Scientific Papers. Series A. Agronomy, Vol. LVII, 2014
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INTERRELATIONSHIPS BETWEEN GRAIN PROTEIN CONTENT AND INDICATORS OF NITROGEN STATUS OF WHEAT PLANT

Olivera NIKOLIC[1], Snezana ZIVANOVIC-KATIC[2], Milivoje MILOVANOVIC[2], Milanko PAVLOVIC[1]

[1]EDUCONS University, 21208 Sremska Kamenica, Vojvode Putnika 87, Serbia
[2]Small Grains Reasearch Center, 34000 Kragujevac, Save Kovacevica 31, Serbia

Corresponding author email: olivera.nikolic@educons.edu.rs

Abstract

Grain protein content is one of indicators of small grains quality. It is important trait, but negatively correlated with grain yield what represents obstacle in small grains breeding on quality and yield, simultaneously. The objective of investigation is to estimate interrelationships between grain protein content and some physiological indicators of nitrogen status in wheat. Tested indicators of nitrogen status in wheat plant are: nitrogen content in aboveground plant part at anthesis (Nanthesis), grain nitrogen content (Ngrain), nitrogen content at straw (Nstraw), nitrogen content at whole matured plant (Ntotal), nitrogen harvest index (NHI), nitrogen reutilization (NreU), nitrogen lost or gained (Npost anthesis) and physiological efficiency of nitrogen (PEN). According to obtained correlation coefficients, NHI and NreU expressed statistically high significant and positive influence on grain protein content. Influence of Npost anthesis was, in some investigation years, statistically high significant and positive, while influence of PEN on grain protein content was statistically high significant and strong, but negative. Influence of indicators of nitrogen accumulation efficiency, like: Nanthesis, Ngrain, Nstraw and Ntotal on grain protein content was insignificant during entire investigation. Such notice could mean higher importance of nitrogen utilization, as a part of process of nitrogen nutrition, in grain protein synthesis. Obtained results could achieve to better understanding of grain protein synthesis and overcoming some evident obstacles in wheat breeding programs.

Key words: *grain protein, nitrogen, wheat plant.*

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most widely grown crop in the word with its unique protein characteristics that serves as an important source of food and energy in human diet. Mature wheat grains contain 8–20% protein, while grain quality is a complex trait resulting from the interactions between numerous protein components (Daniel and Triboi, 2000). The protein content in the wheat grain is dependent on genotype but it is also clearly influenced by environmental variables such as nitrogen application, water access and temperature during growth especially through the grain filling period (Dupont and Altenbach 2003; Tea et al., 2004). The most effective environmental factor on wheat quality is N fertilization. At the same time, the degree of influence is affected by annual weather conditions and by residual soil N (López-Bellido et al., 2001). Therefore, proper management of N fertilizer is essential to ensure high quality wheat production. Nitrogen fertilization management (rate and timing) offers the opportunity for increasing wheat protein content and its quality besides high wheat production. However, excess applications of N are not economically efficient and can reduce protein content as well as create environmental problems (Tayebeh et al., 2011).
The nitrogen fertilizers using and efficiency of wheat plant nitrogen nutrition is topical subject nowadays, especially because of world energetic crises and fertilizers market, as well as requests for agriculture to be more effective and ecosystem protection appeal at the same time (Malesevic et al., 2010). The wheat plant nitrogen nutrition is very complex process and its explanation need following and appreciation a lot of physiological traits and reactions. Basically, the entire process depends on root system activity, mass and absorption capacity, kinds of fertilizers and time of their application, seasonal trends, physiological and genetic factors (Lopez – Bellido et al., 2005; Vuckovic et al., 2005; Bozhkov et al., 2007). The direct evaluation of root system activity, as trait

which works highly on plant absorption efficiency, is hardly deducible in the field conditions. So, there are some certain parameters, suitable to do it indirectly, like: nitrogen content in plant at anthesis or maturity as well as physiological efficiency of nitrogen and nitrogen harvest index as indicators of efficiency of plant nitrogen utilization. By wheat selection and breeding aspects, existance of dependence between these parameters and desirable traits is one of very important questions. Many authors (Anderson et al., 2004; Gallais and Coque 2005) defined that some of these parameters affect grain yield positively.

The aim of this study is to estimate correlation between parameters of nitrogen status and grain protein content in wheat, hoping obtained results could be helpful to overcome obstacles in wheat breeding on yield and quality simultaneously.

MATERIALS AND METHODS

The study was carried out on the property of the Small Grains Research Center in Kragujevac (186 m.a.s.l.), Serbia, during three consecutive seasons (2001/02, 2002/03 and 2003/04). The soil type was smonitza in degradation (Vertisol).

In all three years, the mean temperature was higher than the 30 yr average (Table 1).

There was considerable variability in rainfall amounts and distribution from year to year (Table 2).

The amount of rainfall was most suitable for plant growth in the third season. Rainfall (74.5 mm), received during the germination period (October - November) in the first season was less than in other two (97.00 mm and 111.8 mm) and long-term average (94.73 mm).

The experiment included 30 wheat cultivars and experimental lines, originating from the Serbia: Small Grains Research Center, Kragujevac and Institute of Field and Vegetable Crops, Novi Sad.

The basic processing and pre – sowing preparation of the soil was done using standard procedures.

The randomized complete block experimental design was used with five replicates in rows 1.5 m on, with spacing between rows of 0.20 m. Sowing (200 grains per row) was done by hand (one genotype per row), during the optimal planting period for central Serbian conditions, for winter wheat (29. 10. 2001, 15. 11. 2002 and 06.11. 2003).

Table 1. Average monthly temperatures during the three test growing season and long-term (30-yr) mean (LTM)

M	**Average monthly temperatures (^oC)**			
	2001/02	**2002/03**	**2003/04**	**LTM**
X	13.8	12.2	10.6	11.40
XI	4.6	9.7	8.9	5.90
XII	- 2.4	1.1	2.2	2.13
I	- 0.1	0.7	- 0.9	0.73
II	7.0	- 2.4	3.0	2.42
III	8.9	5.8	7.1	6.43
IV	10.8	10.8	12.8	11.22
V	18.4	19.9	14.5	16.24
VI	21.6	23.3	19.8	19.40
Season average				
	9.18	9.01	8.67	8.43

Table 2. Monthly amounts of rainfall during the three test growing season and long-term (30-yr) mean (LTM)

M	**Monthly amounts of rainfall (l)**			
	2001/02	**2002/03**	**2003/04**	**LTM**
X	10.4	65.5	83.2	47.53
XI	64.1	31.5	28.6	47.20
XII	27.6	39.4	37.2	44.33
I	17.2	59.0	86.4	36.70
II	20.1	19.7	59.5	35.77
III	26.0	2.8	21.3	41.57
IV	63.7	37.2	52.3	50.77
V	38.6	42.3	50.3	65.43
VI	57.2	47.7	61.4	81.27
Total				
	324.9	345.1	483.2	624.43

NPK fertilizer, formulated 8:24:16, was applied at the rate of 300 kg ha^{-1} before sowing each season. Eight grams row^{-1} of nitrogen (260 kg KAN ha^{-1}) was added at the tillering stage of development each season.

Plant samples of each genotype were taken at anthesis (10 plants per replication) and maturity (five plants).

The samples were air – dried and the above – ground weight of the plants at anthesis (DManthesis, g m^{-2}), grain yield (GY, g m^{-2}), weight of straw at maturity (DMstraw, g m^{-2}) and total above – ground biomass at maturity (BY, g m^{-2}) were measured.

All dry vegetative samples and grain were first ground and then plant N concentration was determined by the standard macro- Kjeldahl procedure. Nitrogen content (at anthesis, grain, straw and total at maturity) was calculated by multiplying the N concentration by dry weight (gN m^{-2}).

Moreover, the following parameters, related to dry matter and N accumulation and translocation within the wheat plant during grain filling, were calculated according to Arduini et al. (2006) and Masoni et al. (2007), as follows:

1. Nitrogen reutilization (NreU) = Nanthesis – Nstraw (g m^{-2})
2. Nitrogen lost (–) or gained (Npost - anthesis) = N content at maturity – N content at anthesis (g m^{-2})
3. Physiological efficiency of N (PEN) = GY/Ntotal (g_{grain}/gN) and
4. Grain protein content (GP) = %Ngrain x 5.7 (%)

The simple correlations coefficients between all pairs of variables were determined according to Chaudhary et al. (1999).

RESULTS AND DISCUSSIONS

Correlation coefficients between some traits pair are indicators of nature and significance of their relations and influences. So, correlation coefficients for GP and Npost anthesis (Npa) varied from negative and insignificant (genotypic and phenotypic - 0.08) to positive and high significant (genotypic 0.48** and phenotypic 0.53**) (Table 3).

Many authors (Egle et al., 2008) determined the term „nitrogen excess"conected with post – anthesis nitrogen accumulation. It can be suposed that nitrogen amount could be an important source of components for grain filling as well as for protein synthesis.

At the other side, many authors (Kade et al., 2005; Asseng and Milroy, 2006; Bahrani et al., 2011) found positive and strong correlation between grain protein content and reutilized nitrogen.

In contrast to these observation, relationship between GP and nitrogen reutilization efficiency was very unstabile in our investigation, varied from strong and negative (genotypic - 0.63** and phenotypic – 0.51**) to strong and positive (genotypic 0.29** and phenotypic 0.49**).

It could mean more important role of nitrogen accumulated after anthesis in provide plant with nitrogen substances for protein synthesis. It is obvious that the extreme variation of weather conditions during investigation, among years, influenced different relations between these traits.

Table 3. Correlation coefficients between grain protein content and investigated indicators of plant nitrogen status

Trait	Year	Correlation coefficients genotypic	Correlation coefficients phenotypiuc
		Grain protein (GP)	
Nanthesis	1	- 0.07	-0.06
	2	- 0.60**	-0.43**
	3	0.02	0.17
Ngrain	1	0.06	0.07
	2	0.08	0.11
	3	0.13	0.17
Nstraw	1	-0.13	-0.11
	2	0.13	0.16
	3	-0.17	-0.14
Ntotal	1	-0.03	-0.02
	2	0.11	0.16
	3	0.05	0.07
NHI	1	0.14	0.1*
	2	0.25*	0.39**
	3	0.38**	0.47**
NreU	1	-0.04	-0.01
	2	-0.63**	-0.51**
	3	0.29**	0.48**
Npost Anthesis	1	0.13	0.12
	2	0.49**	0.53**
	3	-0.08	-0.08
PEN	1	-0.73**	-0.54**
	2	-0.87**	-0.73**
	3	-0.65**	-0.54**

In the context of these relationships, results about negative and strong, statistically high significant correlation coefficients between reutilized and post anthesis gained nitrogen (Nicolic, 2009) could be interested.

Although, some authors registered connection between nitrogen harvest index (NHI) and grain protein content as insignificant, their correlation coefficients were positive and high significant in this investigation, no matter if you look at the genotypic (0.14, 0.25* and 0.38**) and phenotypic coefficients (0.1*,0.39** and 0.47**).

Obtained results are in agreement with studies Bahrani et al. (2011) Physiological efficiency of nitrogen (PEN), as another indicator of nitrogen utilization efficiency, was in negative and statistically high significant relation with grain protein content (GP) in this study. It can be explained by claim this parameter expresses ability of plant to use nitrogen for grain filling and yield formatting more than for protein synthesis.

At the other side, parameters belonging to the group of indicators of nitrogen accumulation efficiency, like: Nanthesis, Ngrain, Nstraw and Ntotal, expressed statistically insignificant correlation with GP, mainly.

The exception is nitrogen accumulated up to anthesis at above ground part of plant (Nanthesis), only in second studied year (genotypic – 0.60** and phenotypic – 0.43**). Low correlations of N assimilation prior to anthesis and grain protein concentration or grain protein yield were reported by Charmet et al. (2005).

Such results may lead to the conclusion that nitrogen utilization of earlier accumulated nitrogen reserves in plant, as part of nitrogen nutrition process, has greater role in plant physiological cycles.

CONCLUSIONS

Grain protein content in wheat is complex trait, influenced by many factors, processes and other indicators of plant nitrogen status. The effects of indicators of nitrogen status on grain protein content varied through investigation in dependence on weather and other conditions. The strongest and positive effect on grain protein content was achieved by nitrogen accumulated after anthesis, during reproductive period.

The significant and positive relationship was noticed between grain protein content and nitrogen harvest index, too. Physiological efficiency of nitrogen influenced grain protein content statistically high significant and strong, but negatively.

Generally, indicators of nitrogen utilization efficiency made more significant influence on grain protein content than indicators of nitrogen accumulation efficiency.

ACKNOWLEDGEMENTS

This investigation was supported by Ministry of Education, Science and Technology Development of Serbian Goverment, project TR 31054.

REFERENCES

Anderson A., Johansson E., Oscarson P., 2004. Post – anthesis nitrogen accumulation and distribution among grains in spring wheat spikes. J. Agric. Sci. 142, p. 525-533.

Arduini I., Masoni A., Ercoli L., Mariotti M., 2006. Grain yield, and dry matter and nitrogen accumulation and remobilization in durum wheat as affected by variety and seeding rate. Eur. J. Agron. 25, p. 309-318.

Asseng S., Milroy S.P., 2006. Simulation of environmental and genetic effects on grain protein concentration in wheat. Eur. J. Agron. 25, p. 119-128.

Bahrani A., Heidari H., Abad S., Aynehband A., 2011. Nitrogen remobilization in wheat as influenced by nitrogen application and post-anthesis water deficit during grain filling. African Journal of Biotechnology. 10, 10585-10594.

Bozhkov A.I., Kuznetsova Y.A., Menzyanova N.G., 2007. Interrelationship between the growth rate of wheat roots, their excretory activity and the number of border cells. Russian Journal of Plant Physiology, 54, p. 97-103.

Charmet G., Robert N., Branlard G., Linossier L., Martre P., Triboi E., 2005. Genetic analysis of dry matter and nitrogen accumulation and protein composition in wheat kernels. Theor. Appl. Gen., 111, p. 540-550.

Chaudhary B.D., Pannu R.K., Singh D.P., Singh V.P., 1999. Genetics of metric traits related with biomass partitioning in wheat under drought stress. Ann. Biol., 12, p. 361-367.

Daniel C., Triboi E., 2000. Effects of temperature and nitrogen nutrition on the grain composition of winter wheat: effects on gliadin content and composition. J Cereal Sci. 32, p. 45-56.

Dupont F.M., Altenbach S.B., 2003. Molecular and biochemical impacts of environmental factors on wheat grain development and protein synthesis. J Cereal Sci. 38, p. 133-146.

Egle K., Beschow H., Merbach W., 2008. Assessing post-anthesis nitrogen uptake, distribution and utilization in grain protein synthesis in barley (*Hordeum vulgare* L.) using 15N fertiliser and 15N proteinogenic and non-proteinogenic amino acids. Ann. Appl. Biol. 152, p. 209-221.

Gallais A., Coque M., 2005. Genetic variation and selection for nitrogen use efficiency in maize: a synthesis. Maydica, 50, p. 531-537.

Kade M., Barneix A.J., Olmos S., Dubcovsky J., 2005. Nitrogen uptake and remobilization in tetraploid 'Langdon' durum wheat and a recombinant substitution line with the high grain protein gene Gpc- B1. Plant Breed. 124:343-349.

López-Bellido L., López-Bellido R.J., Castill J.E., López-Bellido F.J., 2001. Effects of long-term tillage, crop rotatioand nitrogen fertilization on bread-making quality of hard red spring wheat. Field Crops Res. 72, p. 197-210.

Lopez-Bellido L., Lopez – Bellido R.J., Redondo R., 2005. Nitrogen efficiency in wheat under rained Mediterranean conditions as affected by split nitrogen application. Field Crop Res., 94, p. 86-97.

Malesevic M., Glamoclija Dj.,. Przulj N., Popovic V., Stankovic S., Zivanovic T., Tapanarova A., 2010. Production characteristics of different malting barley genotypes in intensive nitrogen fertilization. Genetika, 42, p. 323-330.

Masoni A., Ercoli L., Mariotti M. and Arduini I., 2007. Post-anthesis accumulation and remobilization of dry matter, nitrogen and phosphorus in durum wheat as affected by soil type. Eur. J. Agron. 26, p. 179-186.

Niucolic O., 2009. Genetical divergence of wheat genotypes for parameters of nitrogen nutrition efficiency. Doctoral thesis. Faculty of Agriculture Zemun, Belgrade, Serbia, p. 112.

Tayebeh A., Alemzadeh A., Kazemenini S.A., 2011. Wheat yield and grain protein response to nitrogen amount and timing. Australian Journal of Crop Science. 5, p. 330-336.

Tea I., Genter T., Naulet N., Boyer V., Lummerzheim M., Kleiber D., 2004. Effect of foliar sulfur and nitrogen fertilization on wheat storage protein composition and dough mixing properties. Cereal Chem. 81, p. 759-766.

Vuckovic S., Cupina B., Simic A., Prodanovic S., Zivanovic T., 2005. Effect of nitrogen fertilization and undersowing on yield and quality of Cynosuretum cristati - type meadows in hilly-mountainous grasslands in Serbia. Journal of Central European Agriculture, 6, p. 515-520.