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INFLUENCE OF SEED AGE ON QUALITY, GERMINATION AND SEED HEALTH IN SOYBEANS

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Abstract

Due to its multiple uses: in human and animal nutrition but being also successfully used in rotations with the main cereal crops, soybean is one of the most valuable crops in the world. Seed germination can be influenced by a number of factors: genetic, technological, age of storage but also by the presence of pathogens inside or outside the seeds. Seeds infected with pathogens may have low storage capacity, low germination and weight, and low quality of oil and food. There are a number of pathogens that attack soybean seeds and can greatly affect the quality of the seeds. The most common pathogens are: Phomopsis sp., Cercospora kikuchii, Cercospora sojina, Colletotrichum sp., Pernospora manshurica but also a number of pathogens and secondary saprophytes, including Alternaria, Fusarium, Cladosporium and Penicillium (Telenko, 2018). The paper aimed to present the effects of seed age on soybean germination, fungal load and chemical composition. Seeds of Onix soybean variety, from 4 years (2017-2020) were stored in laboratory conditions and used in the experiment. Germination, chemical composition and also sanitary quality were significantly influenced by storage.

Key words*: fertilizers, yield, soybean, seed composition.*

INTRODUCTION

Soybeans are desired on the marketplace as a valuable source of protein used as feed, with food applications and oil which is broadly incorporated into food, feed, and some industrial applications (Clemente and Cahoon, 2009).

The importance of soybean crop is also related to the property of this plant to fix nitrogen in the atmosphere and to use it with high efficiency in the assimilation process, without requiring material efforts from growers. Biological fixation of atmospheric nitrogen has the great advantage that it is a natural and non-polluting process. The symbiosis between soybean plants and *Rhizobium japonicum* bacteria provides over 50% of the nitrogen needed for plant growth and development, after harvesting in the soil remaining between 80-140 kg N/ha (Haş, 2006). Legumes, for both grain and fiber, play an important role in human nutrition and animal feed due to their high protein intake.

Soybean is considered the "golden plant" of mankind and provides over 60% of the world's protein needs and due to its high oil content (over 20%) is considered an oleo-protein plant, providing over 25% of world oil needs. Soybeans are in the first place among oil plants and is ranked 4th in the world in the cultivated area.

Globally, many developing countries are struggling with an acute shortage of protein, the expansion of soybean crops being a future solution to eradicate malnutrition due to this deficit. Soybean seed germination and plant sensitivity in the early stages of vegetation are the main problems of this crop. To meet the requirements of quality for sowing, soybean seed must have a physical purity of over 98% and germination over 80%.

Minimum temperature for seed germination is 7°C, with an optimal temperature between 12-14°C. According to Arif et al. (2013) soybean seed must absorb 50 percent of its weight in moisture to germinate.

Seed germination is a highly complex physiological and biochemical phenomenon in which a number of biological factors acting on the embryo are involved.

Seed age and storage conditions are important factors affecting soybean germination, chemical

composition and also seeds sanitary quality with negative consequences on grain yield (Kandil et al., 2013; Milošević et al., 2004). Several diseases, including *Phomopsis* spp., *Cercospora kikuchii*, *Cercospora sojina*, *Colletotrichum* spp., *Pernospora manshurica*, *Alternaria*, *Fusarium*, *Cladosporium* and *Penicillium* are known to affect soybean crop.
The deterioration of soybeans during storage causes a decrease in the supply of high-quality seeds, being the main limiting factor of the production (Susilawati et al., 2019).

MATERIALS AND METHODS

Onix, an early soybean variety was used in the laboratory experiment that was conducted at the Research and Development Station for Agriculture Turda (RDSA Turda) and at the University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca (UASVM Cluj-Napoca). Germination and seed composition were measured both for fresh seeds and at 1-year-storage interval to 3 years. Soybean seeds were harvested in 4 different years: 2017, 2018, 2019 and 2020 and stored under ambient conditions until 2020. For standard germination test 100 seeds with 4 replications for 8 days were evaluated by using classical method between paper. Germination was performed by counting the germinated seeds and appreciating the germs.
The germinated seeds were counted twice. The first count was made to determine the germination energy, and the second at the end of the germination period, to determine the final germination. The counting was done at each separate repetition, starting with the first, by removing the normally germinated seeds with tweezers. Using the formula purposed by Krishnasamy and Seshu (1990) germination percentage was calculated (Germination (%) = number of seeds germinated/number of seeds tested x 100).
Soybean unground samples were scanned using NIR spectroscopy (Tango, Bruker Optik Gmbh, Ettingen, Germany) to quickly determine soybean seed composition: dry matter, protein content, oil content, stearic acid (18:0), oleic acid (18:1), linoleic acid (18:2), and linolenic acid (18:3).
To determine the health of the seeds, the presence of pathogens on the surface of the grains or under the tegument was also analysed. During the research, the seeds were monitored daily in order to observe and identify certain pathogens that may develop simultaneously with their germination. The identification of pathogens was performed by macroscopic examination of the symptoms on the seeds (exudate, mycelium, spores) and microscopically by obtaining preparations from the affected parts in order to be included in the genera from which they come.

Data analysis

The experimental data was prepared and processed in Polifact for ANOVA test and and the mean comparison was done by Excel (Microsoft, USA).

RESULTS AND DISCUSSIONS

The results from this study reveals that germination is very significant influenced by seed age (Table 1). Fresh seeds had the highest germination (93.5%) with small variation in the first and in the second year of storage. Onix soybean variety germination capacity decreased by 5.5%, 6% and 41.75% when seeds were stored for 1 year, 2 years and 3 years, respectively (Table 2).
Many researchers have also reported that germination, chemical composition and also sanitary quality are significantly influenced by storage (Carvalho et al., 2014; Conceição et al., 2016).

Table 1. ANOVA test for seed germination

Cause of variability	SP	DF	s^2	F
S (Storage)	4401.68	3	1467.2	151.673***
R (Replication)	3.6875	3	1.2291	
SxR	87.0625	9	9.6736	
Error S	87.0625	9	9.6736	
Total	4492.43	15		

Table 2. Effect of storage on seed germination

Storage period		Germination (%)	Difference (%)	Statistical signification
Fresh seeds	93.8	0	Mt.	
1 year	88.0	-5.5	0	
2 years	87.5	-6	0	
3 years	51.75	-41.75	000	

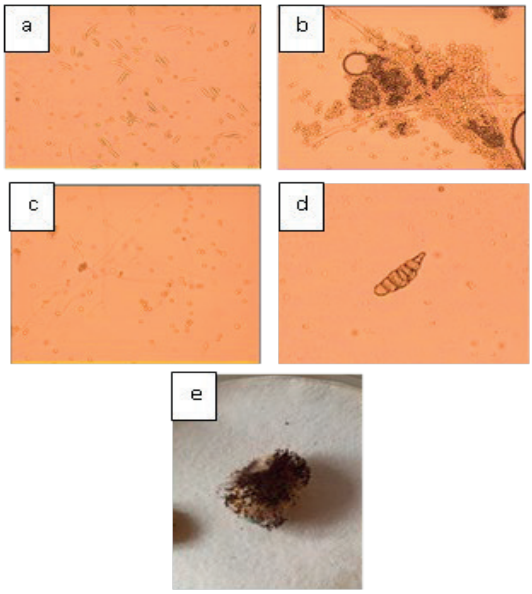

Figure 1. Microorganisms associated to the seeds with 3 years of storage
(a. *Fusarium* sp.; b. *Aspergillus*; c. *Penicillium* sp.; d. *Alternaria*; e. *Rhizopus*)

In our research, seed age affected the abundance of *Fusarium*, *Aspergillus*, *Penicillium*, *Alternaria* and *Rhizopus* species, all being identified when seeds were stored 3 years in the laboratory conditions (Figure 1).

If there was a small infection percentage associated to the seeds with 1 or 2 years of storage, after 3 years seed age caused the highest affectation leading to a significant decrease in germination. According to Lezcano (2015), also for other legumes, the infection of the seeds increased with the passing of the storage time.

Experimental data presented in Figure 2 highlights that seed size (TKW) was not affected by storage time. It varied between 128 g for the seeds harvested in 2018 and 183 g, this value being obtained for the 2019 harvest.

The germination capacity of soybeans is influenced not only by storage but also by sanitary quality of the seeds.

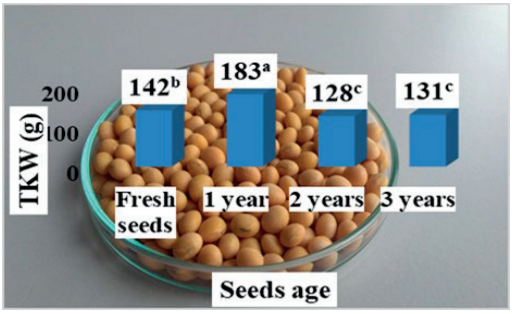

Figure 2. The influence of seed age on TKW

According to Duncan's test with respect to chemical composition of the seeds, after 3 years of storage maximum values for dry matter [%], fat content [%], stearic, oleic and linoleic acid content [%] were obtained. While no significant changes occurred for the fat content, protein content varied from 36.7% to 38.67% but it wasn't influenced by the seed age. Many researchers have reported that storage affected soybean seed composition (Table 3).

Table 4 reveals the estimates of the correlation coefficients evaluated for the studied parameters. Statistically, a positive close correlation between germination and moisture (r = 0.97) can be observed which comply with the reported literature. Also the significant correlations identified between seed size and protein content is very important to support breeding line selection in the soybean breading program. Similar results were obtained by Maestri et al (1998). A strong negative statistical proven correlation was established (r = -0.97) between the fat content and linolenic acid content. The well documented inverse relationship between protein and oil content is shown by the negative value of the correlation coefficients obtained for these quality parameters (Filho et al., 2001).

Table 3. The influence of seed age on quality parameters

No	Parameters	Seed age			
		Fresh seeds	1 year	2 years	3 years
1.	Dry matter [%]	93.30^{b}	93.5^{b}	93.5^{b}	93.8^{a}
2.	Protein [g/100 gDM]	37.23^{bc}	38.67^{a}	36.7^{c}	37.4^{b}
3.	Fat [g/100 g DM]	21.20^{b}	21.17^{b}	22.00^{a}	22.03^{a}
4.	Stearic acid [%]	5.00^{a}	5.30^{a}	5.33^{a}	5.07^{a}
5.	Oleic acid [%]	25.93^{a}	24.80^{b}	25.07^{b}	25.67^{a}
6.	Linoleic acid [%]	51.87^{b}	53.83^{a}	53.73^{ab}	54.10^{a}
7.	Linolenic acid [%]	8.60^{a}	8.27^{a}	7.40^{b}	7.30^{b}

Table 4. Correlation coefficients of studied parameters

	Stearic acid	Oleic acid	Linoleic acid	Linolenic acid	Fat	Moisture	Protein	TKW	Germination
Stearic acid	1	-0.23	0.30	-0.68	0.74	-0.08	-0.75	-0.64	0.02
Oleic acid	-0.23	1	-0.62	0.15	0.04	-0.01	-0.46	-0.57	-0.26
Linoleic acid	0.30	-0.62	1	-0.76	0.59	-0.77	0.23	0.06	-0.59
Linolenic acid	-0.68	0.15	-0.76	1	-0.97^{0}	0.78	0.44	0.60	0.69
Fat	0.74	0.04	0.59	-0.97^{0}	1	-0.70	-0.63	-0.77	-0.65
Moisture	-0.08	-0.01	-0.77	0.78	-0.70	1	-0.02	0.28	0.97*
Protein	-0.75	-0.46	0.23	0.44	-0.63	-0.02	1	0.94*	0.05
TKW	-0.64	-0.57	0.06	0.60	-0.77	0.28	0.94	1	0.38
Germination	0.02	-0.26	-0.59	0.69	-0.65	0.97*	0.05	0.38	1

CONCLUSIONS

The results clearly showed that seed storage for one or two year period does not reduce seed germination while a high decrease in final germination percentage was obtained from seeds stored for three years.

The seeds stored three years germinated but microorganisms such as *Fusarium*, *Aspergillus*, and *Rhizopus* also developed. On the seeds from 2018-2020, the incidence of microorganisms was lower, being present, in small percentages *Fusarium*, *Aspergillus* and *Penicillium*. As the storage time of soybean seeds increases, germination decreases, increasing the risk of installing certain pathogens.

The seed size was not influenced by the storage period, this quantitative character being dependent on the environmental conditions of the year in which the seeds were harvested. Similar results were obtained for the chemical composition of the seeds, the fat and protein content of the grains not being influenced by the age of the seeds.

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