbook of Energy Utilization in Agriculture (Editor: David Pimentel), 109–116. ISBN 0-8493-2661-3, CRC Press, Inc.

Canakci, M., Topakci, M., Akinci, I., Ozmerzi, A. (2004). Energy use pattern of some field crops and vegetable production: case study Antalya region, Turkey. *Energy Conversion and Management*, *46*, 655–666.

Dincer, I., Hussain, M.M., Al-Zahamah, I. (2004). Analysis of sectoral energy and exergy use of Saudi Arabia**.** *International Journal of Energy Research 28*(3), 205–243.

Gungor, C., Ozturk, H.H. (2019). Evaluation of fossil fuel consumption and environmental impacts in sugar beet production as a biofuel raw material. *Fresenius Environmental Bulletin*, *28*.

Muhadar, M.S., Hignett, T.P. (1982). Energy and Fertilizer: Policy implications and options for developing countries. International Fertilizer Centre, Muscle Shoals, Alabama.

Oren, M.N., Ozturk, H.H. (2006). An input-output energy analysis in field crop production in Southeastern Anatolia Region of Turkey. *Journal of Sustainable Agriculture*, *29*(1), 125–136.

Ozkan, B., Akcaoz, H., Karadeniz, F. (2003). Energy requirement and economic analysis of citrus production in Turkey. *Energy Conversion and Management*, *45*(11-12),1821–1830.

Ozkan, B., Akcaoz, H., Fert, C., (2004). Energy input-output analysis in Turkish agriculture. *Renewable Energy*, *29*, 39–51.

Ozturk, H.H. (2011). Energy management in plant production (In Turkish). HASAD Publication, Istanbul, ISBN 975-8377-78-7.

Ozturk, H.H. (2016). Energy use for wheat cultivation in Southeast Anatolia Region of Turkey. *Agricultural Mechanization in Asia, Africa and Latin America*, *47*(3), 47–53.

Ozturk, H.H. (2018). An assessment of carbon dioxide emissions related to fuel consumption for sunflower cultivation. *Fresenius Environmental Bulletin*, *27*(2), 710–719.

Ramirez, C.A., Worrell, E. (2006). Feeding fossil fuels to the soil an analysis of energy embedded and technological learning in the fertilizer industry. *Resources, Conservation and Recycling*, *46*, 75–93.

Singh, G., Singh, S., Sing, J. (2004). Optimization of energy input for wheat crop in Punjab. *Energy Conversion and Management*, *45*, 453–465.

Yamane, T. (1967). Elementary sampling theory, N.J., USA: Prentice Hall Englewood Cliffs.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

AN INVESTIGATION REGARDING THE EFFECTS OF ACCELERATION PRODUCED BY CABIN SUSPENSION SYSTEMS FITTED TO AGRICULTURAL TRACTORS UPON THE OPERATOR, IN TERMS OF OPERATOR HEALTH

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Abstract

Operators of agricultural tractors are exposed to high levels of acceleration. This paper critically examines acceleration data (in the x and y axis) collected through practical tests. Data collected shows that when cabin suspension is fitted to agricultural tractors, operators are exposed to increased levels of acceleration, up to 62% and 25% at 2.5 kph in the x and y axis respectively, when compared to the respective chassis system. Acceleration above 5.98 m/s^2 may result in the operator developing musculoskeletal disorders (MSD). This limit is exceeded in the y axis at 17 kph by a four-point active cabin suspension. Moreover, the mass of the head/torso, will multiply acceleration measured at the seat base, by 2.5, therefore, this limit is exceeded at 6.5 kph in the y axis. Consequently, up to 62% of agricultural tasks have the potential for the operator to sustain MSD. Concerns have been raised regarding the current operational speeds of agricultural tractors; this is due to the potential for acceleration above 5.98 m/s^2, to be generated and transferred to the operator and can result in MSD.

Key words*: acceleration, tractors, cabin suspension.*

INTRODUCTION

Agricultural machinery operates within harsh terrain conditions which can result in high operator exposure to mechanical shocks (acceleration of the human anatomy) (Milosavljevic et al., 2011; Waters et al., 2007; Eager et al., 2016). Moreover, certain types of agricultural machinery (tractors, material handlers) are designed to be multipurpose vehicles, thus compatible with a range of implements/attachments (Caffaro et al., 2016). Furthermore, Langer et al. (2015), adds that the equipment the agricultural tractor is operating can induce mechanical shocks, e.g. a hay/straw baler. Although, Tiemessen et al. (2007) states that an agricultural tractor towing a loaded trailer may reduce mechanical shocks being induced upon the operator. Consequently, health conditions, such as musculoskeletal disorders (MSD), can be developed, although, mechanical shocks are not solely responsible for the development of MSD, other contributing factors include; whole body vibration (WBV), repetition, awkward working position, all the contributing factors can be classified as external motions (Eager et al., 2016; Mayton et al., 2008; Kim et al., 2018; Bovenzi & Betta, 1994).

Rehn et al. (2005; 2009), states that operators of mobile equipment report the development of MSD, in the neck and shoulder region of the human anatomy, as a result of exposure to external motions.

Furthermore, Caffaro et al. (2016) and Eager et al. (2016), state that external motions can damage the spinal structure, and joints of the human anatomy.

Concerns regarding the current operational speed of agricultural machinery have been raised by three authors; Melzi et al. (2014), Achen et al. (2008), Rehn et al. (2005), who suggest that the increased travel speed has the potential to reduce operator comfort and increase exposure to external motions, leading to the development of health conditions. Furthermore, Scarlett et al. (2007), states that WBV increases in correlation with the travel speed of the vehicle.

There are four areas on most agricultural machinery where suspension can be fitted, these consist of the; operator cabin, operator seat, axles, and the tyres (Figure 1) (Melzi et al., 2014).

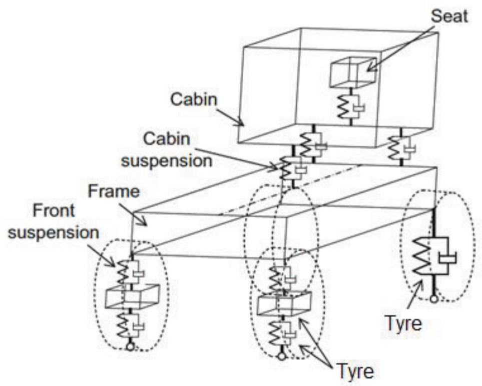

Figure 1. Diagram showing areas suspension can be fitted to agricultural vehicles (Melzi et al., 2014)

Cuong et al. (2013) and Melzi et al. (2014) state that agricultural tractor and machinery manufacturers, do not fit advanced cabin suspension systems to the operator cabin as standard equipment. Instead rubber based anti-vibration mounts are used (Lyashenko et al., 2016). This is despite Hansson (1995) suggesting that suspending the whole operator cabin can reduce mechanical shock transferred to the operator, additionally, Sim et al. (2017) added that operator weight does not detrimentally affect the performance of this type of suspension system. Operator weight is a limiting factor to seat suspension therefore, this can reduce the performance/effectiveness of seat suspension (Sim et al., 2017). According to Cuong et al. (2013), axle suspension can reduce the mechanical shocks experienced by the vehicle being transferred to the operator. Although Achen et al. (2008) states that this type of suspension system can further increase the cost of the vehicle, due to requiring to redesign structural components. Depending on the operating pressure of the tyres, a small amount of mechanical shock can be absorbed by the tyres (Tiemessen et al., 2007). Although, reducing the tyre inflation pressure, to reduce mechanical shocks, in turn increases the heat generated by the tyres and the rolling resistance of the tyres (Donati, 2002).

This paper will evaluate acceleration produced in the *x* and *y* axis (Figure 2), for three variants of cabin suspension. According to Kim et al. (2018), these two axes are not required to be suspended to comply with ISO 2631.

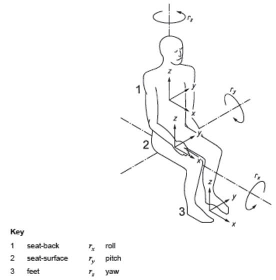

Figure 2. Diagram showing the *x*, *y*, *z* Axis Orientation, (ISO, 1997)

MATERIALS AND METHODS

Four individual elements were used to collected information and data to fully evaluate acceleration produced in the *x* and *y* axis. These four elements consisted of; investigating academic literature, conducting surveys which focused on agricultural workers, conducting interviews with agricultural manufacturers and finally completing practical tests.

The academic literature has been investigated to provide an understanding regarding the current suspension systems used on agricultural tractors and machinery. Additionally, academic literature provided an insight into the health implications for the operator as a result of long-term exposure to mechanical shocks and acceleration.

Surveys were conducted with agricultural workers and agricultural tractor and machinery operators. The surveys were distributed across the North West of the United Kingdom between December 2017 and February 2018 and focused on collecting both qualitative and quantitative data surrounding agricultural tractor operation. Additionally, the surveys collected data about the suspension systems fitted to their (the survey respondents) agricultural tractors, the final question in the survey asked if the respondent had awareness of long-term health effects as a result of exposure to mechanical shock and acceleration.

The interviews with eight agricultural manufacturers were conducted at LAMMA (Lincolnshire Agricultural Machinery Manufacturers Association) on the 17th January 2018, the interviews collected information regarding the current level of cabin suspension systems, which agricultural manufactures install to their agricultural tractors.

Practical tests on three types of cabin suspension systems were completed between November 2017 and February 2018. The practical tests were designed to determine the acceleration in both the *x* and *y* axis. The data has been collected using a data logger, which recorded acceleration (displayed as m/s^2), measuring at 100 Hz. Three types of cabin suspension systems were tested (mechanical, four and three point active). Semi active cabin suspension is another suspension system outlined by Van Iersel (2010), although none were available to test. The three cabin suspension systems were produced by the same agricultural manufacturer, although due to available resources, the current working hours of the agricultural tractors differed when the tests commenced. Additionally, the tyre condition, chassis size and machine age differed between the three agricultural tractors which were tested. The agricultural tractors which were tested all had the same front axle design (suspended beam axle), and the tyre pressures were set to the manufacture's recommendations.

The practical tests involved driving each agricultural tractor over a wooden obstacle which measured 1032 x 280 x 140 mm (length x width x height), at two test speeds, 2.5 and 5 kph. There were three configurations of the wooden obstacle (at each forward speed) allowing the left, right hand and both wheels to negotiate the wooden obstacle; this is shown below in Figure 3 as 1, 2, 3, respectively.

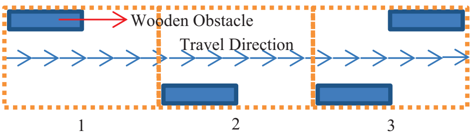

Figure 3. The three test track configurations

The test track varied in length between 11 and 13 metres depending on the forward travel speed 2.5 or 5 kph, respectively. The obstacles shown in Figure 3 above were positioned 4 meters into the course; this allowed the agricultural tractors to reach the desired speed of 2.5 or 5 kph prior to reaching the obstacle. All three agricultural tractors were fitted with a CVT gearbox and cruise control; this allowed consistent travel speeds to be selected.

To provide a comparison between the acceleration produced by the cabin and the chassis, two mounting points for the data logger were selected which could be replicated on all three agricultural tractors, these are shown below in (Figure 4 and 5).

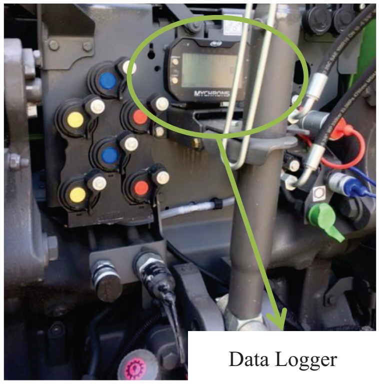

Figure 4. Data Logger Location (Chassis)

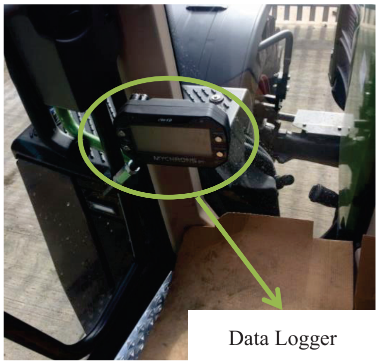

Figure 5. Data Logger Location (Cabin)

The practical tests were repeated 5 times, to provide accurate data collection, and allowing a mean to be calculated. On completion of the data collection Microsoft Excel has been used to post process the data. Post processing of the data determined the mean peak acceleration, for each wavelength on the acceleration traces of each test track configuration (Figure 3), completed at each travel speed. Once the mean peak acceleration had been calculated, for each test track configuration, the mean peak acceleration for the tests completed at the two travel speeds were combined to be provide a mean acceleration at a specific travel speed (2.5 and 5 kph) for each cabin suspension system tested. This data provided the bases to calculate an estimation of acceleration produced at travel speeds higher than 5 kph.

RESULTS AND DISCUSSIONS

Sorainen et al. (1998), states that mechanical shocks (acceleration) are unavoidable when operating machinery on uneven terrains, therefore, the correct equipment to protect the operator should be utilised. Hansson (1995) suggests cabin suspension should be fitted to agricultural tractors, reducing the operator exposure to acceleration.

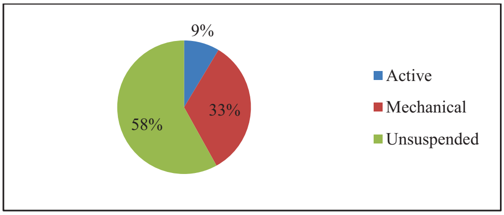

Figure 6. Chart showing the cabin suspension system fitted to the agricultural tractors surveyed

As shown in Figure 6, 58.0% of the cabins fitted to the agricultural tractors surveyed were unsuspended. This is despite one of the manufacturers interviewed commenting that customers expect cabin suspension systems to be offered as standard on large agricultural tractors. Although, as shown in Figure 7, the mean age of the agricultural tractors fitted with an unsuspended cabin is 19.56 years. Additionally, as show in Figure 7, the mean age of the agricultural tractors with fully suspended axles, is higher than the mean age of the agricultural tractors fitted with active cabin suspension, this could be explained due to only 4.0% of the agricultural tractors surveyed being fitted with fully suspended axles. Moreover, as shown in Figure 8, the mean yearly operational hours for the unsuspended cabins is 432, as expected Figure 8 shows that the yearly operational hours positively correlate to the increased level of suspension system installed to the vehicle. Furthermore, Figure 10, shows that unsuspended cabins are used in rough terrain conditions (yards and fields) for the highest yearly percentages, 21.46% and 14.94%, respectively, when compared against the other suspension systems which were considered in this report. Therefore, there is a potential for increased operator exposer to acceleration in these terrain conditions.

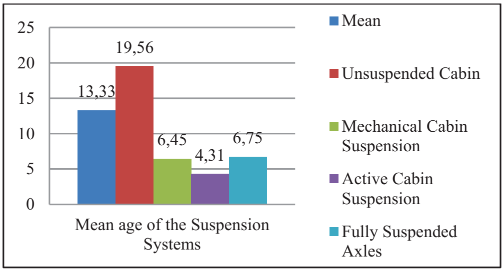

Figure 7. Chart showing the mean age of the agricultural tractors surveyed, categorised by suspension systems

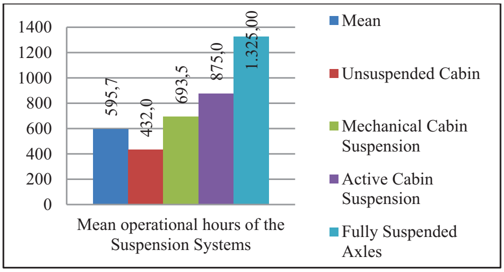

Figure 8. Chart showing the mean operational hours of the agricultural tractors surveyed, categorised by suspension systems

According to Johanning (2015), there is a correlation between increased spinal disorders and increased intensity/ duration of exposure to mechanical shocks. Milosavljevic et al. (2010), adds this is due to increased spinal loading in the lumbar vertebrae of the spinal structure, (Figure 9). This increased loading is through the intervertebral end plates (the top and bottom of each vertebral body), and the lumbar intervertebral discs, this is shown below in Figure 11.

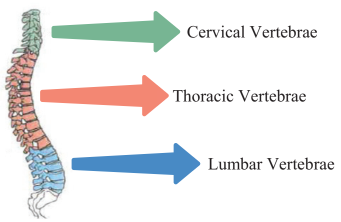

Figure 9. The vertebral column (Marieb, 1998)

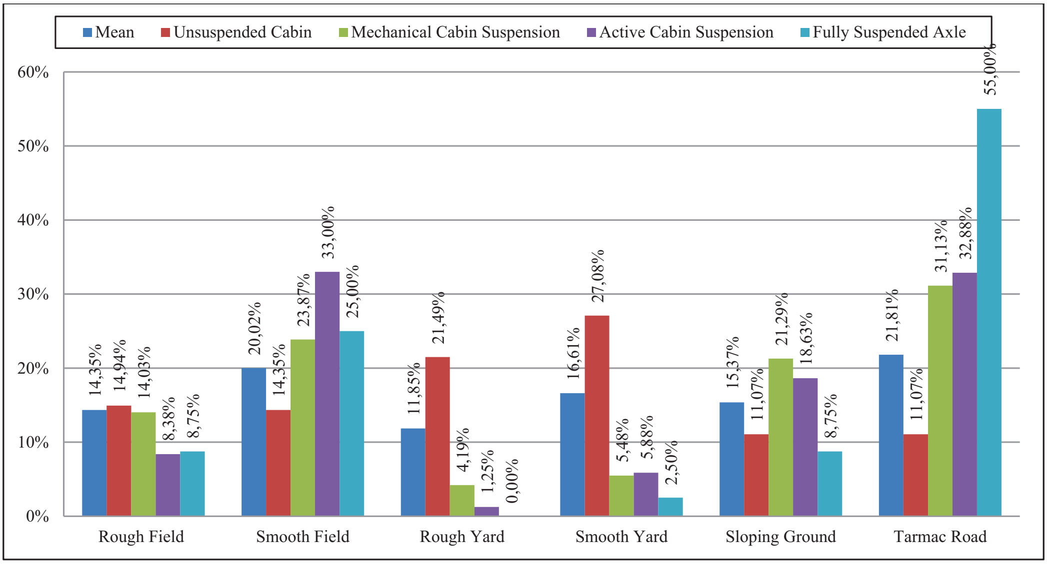

Figure 10. Chart showing the mean percentage of yearly operation in given terrains, for each suspension system respectively

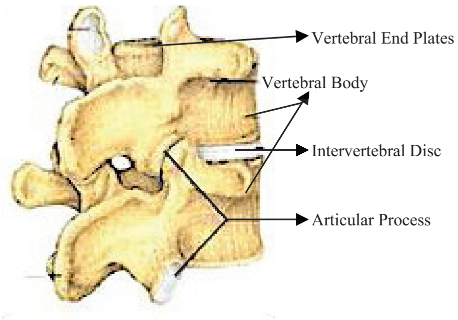

Figure 11. Annotation of the Lumber Vertebrae (Marieb, 1998)

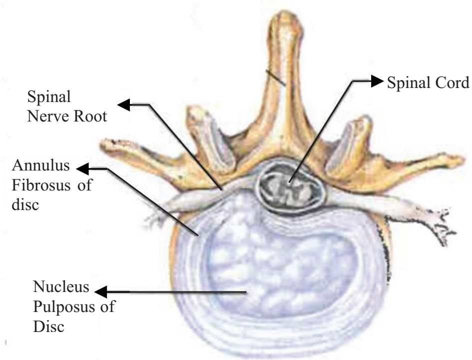

Figure 12. Annotation of the vertebrae (Marieb, 1998)

Waters et al. (2007) and Bovenzi (2006) state that the lumber region is the first area of the spinal structure which is susceptible to damage when exposed to mechanical shock. Additionally, Wikström et al. (1994) and Waters et al. (2008) state that when the spine is in a twisted position the spinal structure is at a higher risk of being damaged by mechanical shocks. Moreover, this is a common occurrence for operators of agricultural machinery, as they are required to frequently turn their head to inspect the rear of the machine (Wikström et al., 1994). Furthermore, Waters et al. (2007), states that vehicle induced mechanical shocks contributes to 36.0% of lower spinal injuries.

Waters et al. (2007) states that the vertebral end plates can deform/misalign under load (mechanical shock), this allows the hydrated nucleus pulposus (Figure 12) to herniate the endplate. This results in a pressure reduction of the nucleus pulposus, the decompressed disc bulges and loses height (degeneration of the intervertebral disc). Additionally, Bovenzi and Betta (1994), states that degeneration of the intervertebral discs is a contribution factor to lower back pain. Moreover, Kim et al. (2018) and Milosavljevic et al. (2010) add the balance and vision of the operator may be reduced as a result of degenerating intervertebral discs. Furthermore, Sorainen et al. (1998), states that intervertebral discs loose water throughout the day, and exposure to mechanical shocks accelerates the reduction of water in the intervertebral discs. When the intervertebral discs loose water they start to narrow, this reduces the function of the spine. Additionally, the nutrient pathways to the spinal articular processes are damaged with exposure to mechanical shock (Milosavljevic et al., 2010).

The intervertebral discs are the largest none vascular part of the human anatomy which has no biological repair process, and therefore, should be protected from exposure to mechanical shock (Wikström et al., 1994). Moreover, operator exposure to acceleration can decrease operator productivity (Taghizadeh-Alisaraei, 2017).

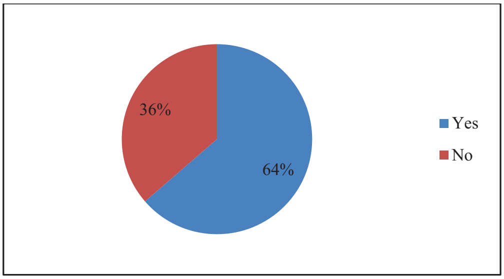

Figure 13. Chart showing the respondents awareness to health effects after exposure to mechanical shocks

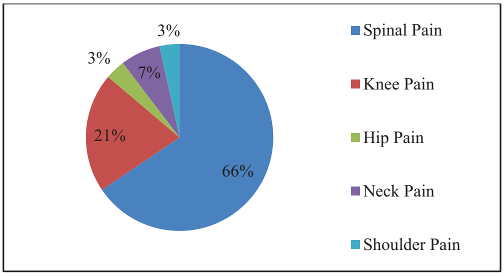

Figure 14. Chart showing areas of the human anatomy which respondents felt the prevalence of pain when exposed to mechanical shocks, whilst or shortly after operating agricultural machinery

As shown in Figure 13, 64.0% of respondents were aware of the potential to develop health issues as a result of exposure to mechanical shocks. Additionally, 66.0% of these respondents related exposure to mechanical shocks to developing spinal pain, as shown in Figure 14. Although, as shown in Figure 13 and described by Solecki (2012), there is a proportion of the respondents who are not aware of the health effects as a result of exposure to mechanical shocks.

According to Wikström et al. (1994), acceleration/mechanical shock above 5.98 m/s^2 (threshold limit) can cause damage to the intervertebral discs, additionally, mechanical shock/acceleration in the *x* and *y* axis are accelerated by the mass of the head/torso. Additionally, Kim et al. (2018) adds that acceleration measured at the head/torso is 2.5 times greater than acceleration measured at the base of the seat.

As shown in Table 1 below, in all but one test scenario the cabin produced higher levels of acceleration, than the acceleration measured at the chassis. Furthermore, the increased acceleration of the cabin is up to 62.0% and 25.0% higher than the chassis at 2.5 kph, in the *x* and *y* axis respectively, for the mechanical cabin suspension system tested. The difference between cabin and chassis acceleration (in the *x* axis) does reduce with the implementation of increased cabin suspension systems. Although, acceleration in the *y* axis does not display the same trend (with regards to reduction), seen for cabin/chassis acceleration in the *x* axis.

Table 1. Summary of the practical test data

		X Axis		Y Axis	
Cabin Suspension System	Travel Speed (kph)	2.5	5	2.5	5
Mechanical (Cabin Acceleration) (m/s^2)		0.745	0.981	0.824	1.530
Mechanical (Chassis Acceleration) (m/s^2)		0.461	0.686	0.657	1.402
Percentage Difference (Cabin/Chassis)		*62%*	*43%*	*25%*	*9%*
Four Point Active (Cabin Acceleration) (m/s^2)		0.637	0.912	0.951	1.814
Four Point Active (Chassis Acceleration) (m/s^2)		0.490	0.657	1.049	1.167
Percentage Difference (Cabin/Chassis)		*30%*	*39%*	*-9%*	*55%*
Three Point Active (Cabin Acceleration) (m/s^2)		0.588	0.775	0.598	1.451
Three Point Active (Chassis Acceleration) (m/s^2)		0.500	0.755	0.481	1.089
Percentage Difference (Cabin/Chassis)		*18%*	*3%*	*24%*	*33%*

Figure 17 and Figure 18 shows an estimated acceleration in the *x* and *y* axis for travel speeds from 2.5 to 40 kph, for the three tested cabin suspensions, in addition, an estimated acceleration produced by an unsuspended cabin is shown. The estimated acceleration produced by an unsuspended cabin is a combination of the estimated acceleration measured on all of the chassis tested.

As shown in Figure 17, acceleration at speeds above the maximum test speed (5 kph), is below the 5.98 m/s^2 threshold limit, for all the cabin suspension systems considered. Although when the acceleration data is multiplied by 2.5, to determine the acceleration of the head/torso, the 5.98 m/s^2 threshold limit, is exceeded at 20, 18.5, 27, 25 kph by the mechanical, four/three-point active cabin suspension system and an unsuspended cabin respectively (in the *x* axis). Furthermore, at 40 kph the three-point active suspension system reduces the acceleration in the *x* axis by 9.6%, when compared to the acceleration produced by an unsuspended cabin.

As shown in Figure 18, acceleration measured in the *y* axis is up to 50.0% greater than acceleration measured in the *x* axis, (at 2.5 kph). Additionally, the threshold limit of 5.98 m/s^2, is exceeded by all the cabin suspension systems considered in this report between 2.5 and 30 kph. Moreover, when the acceleration data is multiplied by 2.5, to determine the acceleration of the head/torso. The 5.98 m/s^2 threshold limit is exceeded between 2.5 and 11 kph. As shown in Figure 18 the unsuspended cabin produced the lowest levels of acceleration in the *y* axis, followed by the mechanical, three/four-point active cabin suspension system. Despite the mechanical cabin suspension system recording the second lowest acceleration trace in the *y* axis at 40kph, the acceleration trace is 29.0% higher than the acceleration produced by an unsuspended cabin. Moreover, the three/four-point active cabin suspension acceleration trace is 39.5% and 41.7% higher than the acceleration produced by an unsuspended cabin, at 40 kph.

As shown in Figure 15, between 52.0% and 62.0% of the given field operations completed with agricultural machinery using one of the three cabin suspension systems tested have a potential for the operator to cause damage to their intervertebral discs. Although, it is interesting to note due to the low speed of the field operations (maximum of 10 kph), the acceleration threshold of 5.98 m/s^2 is not exceeded by an unsuspended cabin, in the *y* axis. According to Taghizadeh-Alisaraei (2017), operator exposure to acceleration is greater in developing countries, due to agricultural tractors not utilising the latest technologies, and are being operated for long periods of time in rough terrain conditions.

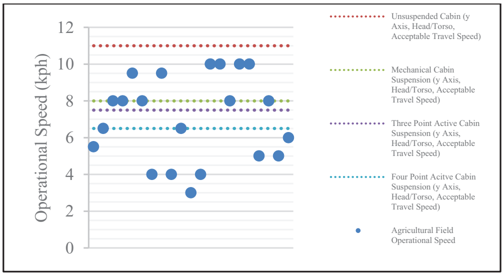

Figure 15. Chart showing the typical operational speeds for a range of field activities described by Landers (2000), additionally, the chart shows the acceptable travel speed (ensuring acceleration is not above 5.98 m/s^2 in the *y* axis (head/torso), preventing damage occurring to the operator's intervertebral discs) for all of the cabin suspension systems considered in this report

UK Legislation, amended in 2015, increases the travel speed of conventional agricultural tractors to 40 kph, from 32 kph without any design alterations (Collins, 2015). As shown in Table 2, increasing the travel speed by 8 kph, the estimated acceleration (in the *y* axis for the head/torso) is increased by up to 115.0% (32 to 40 kph). Additionally, at 40 kph the estimated acceleration, (in the *y* axis for the head/torso), is up to 481.0% above the 5.98 m/s^2 threshold limit described by Wikström et al. (1994).

As shown above in Figure 8, the surveyed agricultural tractors are operated between 432.0 and 1,325.0 hours per year, depending on the suspension system installed to the vehicle. Furthermore, Johanning (2015), Bovenzi (1996), and Kittusamy & Buchholz (2004), state that the period of exposure to acceleration, is a high contribution to operators sustaining lower back injuries. Moreover, as shown in Figure 16, the operational hours of agricultural tractors vary depending on the month. This is due to the agricultural activities which require an agricultural tractor peaking twice a year, (month 4 and 8), these activities include soil cultivation, harvesting and produce transport. Additionally, Solecki (2012), states that agricultural tractors may be operated for up to 16.3 hours during peak operational periods of the year.

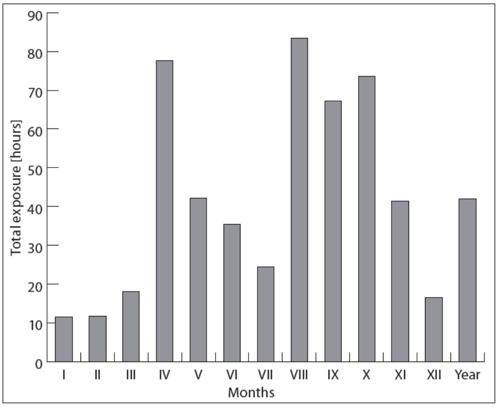

Figure 16. Monthly Operation Hours of agricultural tractors (Solecki, 2012)

Commercial vehicles which are used for hire or reward are subject to stringent daily and weekly operator hour regulations, additionally, regular breaks are required to be taken (EOS, 2017). Commercial vehicles and agricultural tractors used for purposes relating to; agriculture, horticulture, forestry, farming, or fishery, are exempt from the EU operator hour regulations, as-long-as the vehicle is operated within a 100km radius of the business site (Government Digital Service, 2015). Although, the operators are still subject to UK domestic driver hour regulations where a daily maximum of 10 hours applies, only time operating on public highways are counted towards the 10 hours daily limit (Government Digital Service, 2015). Moreover, operators are eligible to opt-out from the UK domestic driver hour regulations (EOS, 2017). Therefore, operators may be exposed to acceleration for extended periods of time.

Table 2. Table showing the % increase in estimated acceleration between 30 and 40 kph travel speed

	x Axis	32 kph (m/s^2)	% above 5.98 m/s^2	40 kph (m/s^2)	% above 5.98 m/s^2	Percentage increase between acceleration above 5.98 m/s^2 at 32 kph and 40 kph
Estimated Acceleration	Unsuspended	3.02	-49%	3.71	-38%	11%
	Mechanical	3.52	-41%	4.28	-29%	13%
	Four Point	3.87	-35%	4.76	-20%	15%
	Three Point	2.79	-53%	3.38	-43%	10%
	x Axis					
Estimated Head/Torso Acceleration	Unsuspended	7.55	26%	9.27	55%	29%
	Mechanical	8.81	47%	10.69	79%	31%
	Four Point	9.70	62%	11.90	99%	37%
	Three Point	6.96	16%	8.45	41%	25%
	y Axis					
Estimated Acceleration	Unsuspended	6.54	9%	8.11	36%	26%
	Mechanical	9.16	53%	11.41	91%	38%
	Four Point	11.13	86%	13.90	132%	46%
	Three Point	10.67	78%	13.40	124%	46%
	y Axis					
Estimated Head/Torso Acceleration	Unsuspended	16.33	173%	20.26	239%	66%
	Mechanical	22.54	277%	28.54	377%	100%
	Four Point	27.84	366%	34.74	481%	115%
	Three Point	26.66	346%	33.49	460%	114%

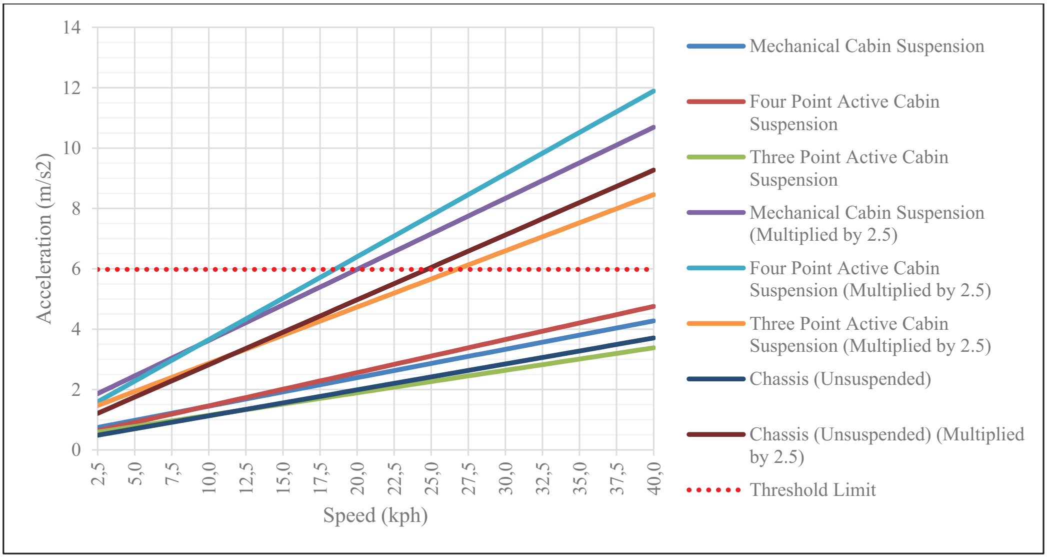

Figure 17. Chart showing the Estimated Acceleration in the *x* Axis

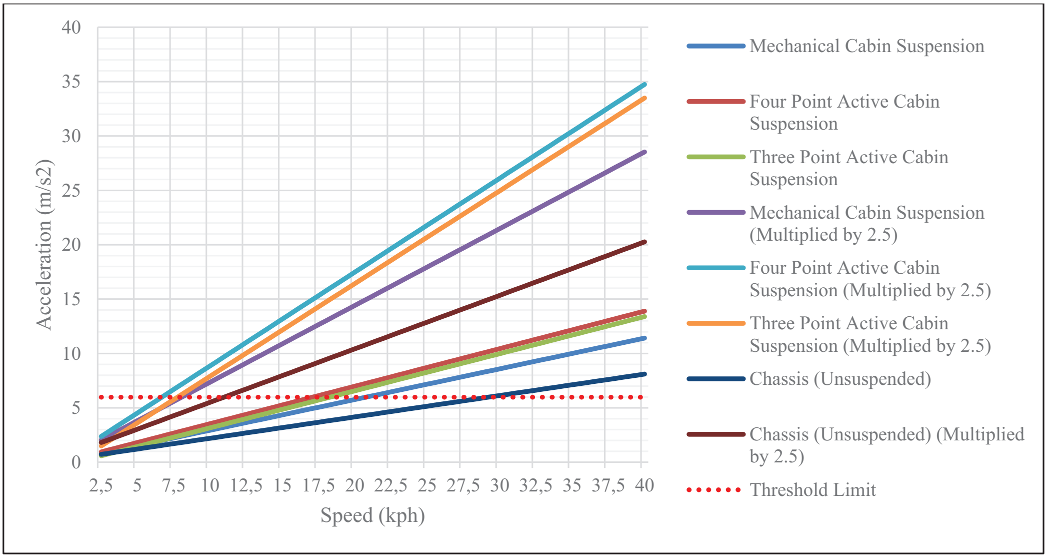

Figure 18. Chart showing the Estimated Acceleration in the *y* Axis

CONCLUSIONS

Surveys have shown that 58.0% of agricultural tractors operating in the North West of the UK, are not fitted with cabin suspension. Furthermore, the surveys have outlined that the agricultural tractors without cabin suspension complete the highest yearly operation (36.4%) in rough terrain conditions, when compared to the other cabin suspension systems considered.

Acceleration above 5.98 m/s^2, has the potential to cause damage to the fibres of the intervertebral discs. Data which has been collected shows that this limit is exceeded in the *y* axis between 2.5 and 30 kph for all of the cabin suspension considered in this report. Although, when the data is multiplied by 2.5 to estimate the acceleration experienced by the head/torso, this limit is exceeded in the *x* axis between 2.5 and 25 kph and exceeded in the *y* axis between 2.5 and 11 kph. Consequently, up to 62.0% of field activities have the potential for the operator to exceed the 5.98 m/s^2 threshold limit.

Only the estimated acceleration in the *x* axis, produced by the 3 point active cabin suspension is lower than the estimated acceleration produced by an unsuspended cabin, although this is only a reduction of 9.6% at 40 kph. All the other tested cabin suspension systems produce an increased level of acceleration when compared to an unsuspended cabin.

Moreover, the collected data has shown that acceleration in the *y* axis is up to 50.0% higher than acceleration measured in the x axis.

Therefore, to protect operators from exposure to acceleration in the *x* and *y* axis, improvements with regards to cabin suspension systems fitted to the cabin is required, due to exposure to acceleration having a degenerative effect on the operator's spinal structure (Waters et al., 2007; Bovenzi, 2006).

Concerns have been raised regarding the increase in operations speeds (32 to 40 kph) of agricultural tractors on public roads in the UK, due to a potential increase in the estimated acceleration of the head/torso in the *y* axis of 115.0%.

Furthermore, agricultural tractor operators, operating on private land are exempt from daily and weekly hour regulations, and therefore extended periods of exposure to acceleration could be encountered (up to 16.5 hours daily, during peak periods of the year) (Solecki, 2012).

Finally, Kim et al. (2018), states that currently the industry standard (ISO 2631), only requires the z axis to be suspended, even though this report has outlined that both the *x* and *y* axis have the potential to produce excessive levels of acceleration.

REFERENCES

Achen, A., Toscano, J., Marjoram, R. Clair, K., McMahon, B., Goelz, A., & Shutto, S. (2008). Semi-Active Vehicle Cab Suspension using Magnetorheological (MR) Technology. *Proceedings of the JFPS International Symposium on Fluid Power*, *7*, 561–564.

Bovenzi, M. (2006). Health risks from occupational exposure to mechanical vibration. *La Medicina del Lavoro, 27*, 58–64.

Cuong, D., Zhu, S., Zhu, Y. (2013). Effects of tyre inflation pressure and forward speed on vibration of an unsuspended tractor. *Journal of Terramechanics*, *50*, 185–198.

Donati, P. (2002). Survey of Technical Preventative Measures to Reduce Whole-Body Vibration Effects when Designing Mobile Machinery. *Journal of Sound and Vibration*, *253*, 169–183.

Eager, D., Pendrill, A., Reistad, N. (2016). Beyond velocity and acceleration: jerk, snap and higher derivatives. *European Journal of Physics*, *37*.

Hansson, P. (1995). Optimization of agricultural tractor cab suspension using the evolution method. *Computers and Electronics in Agriculture*, *12*, 35–49.

Johanning, E. (2015). Whole-body vibration-related health disorders in occupational medicine – an international comparison. *Ergonomics*, *58*, 1239–1252.

Kittusamy, N., & Buchholz, B. (2004). Whole-body vibration and postural stress among operators of construction equipment: A literature review. *Journal of Safety Research*, *35*, 255–261.

Langer, T., Ebbesen, M., Kordestani, A. (2015). Experimental analysis of occupational whole-body vibration exposure of agricultural tractor with large square baler. *International Journal of Industrial Ergonomics*, *47*, 79–83.

Marieb, E. (1998). *Human anatomy & physiology*. 4th ed. California, USA: Benjamin/Cummins Science Publishing.

Mayton, A., Kittusamy, N., Ambrose, D., Jobes, C., Legault, M. (2008). Jarring/jolting exposure and musculoskeletal symptoms among farm equipment operators. *International Journal of Industrial Ergonomics*, *38*, 758–766.

Rehn, B., Nilsson, T., Lundström, R., Hagberg, M., Burström, L. (2009). Neck pain combined with arm pain among professional drivers of forest machines and the association with whole-body vibration exposure. *Ergonomics, 52*, 1240–1247.

Scarlett, A., Price, J., Stayner, R. (2007). Whole-body vibration: Evaluation of emission and exposure levels arising from agricultural tractors. *Journal of Terramechanics*, *44*, 65–73.

Sim, K., Lee, H., Yoon, J., Choi, C., Hwang, S. (2017). Effectiveness evaluation of hydro-pneumatic and semi-active cab suspension for the improvement of ride comfort of agricultural tractors. *Journal of Terramechanics*, *69*, 23–32.

Sorainen, E., Penttinen, J., Kallio, M., Rytkönen, E., Taattola, K. (1998). Whole-Body Vibration of Tractor Drivers During Harrowing. *American Industrial Hygiene Association Journal*, *59*, 642–644.

Taghizadeh-Alisaraei, A. (2017). Analysis of annoying shocks transferred from tractor seat using vibration signals and statistical methods. *Computers and Electronics in Agriculture*, *141*, 160–170.

Tiemessen, I., Hulshof, C., Frings-Dresen, M. (2007). An overview of strategies to reduce whole-body vibration exposure on drivers: A systematic review. *International Journal of Industrial Ergonomics*, *37*, 245–256.

Van Iersel, S. (2010). Passive and semi-active truck cabin suspension systems for driver comfort improvement. Master's thesis. Eindhoven University of Technology Department of Mechanical Engineering Dynamics & Control.

Waters, T., Genaidy, A., Viruet, H., Makola, M. (2008). The impact of operating heavy equipment vehicles on lower back disorders. *Ergonomics, 51*, 602–63.

Scientific Papers. Series A. Agronomy, Vol. LXII, No. 1, 2019
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

THE QUALITY OF BIOMASS AND FUEL PELLETS FROM JERUSALEM ARTICHOKE STALKS AND WHEAT STRAW

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Abstract

In recent years, the biomass market has constantly increased. The densification of plant biomass would contribute to improving its efficiency as a fuel by increasing its homogeneity and allowing a wider range of lignocellulosic materials to be used as fuel. The eco-friendly solid fuels, such as pellets, have become rapidly a viable alternative to fossil fuels, due to their high energy content, which makes them suitable for use by small households and by industrial consumers. The knowledge of the physical and mechanical properties of biomass is important for the design and efficient operation of equipment for handling, storing and processing such materials. Jerusalem artichoke, Helianthus tuberosus, is native to North America. Its tubers were previously used as raw material, food, folk remedy and animal fodder. Its potential yield and low requirements meant that it could be of interest in the renewable energy sector, tubers can be used for biogas or ethanol production, and the aboveground parts – for pellets and briquettes. The objective of this research was to evaluate some physical and mechanical properties of dry biomass and pellets from Jerusalem artichoke stalks and wheat straw collected from the experimental field of the National Botanical Garden (Institute), Chişinău. The physical and mechanical properties were determined according to the European standards accepted in the Republic of Moldova; the production of solid fuels, pellets – by the equipment developed in the Institute of Agricultural Technique "Mecagro", Chişinău. The pellets were produced from Jerusalem artichoke biomass and mixture of Jerusalem artichoke and wheat straw with a percentage: 0%, 30%, 50%, 70% and 100%. It was determined that the bulk density of the chaffs milled by a 6 mm sieve ranged from 163 to 231 kg/m^3, the ash content – from 2.12 to 4.93%, the gross calorific value - from 17.4 to 19.1 MJ/kg. The biomass of the species Helianthus tuberosus was characterized by high gross calorific value and moderate ash content. The physical and mechanical properties of fuel pellets varied depending on the mixture ratio: the moisture content ranged from 13.8 to 14.1%, the bulk density - from 582 to 685 kg/m^3, the specific density - from 880 to 1008 kg/m^3 and the net calorific value - from 15.6 to 17.71 MJ/kg.

Key words: *biomass, Jerusalem artichoke stalks, pellet, physical and mechanical properties, wheat straw.*

INTRODUCTION

In an era of accelerating change, the imperative to limit climate change and achieve sustainable economic growth is strengthening the momentum of the global energy transformation. The global energy landscape continues to shift toward decarbonisation, digitalization and decentralization in combination with improving energy efficiency. The world's demand for energy grew by 2.1% in 2017, compared with 0.9% in the previous year; a quarter of the rise was met by renewables, according to new data from the International Energy Agency (IEA).

In recent years, the biomass market has constantly increased in the world, and the production of renewable energy has developed at the regional and national levels too. Phytomass, agricultural crop residues along with energy crops, represent a good opportunity, since they can be converted into various types of fuels or energy via thermal, physical and biological processes. Harvested phytomass, because of its irregular shape and size and low bulk density, is very difficult to handle, transport, store and use in its original form. The densification of plant biomass would contribute to improving its efficiency as a fuel by increasing its homogeneity and allowing a wider range of lignocellulosic materials to be used as fuel. Kaliyan & Morey (2009) conducted a broad review of factors affecting the strength and durability of densified biomass products and made recommendations regarding the selection of

processing parameters in the production of agglomerates of acceptable quality and durability. The eco-friendly solid fuels, such as pellets, have become rapidly a viable alternative to fossil fuels, due to their high energy content, which makes them suitable for use by small households and by industrial consumers (Gudîma, 2018).
Currently, cereal straw is the most commonly utilized raw material for the production of heat energy by direct combustion process in Moldova. Cereal straw is sometimes used as feed, bedding for animals and as a soil amendment, incorporated into the ploughed layer or used as mulch. There are also pellets produced from straw, but they are not so common because such pellets produce more ash than pellets from wood (3-5%), which causes users to clean and service their boilers more frequently (Smaga et al., 2018).
The identification, mobilisation and cultivation of new species for biomass production depend on the climate and soil conditions, the availability and cost of reproductive material, the market demand for the given type of bio fuel, technological, organizational and economic conditions and other factors. The debate related to agricultural land to be used for food or fuel production in a world that still sees starvation and malnutrition of a part of its population is fierce and the identification and cultivation of species with multiple utility is a subject of topical interest (Roman et al., 2016).
One such species is Jerusalem artichoke, otherwise known as topinambour, *Helianthus tuberosus* L., which belongs to the family *Asteraceae*, is native to North America, has strong, vigorous stems, sometimes branched at the base, grows 2.5-3.0 m tall, but can reach even 5 m. Its large leaves reach the length of 20 cm, and are arranged on the opposite sides of the stem, alternately to one another; they are broad ovoid-acute in shape, with dentate edges, covered with rough hair. It produces flower heads with bright yellow petals, which resemble sunflower, with a bunch of small florets (10-20) in the center. The fruit is a hairy achene containing a mottled black or brown seed, 5 mm long x 2 mm wide. In the underground part, there are unevenly shaped tubers, round or elongated, with bumps, ranging in size between 2 and 10 cm, their color can vary from brown to white, red and even purple.
The analysis of literature sources has revealed that Jerusalem artichoke can be useful in many ways. It is interesting because of the high productivity and possibilities of cultivation on marginal land, the ability to absorb carbon from the air and to release oxygen, the suitability for the creation of effective green belts around industrial centers. This species is studied and cultivated in Europe, Asia and America, has many application areas such as: tubers in human nutrition and animal feed, source of inulin for pharmaceutical industry, functional and dietary products, biologically active and food supplements; aboveground parts as green or ensiled forage, raw material for the production of various chemicals etc. (Kays &Nottingham, 2007; Heuze et al., 2015; Ţîţei et al., 2013; Ţîţei & Coşman, 2016). Its potential yield and low requirements mean that it can be of interest in the renewable energy sector (Gunnarsson et al., 2014; Kowalczyk-Juśko et al. 2012; Szostek et al., 2018; Ţîţei, 2015; Zapalowska & Bashutska, 2017).
The growing market demand for renewable fuels has stimulated the search for new biomass types suitable for pelletizing. Biomass materials are frequently mixed to obtain and improve the properties and the quality of pellets (Gageanu et al., 2017; Gudîma, 2018; Lisowski et al., 2018; Miranda et al., 2015). A literature review also points to the need to identify new kinds of fuels by mixing well-known and new materials, and the characteristics of these materials must be established. There is little information in the existing literature regarding the properties of compaction of Jerusalem artichoke stalks in pellet production and the quality characteristics of pellets from mixtures of Jerusalem artichoke and cereal straws.
The objective of this research was to evaluate some physical and mechanical properties of dry biomass and pellets: from Jerusalem artichoke stalks and wheat straw, and their mixtures.

MATERIALS AND METHODS

The cultivar 'Solar' of Jerusalem artichoke, *Helianthus tuberosus,* created in the National Botanical Garden (Institute), registered in

2014, in the Catalogue of Plant Varieties and patented in 2016, by the State Agency on Intellectual Property of the Republic of Moldova patent nr. 205/31.05.2016, served as research subject.

Jerusalem artichoke stalks were collected from the experimental field at the end of January. Stalks and wheat straw bales were chopped. The chopped biomass of pure Jerusalem artichoke stalks and wheat straw and their biomass mixtures (30%, 50%, 70%) were milled in a beater mill equipped with a sieve with diameter of openings of 6 mm using an equipment developed in the Institute of Agricultural Technique "Mecagro", Chişinău. The pellets were produced from Jerusalem artichoke biomass and a mixture of Jerusalem artichoke and wheat straw with a percentage: 0%, 30%, 50%, 70% and 100%. A scientific research on the biomass for the production of solid biofuel was carried out and it included several steps: the moisture content of the plant material was determined by CEN/TS 15414 in an automatic hot air oven MEMMERT100-800; the content of ash was determined at 550ºC in a muffle furnace HT40AL according to CEN/TS 15403; automatic calorimeter LAGET MS-10A with accessories was used for the determination of the calorific value, according to CEN/TS 15400; the particle size distribution was determined using standard sieves, the collected particles in each sieve were weighed; the cylindrical containers were used for the determination of the bulk density, calculated by dividing the mass over the container volume; the pelleting was carried by the equipment developed in the Institute of Agricultural Technique "Mecagro"; the mean compressed (specific) density of pellets was determined immediately after removal from the mould as a ratio of measured mass over calculated volume.

RESULTS AND DISCUSSIONS

It is known that moisture and leaf share in the biomass harvested to produce solid fuel influence the costs of transport, storage, drying and processing, and the thermophysical properties of biofuel reduce the final usable energy and thus the efficiency of the energy system, contributing at the same time to the increased emission of pollutants. There is a practical limit of autogenous combustion at about 67% moisture. The high amount of moisture in biomass decreases the heating value of fuel, which in turn reduces the conversion efficiency, as a large amount of energy would be used for the initial drying step during the conversion processes (Marian, 2016).

Table 1. Moisture and leaf contents in the aboveground parts of *Helianthus tuberosus*

Harvesting period	*Helianthus tuberosus*	
	moisture content, %	leaf content, %
September	62.1	23.1
October	35.0	15.1
November	28.0	4.0
December	17.7	0.7
January	14.3	0.5

We could mention that, in the middle of September, when the harvest of tubers started, the moisture and leaf contents of aboveground parts were 62.1% and 23.1%, respectively. Under the climatic conditions with temperatures below 0°C, snow and wind, in October-January, the defoliation and dehydration of the stems intensified, the leaf and capitulum content in *Helianthus tuberosus* aboveground biomass decreased from 15.1% to 0.5%, and the moisture content decreased from 35.0 % to 14.3%, respectively (Table 1). Other authors presented similar results, for example, in Poland, the moisture content of *Helianthus tuberosus* stalks in November-April decreased from 63.49% to 17.93% (Stolarski et al., 2014).

Biomass particle size and its distribution is an important parameter used for handling, storage, conversion, dust control systems and the combustion behaviour of biomass fuels. In the case of pure Jerusalem artichoke milled chaffs, we obtained the highest percentage of particles larger than 4 mm (10.2%), and the lowest values for the particles of 1 mm (6.7%). Moreover, the fractions 1-4 mm were relatively high, in the case of pure milled wheat straw (Table 2). This is probably an effect of the morphological nature of Jerusalem artichoke stalks, the high level of pith microstructures and the moisture content of the used materials, milled chaffs, influences the passage of particles through the sieve meshes.

The mixture of Jerusalem artichoke and wheat straw 50:50 and 70:30 resulted in a reduction in the proportion of longer particles and in an increase in the share of the smallest particles (below 2 mm).

It is known that low ash and moisture contents increase combustibility, high bulk density fuel is convenient to be transported, and high pelletizing percent makes higher yield of the pelletizing process. It was determined that Jerusalem artichoke milled chaffs were characterized by a low ash content (2.12%) and a high moisture content (14.3%), as compared with pure wheat straw. In the prepared mixtures of Jerusalem artichoke with wheat straw, there was a lower ash and higher moisture content than in pure wheat straw (Table 3). Mani et al. (2006), reported that moisture in the material, during the compaction, increases the bonding via Van der Wall's forces, thereby increasing the contact area of particles.

It was established that the bulk density and the gross calorific value of the Jerusalem artichoke milled chaffs constituted 231 kg/m^3 and 19.1 MJ/kg, these indices were the lowest in pure wheat straw milled chaffs: 163 kg/m^3 and 17.4 MJ/kg, respectively. We determined that the ratio of Jerusalem artichoke in mixture makes the bulk density of milled chaffs vary from 178 to 224 kg/m^3 and its gross calorific value - from 18.0 to 18.6 MJ/kg (Table 3).

Table 2. Particle size distribution of milled chaffs, %

Particle size	100% wheat straw	100% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke
<5mm	0.1	2.2	0.6	0.7	0.5
4-5mm	2.9	8.0	5.2	4.6	4.7
3-4mm	31.7	27.9	33.2	20.8	19.8
2-3mm	31.8	34.5	29.2	31.9	31.6
1-2mm	20.6	20.6	19.6	26.1	26.9
1mm	12.8	6.7	12.2	15.8	16.5

Table 3. Some physical and mechanical properties of biomass and pellets

Indices	100% wheat straw	100% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke	70% wheat straw +30% Jerusalem artichoke
moisture content of milled chaffs,%	11.6	14.3	11.8	13.8	13.0
ash content of milled chaffs,%	4.93	2.12	3.98	3.42	2.93
bulk density of milled chaffs, kg/m^3	163	231	178	197	224
gross calorific value of milled chaffs, MJ/kg	17.4	19.1	18.0	18.3	18.6
moisture content of pellets, %	13.4	12.3	14.1	13.9	14.1
ash content of pellets, %	4.87	2.10	3.94	3.45	2.87
specific density of pellets, kg/m^3	1007	880	1008	947	884
bulk density of pellets, kg/m^3	685	582	636	615	601
net calorific value of pellets, MJ/kg	15.6	17.7	16.2	16.7	17.2

In Poland, Kowalczyk-Jusko et al. (2012) stated that the calorific value of biomass of the examined varieties of Jerusalem artichoke varied within narrow limits 16.10-16.30 MJ/kg, while the ash content was 5.4-5.6%; Stolarski et al. (2014) mentioned that the ash content of *Helianthus tuberosus* stalks, from November to April, decreased from 5.26% to 3.02%, but the gross calorific value grew insignificantly from 18.45 to 18.59 MJ/kg dry matter.

In our study, has been established that after pressure agglomeration of the biomass, the average moisture in the pellets changed, thus it decreased by 2%, in pellets from pure Jerusalem artichoke biomass and increased in pellets from pure wheat straw, 30% and 70% mixtures, reaching the absolute values of 13.1-14.1%, the ash content of all prepared pellets did not change essentially as compared with milled chaffs. The specific density of pellets made from pure Jerusalem artichoke biomass and pure wheat straw was 880 kg/m^3 and 1007 kg/m^3, while the bulk density was 582 kg/m^3 and 685 kg/m^3, respectively. It was determined that the specific and bulk densities of pellets were the highest in the products manufactured from Jerusalem artichoke biomass mixture and they attained values of 884-1008 kg/m^3 and 601-636 kg/m^3, respectively. The net calorific value of pellets ranged from 15.6 MJ/kg to 17.7 MJ/kg and decreased with an increase in the proportion of wheat straw in the mixture (Table 3).

There are different results reported in research studies conducted by other authors. Kowalczyk-Jusko et al. (2012) stated that the calorific value of biomass of the examined varieties of Jerusalem artichoke varied within narrow limits 16.10-16.30 MJ/kg, while the ash content was 5.4-5.6%. Zapalowska & Bashutska (2017) noted that Jerusalem artichoke pellets were characterized by lower moisture content (6.81%), moderate ash content (2.04%), higher energy value (18.85 MJ/kg) and resistance to crushing (1024.57 Newton), as compared with pellets made from conifers and deciduous trees. Hăbăşescu (2011) stated that the pellets from sunflower stems had net calorific value of 14.8 MJ/kg and the ash content was 3.78%, the pellets from corn stems were characterized by 14.2 MJ/kg and 5.14%, respectively, and wheat straw - by 14.3 MJ/kg and 6.25%, respectively. According to Ivanova et al. (2015) the ash content in wheat straw pellets reached 6.33% and the calorific value 17.60 MJ/kg. Gageanu et al. (2017) stated that the pellets obtained from wheat straw had 8.0% moisture, 6.03% ash, 15.6 MJ/kg net calorific value and 382.3 kg/m^3 bulk density; from biomass mixture 40% wheat straws and 60% forestry residues - 8.1% moisture, 5.11% ash, 16.9 MJ/kg net calorific value and 456.3 kg/m^3 bulk density, but pellets from 50% wheat straw and 50% corn cobs - 8.37% moisture, 3.88 % ash, 16.2 MJ/kg net calorific value and 386.4 kg/m^3 bulk density.

The results obtained Miranda et al. (2015) for wheat straws pellets were: 9.4 % moisture, 9.10 % ash, 18.25 MJ/kg gross calorific value and 13.73 MJ/kg net calorific value and 620 kg/m^3 bulk density. Swietochowski et al. (2018) determined that the properties of wheat pellets and wood pellets used for tests differed significantly, wheat pellets contained 8.52% moisture, were characterized by 94.66% durability, 13.79 MJ/kg net calorific value, 587.5 kg/m^3 bulk density and 1151 kg/m^3 specific density, but wood pellets - 3.53% moisture, 98.31% durability, 16.79 MJ/kg net calorific value and 671.1 kg/m^3 bulk density and 1213 kg/m^3 specific density, respectively.

The bulk density of pellets made from miscanthus, wheat straw and corn stover was 580, 490 and 635 kg/m^3, respectively (Jackson, 2015). Lisowski et al. (2018) mentioned that the pellets made from hay, straw and their blend at the ratio of 50:50, have moisture content 5.56-5.87%, specific density of 974-1102 kg/m^3, while the bulk density values were 478-514 kg/m^3, the calorific value of pellets was 16.07-17.00 MJ/kg, the highest values were achieved by the pellets made from the blend of biomass.

Kowalczuk et al. (2012) mentioned the following calorific values: pellets from wheat straw 15.960 MJ/kg, rye straw 15.865 MJ/kg and pellets made from maize straw - 15.868 MJ/kg.

CONCLUSIONS

It was determined that the bulk density of the chaffs milled by a 6 mm sieve ranged from 163 to 231 kg/m^3, the ash content - from 2.12 to 4.93%, the gross calorific value - from 17.4 to 19.1 MJ/kg.

The biomass of the species *Helianthus tuberosus* was characterized by high gross calorific value and moderate ash content. The physical and mechanical properties of fuel pellets varied depending on the mixture ratio: the moisture content ranged from 13.8 to 14%, the bulk density - from 582 to 685 kg/m^3, the specific density - from 880 to 1008 kg/m^3 and the net calorific value - from 15.6 to 17.71 MJ/kg.

REFERENCES

Gageanu, I., Voicu, G., Vladut, V., Voicea, I. (2017). Experimental research on influence of recipes used on quality of biomass pellets. *Engineering for rural development*, 785–791.

Gudîma, A. (2018). Technology of obtaining of ENPlus pellets from agricultural waste in the conditions of the Republic of Moldova. PhD thesis, Chişinău. (155 p.) http://hdl.handle.net/123456789/2702

Gunnarsson, I.B., Svensson, S.E., Johansson, E., Karakashev, D., Angelidaki, I. (2014). Potential of Jerusalem artichoke (*Helianthus tuberosus* L.) as a biorefinery crop. *Industrial Crops and Products*, *56*, 231–240.

Heuze, V., Tran, G., Chapoutot, P., Bastianelli, D., Lebas, F. (2015). Jerusalem artichoke (*Helianthus tuberosus*). Feedipedia.

www.feedipedia.org/node/544

Hăbăşescu, I. (2011). Sursele energiei regenerabile şi echipamentul pentru producerea lor. *Akademos, 2*(21), 82–86.

Ivanova, T., Kavalek, M., Havrland, B., Kolarikova, M., Skopec, P. (2015). Comparison of technologic parameters of pellets and other solid fuels produced from various raw materials. *Agronomy Research*, *13*(2), 303–310.

Jackson, J.J. (2015). Optimal uses of biomass resources in distributed applications. Theses and Disser. Biosystems and Agricultural Engineering, 34.

Kaliyan, N., Vance Morey, R. (2009). Factors affecting strength and durability of densified biomass products. *Biomass and Bioenergy*, *33*(3), 337–359.

Kays, S.J., Nottingham, S.F. (2007). Biology and Chemistry of Jerusalem artichoke (*Helianthus tuberosus* L.) London: CRC Press, Taylor and Francis Group.

Kowalczuk, J., Zarajczyk, J., Sobczak, P., Mazur, J., Rudy, S., Cycan, D., Winiarczyk, M., Jadwisienczak, K., Zalewska, K. (2012). The usefulness of briquettes and pellets from selected plant materials for energy purposes. *Teka. Commission of Motorization and Energetics in Agriculture*, *12*(2), 311–313.

Kowalczyk-Jusko, A., Jozwiakowski, K., Gizinska, M., Zarajczyk, J. (2012). Jerusalem artichoke (*Helianthus tuberosus* L.) as renewable energy raw material. *Teka. Commission of motorization and energetics in agriculture*, *12*(2), 117–121.

Lisowski, A., Matkowski, P., Dąbrowska, M., Piatek, M.,Swietochowski, A., Klonowski, J., Mieszkalski, L., Reshetiuk, V. (2018). Particle size distribution and physicochemical properties of pellets made of straw, hay, and their blends. *Waste Biomass Valorization,* 1–13. doi.org/10.1007/s12649-018-0458-8

Mani, S., Tabil, L. G., Sokhansanj, S. (2006). Effects of compressive force, particle size and moisture content on mechanical properties of biomass pellets from grasses. *Biomass Bioenergy, 30*, 648–654.

Marian, G. (2016). Biocombustibilii solizi, producere şi proprietăţi. Chișinău.

Miranda, T., Montero, I., Sepulveda, F.J., Arranz, J.I., Rojas, C.V., Nogales, S. (2015). A review of pellets from different sources. *Materials*, *8.* 1413–1427.

Roman, G. V., Ion, V., Epure, L.I., Băşa, A.G. (2016). *Biomasa. Sursă alternativă de energie*. București RO: Ed. Universitară.

Smaga, M., Wielgosinski, G., Kochanski, A., Korczak, K. (2018). Biomass as a major component of pellets. *Acta Innovations*, *26*, 81–92.

Stasiak, M., Molenda, M., Banda, M., Wiacek, J., Parafiniuk, P., Gondek, E. (2017). Mechanical and combustion properties of sawdust-straw pellets blended in different proportions. *Fuel Processing Technology, 156*. 366–375.

Stolarski, M.J., Krzyzaniak, M., Snieg, M., Slominska, E., Piorkowski, M., Filipkowski, R. (2014). Thermophysical and chemical properties of perennial energy crops depending on harvest period. *International Agrophysic*, *28*, 201–211.

Swietochowski, A., Dabrowska, M., Lisowski, A. (2018). Friction properties of pellets made of wood and straw. *Engineering for Rural Development*, 1815–1820.

Ţîţei, V., 2015. Plant species for renewable energy production in the Republic of Moldova. *Scientific Papers. Series Agronomy*, *58*, 425–431.

Ţîţei, V., Coșman, S., 2016. Biochemical characteristics of the *Asteraceae* species silage and possible use as a feedstock for livestock and biogas production in Republic of Moldova. *Research Journal of Agricultural Science, 48*(2), 105–112.

Ţîţei, V., Teleuță, A., Muntean, A. (2013). The perspective of cultivation and utilization of the species *Silphium perfoliatum* L. and *Helianthus tuberosus* L. in Moldova. *Bulletin UASMV Cluj-Napoca*, S*eries Agriculture, 70*(1), 160–166.

Zapalowska, A., Bashutska, U. (2017). Qualitative analysis of pellets produced from Jerusalem artichoke (*Helianthus tuberosus* L.). *Proceedings of the Forestry Academy of Sciences of Ukraine, 15*, 124–128.

ISSN 2285 – 5785
ISSN-L 2285 – 5785