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# Evaluation of Herbicides on the Content and Yield of Raw Protein in Wheat (*Triticum Aestivum* L.)

Margarita Nankova\*, Zornitsa Petrova, Genoveva Bankova - Atanassova, Emil Penchev, Georgi Sabev

Dobrudzha Agricultural Institute – General Toshevo \*Corresponding author: nankova\_margo@abv.bg

**Abstract** The investigation was carried out at Dobrudzha Agricultural Institute – General Toshevo during 2012 – 2014 in four-field crop rotation (wheat-maize-wheat-sunflower). Cultivars Aglika and Enola were sown at two dates – optimal (1<sup>st</sup> – 15<sup>th</sup> October) and late (after 20<sup>th</sup> November). The following set of herbicides was tested: Derby super VG – 3,3 g/da; Palace 75WG + oil – 25+100 g/ml/da; Granstar 75DF – 1,5 g/da; Lintur 70VG – 15 g/da; Husar Max OD - 100 ml/da; Secator OD - 10 ml/da and Mustang 306,25CK - 80 ml/da. The herbicides were applied at three dates: 1st date A - stage 29 according to Zadoks (Tillering stage); 2nd date B - stage 32 according to Zadoks (Stem elongation) and 3<sup>rd</sup> date C – stage 37 according to Zadoks (*Emergence of flag leave*). The experiment included two control variants K1 - weed-free control (manually weeded from the emergence of the first weeds till heading of wheat), and  $K_2$  (nil) – a control variant infested till harvesting of the crop. A high level of significance of the tested herbicides on the values of raw protein in grain and protein yield from the investigated cultivars was determined under the conditions of the experiment. The years of investigation had highest effect on the values of the tested characteristics. Best results were obtained in 2012. The two cultivars formed grain with the highest raw protein content in 2014. Regardless the sowing data the grain of cultivar Enola reached the maximum content of raw protein at the tillering stage of herbicides application, on the contrary of cultivar Aglika. The grain of cultivar Enola contained raw protein 9.33% more then cultivar Aglika average for the trail. The auxin type herbicide Lintur 70 VG cased the increasing the raw protein content in maximum degree in comparison the other herbicides. The exceeding to the weed-free control was 16.67% average for the years. At the late date of herbicide application, the amount of protein decreased in both cultivars. Averaged for the investigated period, the size of protein yield of cultivar Enola was with 18.7 % higher than the yield of cultivar Aglika. Cultivar Aglika has well expressed tolerance to the stages of herbicide application in comparison with cultivar Enola. Aglika's raw protein yield was higher at the stem elongation stage (32 Zadoks) of herbicides application. Cultivar Enola differ from cultivar Aglika with the greatest raw protein yield at the tillering stage (29 Zadoks) of herbicides application. In all three years of the investigation, highest yield of raw protein was obtained from the manually weed-free control (K<sub>1</sub>) and lowest – from the infested control (K<sub>2</sub>). Averaged for the tested variants of the trial, only after the application of the herbicides Derby super VG and Secator OD the raw protein yield remained at the level of the weed-free control (K1). There was a strong positive correlation of grain yield with raw protein yield. Averaged for the period of investigation, the sowing date of cultivar Aglika did not show significant effect on the values of the correlation coefficients. In cultivar Enola, the late sowing date lead to high increase of the correlation. This tendency was best expressed in harvest year 2014 (r=,875\*\*). The regression models obtained for the investigated years once again confirmed the high proportional correlation of grain yield with protein.

**Keywords:** wheat, herbicides, stages of application, protein yield, cultivars, dates of sowing

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#### 1. Introduction

The content of protein in wheat grain as influenced by different factors has been studied for more than 100 years [1]. Protein is the principal nitrogen-containing constituent in the wheat kernel. Therefore, soil and climatic factors that affect the N nutrition of the wheat plant will cause differences in wheat grain protein [2]. Herbicides can influence wheat protein levels by directly affecting the nitrogen uptake by the roots or the N metabolism and

protein synthesis within the plant [3,4,5]. Kumar reported that herbicides were able to gradually reduce the carbohydrate and protein content from lower to higher concentration of herbicides [6].

The complex methods for control of weeds in the cereal crops include various agronomy practices for restriction of their distribution and for ensuring better conditions for the growth and development of the cultural plants [7]. Under the conventional technologies for growing of common wheat, the control of the weed vegetation is a significant part of the entire agro technology of the crop.

Without efficient control of the weed vegetation, higher yields and quality production are unthinkable [8,9,10,11]. The development of weeds may be critical for the quality of the produce [12].

Delchev and Petrova [13] reported that there are insufficient investigations on the effect of the herbicides on grain quality in Bulgaria, and most of these investigations are focused on durum wheat [14,15,16]. Liu et al. concluded that the information worldwide was also scarce [17].

Mungova et al. found out considerable increase of the technological quality of wheat kernel when comparing a untreated check variant to variants with combined application of Akurat 60 VG at dose 1 g/dka, Impact 25 SC at dose 50 ml/dka and various fertilizers for foliar treatment – Amalgerol, Vertex H 34, Foliar extra, etc [18].

The aim of this investigation was to evaluate the effect from the use of a set of herbicides at different stages of the development of cultivars Aglika and Enola (*Triticum aestivum* L.) on the protein content and the raw protein yield after two dates of sowing.

## 2. Material and Methods

The investigation was carried out at Dobrudzha Agricultural Institute – General Toshevo during 2012 – 2014 in four-field crop rotation (wheat-maize-wheat-sunflower). Cultivars Aglika and Enola were sown in Haplic Chernozems at two dates – within the optimal

 $(1^{st}-15^{th}\ \text{October})$  and late (after  $20^{th}\ \text{October})$ . The mineral fertilization of wheat was done with conventional fertilizers at norm  $N_{12}P_{10}K_0$ .

It was created an artificial weeds background before wheat sowing. We used the most widespread weeds in the region: from annual broad-leaved - Veronica hederifolia (L.), Sinapis arvensis (L.), Galium tricorne Stok, Matricaria inodora (L.), Anthemis arvensis (L.), Consolida regalis Gray) and from perennial broad-leaved - Cirsium arvense (L.) and Convolvulus arvensis (L.)).

The tested herbicides were from the group of sulfonylureas characterized with two types of impact: auxin type and inhibitors of acetolactate synthase.

The auxin type herbicides (hormone-like preparations) inhibit the transportation of the auxin phyto hormone in plants. They case yellowing and wilting of leaves, epinastic deformations of petioles, stems, leaves and tumor growth on stems. The commercial preparations used were Lintur 70 VG and Mustang 306,25CK.

The inhibitors of acetolactate synthase cause the perishing of weeds within several weeks, and stunted growth, dwarfing, necrosis and chlorosis of the shoots are observed. These symptoms are caused by insufficient amino acids (leucine and isoleucine) leading to exhaustion and perishing of weeds. The following used commercial preparations belong to this group: Derby super VG, Granstar 75DF, Secator OD, Palace 75WG and Husar Max OD (Table 1).

Table 1. Characteristics of the tested herbicides

Name	Active substance	Applied norm	Action against				
Derby super VG	150,2 g/kg florasulam +300,5 g/kg aminopyralid-potassium	3,3 g/da	Broad-leaved weeds				
Palace 75 WG + oil	75 g/kg pyroxulam	25+100 g/ml/da	Broad-leaved + grass weeds				
Granstar 75 DF	750g/kg tribenuron-metil	1,5 g/da	Broad-leaved weeds				
Lintur 70 VG	4,1% triasulfuron +65,9% dicamba	15 g/da	Broad-leaved weeds				
Husar Max OD	8 g/l mesosulfuron +8 g/l iodosulfuron +23 g/l antidot	100 ml/da	Broad-leaved + grass weeds				
Secator OD	106 g/l amidosulfuron + 25 g/l iodosulfuron	10 ml/da	Broad-leaved weeds				
Mustang 306,25CK	6,25 g/l florasulam +300 g/l 2-4D - ester	80 ml/da	Broad-leaved weeds				

The herbicides were applied at three dates:

1<sup>st</sup> date A – stage 29 according to Zadoks – Tillering stage (main shoot and 9 or more tillers);

2<sup>nd</sup> date B – stage 32 according to Zadoks – Stem elongation (2<sup>nd</sup> note detectible)

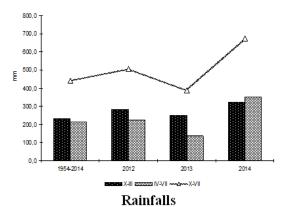
and 3<sup>rd</sup> date C – stage 37 according to Zadoks – Emergence of flag leaves (Flag leaf ligule/collar just visible) [19].

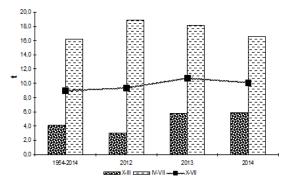
Two control variants were involved in the experiment:  $K_1$  – weed-free control (manual weeding from emergence

of the first weeds till heading of wheat) and  $K_2$  (*nil*) – infested control till harvesting of the crop.

Total nitrogen in grain was determined using Kjeldahl method and protein (N x 5,7) - BDS ISO 1871:2014.

The data were analyzed as completely randomized design using analysis of variance (ANOVA) test. All statistical analysis was carried out using the SPSS Inc. computer program, at 0.05 level of significance.





Air temperatures

Figure 1. Characteristics of the meteorological elements

meteorological elements, precipitation, were in all three years of the investigation considerably variable in comparison to the mean values of the long-term period (1954 - 2014) (Figure 1). The growth season of wheat during harvest year 2014 was characterized with the highest sum of rainfalls – 674.7 mm, which exceeded the mean long-term value with 52.9 % and was almost equal to the sum of autumn-and-winter rainfalls and the rainfalls during April – July. This was the most humid year apart from 2010 for the 60-year period of meteorological observations in the region.

Harvest year 2012 was also characterized with more rainfalls during the growth season as compared to the mean long-term values - with 14.7 %. The autumn-andwinter rainfalls were with 26.8 % higher than the April – July rainfalls. Harvest year 2013 was with the lowest sum of precipitation during the growth season of the crop -386.0 mm. The vegetation rainfalls during April – July were 136.2 mm, which resulted in drought at the critical stages of wheat development in May and June.

Averaged for the three years, higher mean temperatures during the growth season of wheat were measured in 2013 (10.7°C) and in 2014 (10.2°C) due to the higher mean monthly temperatures during the autumn-and-winter period. During the autumn-and-winter period of 2012, the mean air temperatures were lower than the mean longterm values. This year of the investigation, however, was characterized with higher mean monthly temperatures during the growth season of wheat from April to July in comparison to both 2013 and 2014, and to the mean values of the long-term period.

The applied statistical methods are: Disspertion analysis, Correlation analysis and Regression analysis. The data were analysed by SPSS 19.0 and MS Excel 2013 (graphics).

#### 3. Results

The analysis of the variances for protein content in grain of both cultivars revealed strongly expressed correlations of the values of the index with the independent and combined effect of the factors tested in the experiment (Table 2). Undoubtedly, most expressed was the effect of the meteorological conditions during the years of investigation. At the late date of sowing, the combination of the meteorological components had higher influence on the concentration of protein in the grain of both cultivars in comparison to the optimal date of sowing.

Table 2. Protein content - analysis of variances (n - 0.05)

Source	df	AGLIKA			ENOLA				
		Optimal date	Optimal date of sowing Late date of sowing C		Optimal date	of sowing	Late date of sowing		
		F	Sig.	F	Sig.	F	Sig.	F	Sig.
Years (1)	2	3802,7	,000	7307,6	,000	9912,5	,000	5961,6	,000
Date of herbicide application (2)	2	1528,1	,000	465,8	,000	262,9	,000	251,9	,000
Set of herbicides (3)	8	374,6	,000	206,7	,000	194,3	,000	51,8	,000
1 x 2	4	410,2	,000	20,1	,000	681,4	,000	46,4	,000
1 x 3	16	138,6	,000	66,3	,000	72,0	,000	59,0	,000
2 x 3	16	153,2	,000	42,9	,000	118,4	,000	24,4	,000
1 x 2 x 3	32	54,8	,000	70,2	,000	38,4	,000	38,3	,000

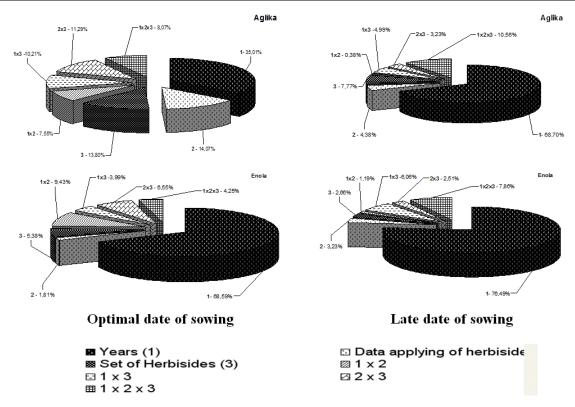


Figure 2. Strength of the factors' effect on grain protein content

The analysis demonstrated that the protein in the grain of cultivar Enola was affected to a higher degree by the meteorological factor than cultivar Aglika. The strength of this effect was 68.39 % at the optimal date of sowing, and 76.49 % at the late date of sowing (Figure 2). The combination of the meteorological components in cultivar Aglika when sown at the optimal date was also of primary importance, but the strength of their independent influence on the concentration of protein in grain was only 35.01 %, which resulted in higher effect of the other factors and their combined interaction. Thus the date of herbicide application was the factor second in strength of effect on the concentration of nitrogen in grain, followed by the set of the tested herbicides.

The set of tested herbicides had higher effect on the protein in grain regardless of the date of sowing in comparison to cultivar Enola. Although the effect was significant, in cultivar Enola the date of herbicide application had lower influence on the values of protein in comparison to cultivar Aglika. The strength of the combined interaction of the factors revealed considerable differentiation according to the cultivar and the date of its sowing. A permanent tendency was observed toward greater strength of the interaction between the three factors on the values of protein after the late date of sowing in comparison to the optimal date, this effect being better expressed in cultivar Aglika than in cultivar Enola.

The raw protein yield was significantly influenced by the level of the tested factors and their combined interactions (Table 3). The meteorological conditions of the year were the determining factor for this index. Their effect was better expressed on cultivar Aglika than on cultivar Enola. A well expressed differentiation was observed in the independent and combinative interaction between the factors in the respective cultivars according to the stage of application of the herbicides.

Table 3. Protein yield - analysis of variances  $(p - \theta, \theta 5)$ 

			AGLII	KA	ENOLA				
Source		Optimal date of sowing		Late date of sowing		Optimal date of sowing		Late date of sowing	
		F	Sig.	F	Sig.	F	Sig.	F	Sig.
Years (1)	2	15023,7	,000	10214,1	,000	9397,9	,000	5285,9	,000
Date of herbicide application (2)	2	914,4	,000	852,7	,000	2332,1	,000	1290,6	,000
Set of herbicides (3)	8	1096,1	,000	1484,6	,000	919,5	,000	351,7	,000
1 x 2	4	1026,1	,000	399,3	,000	356,5	,000	55,1	,000
1 x 3	16	226,3	,000	163,5	,000	120,8	,000	42,2	,000
2 x 3	16	445,1	,000	259,2	,000	173,8	,000	81,2	,000
1 x 2 x 3	32	134,2	,000	107,2	,000	51,4	,000	28,6	,000

In both cultivars the set of tested herbicides was the factor second in strength of effect (Figure 3). Its influence at the late date of sowing was stronger on cultivar Aglika (25.93 %) in comparison to cultivar Enola (14.75 %). At the optimal date of sowing the set of herbicides had higher

effect on the raw protein yield of cultivar Enola (19.06 %). The independent effect of the stage of application of the herbicides, although significant, was comparatively weaker on cultivar Aglika at both dates of sowing – from 3 to 4 %.

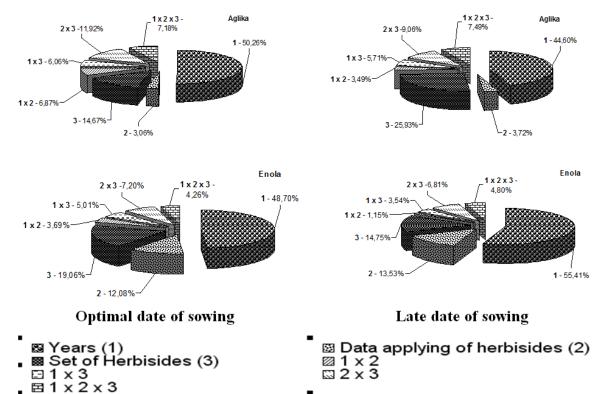


Figure 3. Strength of the factors' effect on grain protein yield

In cultivar Enola the strength of effect of this factor increased more than three times, with a tendency towards higher strength at the late date of sowing. Among the combined interactions, best expressed was the interaction date of application of herbicides x type of herbicides. This interaction had stronger influence on the values of the index in cultivar Aglika in comparison to Enola. The full combinative interaction between the factors also had higher effect on the raw protein yield from cultivar Aglika than Enola, without significant differences between the two dates of sowing.

#### 4. Discussion

The grain from the two cultivars in 2013 was with lower concentration of protein in comparison to the other years of the investigation (Table 4). Most probably the higher air temperatures in May and June, accompanied with strong deficiency of soil moisture, had negative influence on its concentration in kernel. At the same time the raw protein yield, averaged for the cultivars, were 65.33 kg/da and was less than the yield obtained in 2012 with only 3.32 g/da.

Table 4. Protein content and protein yield according to years of investigation

		AGI	IKA		ENOLA			
Years of investigation	Optimal dat	te of sowing	Late date of sowing		Optimal date of sowing		Late date of sowing	
	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da
2012	10,15 b	60,6 b	11,32 b	60,6 c	12,01 b	76,2 c	12,63 b	77,2 c
2013	8,87 a	63,8 c	9,09 a	59,5 b	9,10 a	69,0 b	9,71 a	69,0 b
2014	11,72 с	43,7 a	13,02 с	44,7 a	13,06 с	54,6 a	13,81 с	49,1 a

Different letter in the same row indicated significant difference (p<0.05) according to Waller-Duncan test.

Regardless of the fact that in 2014 the highest values of raw protein content were obtained, averaged for the cultivars (12.90 %). The mean yield of raw protein was lowest for the investigated period -48.03 kg/da. It was lower than the average yield for 2012-2014 (60.67 kg/da) with 20.83 %, and lower than the yield of 2012 (68.65 kg/da) with 30.04 %.

In all three years of investigation cultivar Enola exceeded cultivar Aglika with averagely 18 % by size of the raw protein yield. In both cultivars the late date of sowing lead to slight decrease of the values of the index in comparison to the optimal date of sowing.

As already commented above, the stage of application of the tested herbicides lead to significant variations of raw protein content in grain and the obtained yields from it. Grain quality, as a polygenically dependant trait, is a combination of biological, physical and chemical,

technological and utility characteristics, depending on a number of factors [16].

The use of herbicides during the growth season is one of those factors with considerable importance for the expression of the biological potential of plants. The results from our investigation showed that in cultivar Aglika, regardless of the date of sowing, a tendency was observed toward some increase of the raw protein content after the late application of herbicides at stage 37 (Zadoks) emergence of flag leaf (Table 5). In cultivar Enola the late application of herbicides at both sowing dates had the opposite effect – slight decrease of the raw protein content. Both cultivars, regardless of the stage of herbicide treatment, formed grain with higher concentration of protein after the late date of sowing as compared to the optimal one. In cultivar Aglika this mean increase was with 8.8 % for all variants of the experiment, and in cultivar Enola – with 5.6 %.

Table 5. Protein content and protein yield according to the date of herbicides application and stages of wheat development (Zadoks)

Stage of development		AGL	IKA		ENOLA			
	Optimal dat	te of sowing	Late date of sowing		Optimal date of sowing		Late date of sowing	
Stage of development	Protein	Protein	Protein	Protein	Protein	Protein	Protein	Protein
	content, %	yield, kg/da	content, %	yield, kg/da	content, %	yield, kg/da	content, %	yield, kg/da
Stage 29	9,2 a	54,1 a	10,6 a	53,4 a	11,6 b	71,0 c	12,5 c	72,1 c
Stage 32	10,6 b	59,1 c	11,4 b	57,9 b	11,6 b	68,3 b	11,9 b	65,3 b
Stage 37	11,0 с	54,9 b	11,5 b	53,5 a	11,0 a	60,4 a	11,7 a	57,8 a

Different letter in the same row indicated significant difference (p<0.05) according to Waller-Duncan test.

The row protein yield as a function of two values (concentration of protein in grain and grain yield) is subjected to differentiation according to the date of sowing, the stage of herbicide application and the type of cultivar. The obtained mean results for the wheat crop showed that the use of the herbicides at a late stage of its development (emergence of flag leaf) caused significant decrease of the yield from raw protein – with about 10 % in comparison to the application of herbicides at tillering stage (29 Zadoks).

The genotypic differentiation in the response of the cultivars was very strong. For cultivar Enola the late application of herbicides after tillering stage decreased the protein yield at both sowing dates of the cultivar. This decrease reached 20 % after late sowing. This is indicative

of high susceptibility with regard to grain yield since the decrease of protein in grain was not so strongly expressed. In relation to the obtained mean values of raw protein yield, cultivar Aglika was with well expressed tolerance to the stages of herbicide application. In practice, there was no decrease in the values of the index at the late date, and after their application at stage 32 (Zadoks) – stem elongation, a mean increase of raw protein yield with 9 % was determined in comparison to herbicide application at tillering. Regardless of this fact, averaged for the period of investigation, cultivar Enola exceeded cultivar Aglika with 18.6 % by raw protein yield. This tendency was present in all stages of application of the herbicides at both sowing dates of the cultivars, highest exceeding being

determined after their use at tillering stage (29 Zadoks) – averagely with 33.1%.

The herbicides tested in the trial, as well as the two check variants, differentiated the mean values of raw protein content in grain of the investigated cultivars from 10.7% in  $K_1$  to 12.4% in Lintur 70 VG (Table 6). The infested control (K<sub>2</sub>) and all tested herbicides increased the concentration of protein in grain in both cultivars after the two dates of their sowing. According to Peltzer and Bowran, some of the herbicides increased the grain protein of wheat in the absence of weeds provided that there was adequate available nitrogen [20]. These tended to belong to specific mode of action groups; acetolactate inhibitors (Group A), inhibitors of photosynthesis (Group C) and the phenoxyalkanoic acids of Group I. According to Singh and Verma (2015) the possible mechanism for this increase in crude protein content might be due to more uptake of N<sub>2</sub> in grain and the reduction of nitrate to ammonia by activities of complex enzyme resulting in production of more amino acids, which are the main constituent of protein [21].

Highest differentiation with regard to the values of raw protein yield was found between the two control variants. Averaged for the period, the yield from the infested control was the lowest obtained and conceded to the manually weeded variant with 18.5 kg/da.

Averaged for the dates of sowing, cultivar Aglika reached a mean maximum of raw protein yield after the use of Secator OD -60.0~kg/da, followed by  $K_1-59.3~kg/da$ . In cultivar Enola, the highest values of the index were obtained from the variants with Derby super VG -71.6~kg/da,  $K_1-71.3~kg/da$  and Secator OD -70.4~kg/da. The herbicides inhibitors of acetolactate synthase with action against broad-leaved and grass weeds (Palace 75 WG and Husar Max OD) lead to the lowest mean yields of raw protein obtained from both cultivars. They, however, significantly exceeded the infested control variant.

Table 6. Protein content and protein yield according to the set of herbicides

		AGL	IKA		ENOLA			
Variants of treatment with	Optimal date of sowing		Late date of sowing		Optimal date of sowing		Late date of sowing	
herbicides	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da	Protein content, %	Protein yield, kg/da
K <sub>1</sub> - check	9,4 a	58,4 e	11,0 c	60,2 j	10,7 a	70,9 f	11,5 a	71,6 f
K <sub>2</sub> - check	9,8 bc	44,0 a	10,9 b	40,5 a	11,3 cd	53,0 a	12,0 c	49,7 a
Derby super VG	9,8 c	57,5 d	11,1 cd	58,4 e	11,8 g	72,8 g	12,3 e	70,3 e
Palace 75 WG	9,7 b	52,5 b	11,0 c	54,6 c	11,1 b	63,4 b	12,2 e	65,3 cd
Granstar 75 DF	10,4 e	57,5 d	10,8 b	54,4 c	11,7 f	69,7 e	11,7 b	64,8 c
Lintur 70 VG	12,0 g	60,2 f	12,6 f	56,6 d	12,4 h	67,0 d	12,7 f	63,3 b
Husar Max OD	10,4 e	56,7 с	10,6 a	53,7 b	11,2 bc	65,1 c	12,0 c	64,7 c
Secator OD	10,6 f	60,6 j	11,2 e	59,3 f	11,3 d	70,9 f	12,2 de	69,8 e
Mustang 306,25CK	10,2 d	56,7 c	11,1 c	56,9 d	11,2 b	66,5 d	12,1 d	66,2 d

Different letter in the same row indicated significant difference (p<0.05) according to Waller-Duncan test.

The genotypic specificity in the response of the two cultivars, averaged for the tested herbicides and the control variants, was very well expressed (Figure 4). The mean raw protein yields were among those obtained from the two control variants, tending towards the yields

obtained from the manually free-weed check  $(K_1)$ . This fact is an indication that regardless of the various effects of the tested set of herbicides, they all have positive influence to different extent on the productivity of wheat with regard to protein.

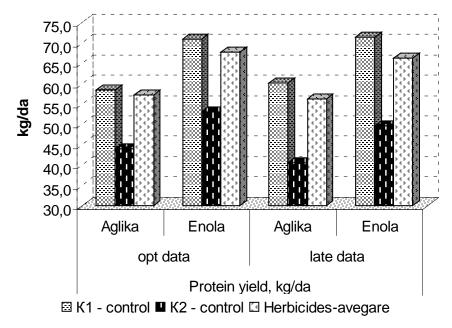


Figure 4. Raw protein yield averaged for the tested herbicides in comparison to the control variants during 2012-2014

The correlation of grain yield with raw protein yield was significant for both cultivars (Table 7). The values of

the correlation coefficients varied considerably depending on the year and the date of sowing. In cultivar Aglika, these values were lower in 2013 (a mean of ,512\*\*), and in cultivar Enola – in 2012 (a mean of ,772\*\*). The late date of sowing increased the correlation of grain yield with raw protein yield in both cultivars. The investigation showed that the mean values of this correlation

were ,648\*\* in cultivar Aglika, and ,812\*\* in cultivar Enola. In years with high sum of autumn-and-winter rainfalls and high sum of the rainfalls during the growth season (2014), the correlation of grain yield with raw protein yield was also stronger.

Table 7. Correlations between grain yields and raw protein yields (Pearson Correlation)

Years	Agi	lika	Enola		
Tears	Optimal date of sowing				
2012	,761(**)	,756(**)	,723(**)	,820(**)	
2013	,438(**)	,586(**)	,807(**)	,863(**)	
2014	,510(**)	,839(**)	,781(**)	,875(**)	

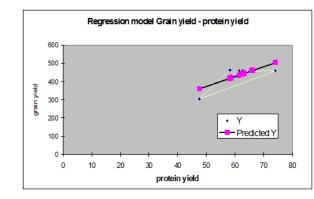
The significant correlations between grain yield and raw protein obtained for the investigated years were further specified by applying regression analysis. The regression models obtained for both dates of sowing of the cultivars during the investigated years once again confirmed the high proportional correlation of grain yield with protein (Table 8). A deviation from this tendency was found for the optimal date of sowing of cultivar Aglika in

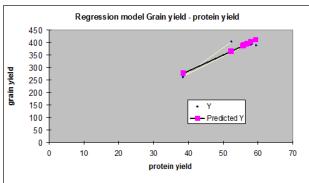
2013. This was most probably due to the critical soil drought during grain filling accompanied with extreme high air temperatures. The graphic representation of the models confirms the obtained results. In cultivar Enola these results were more stable for the two dates of sowing (Figure 5).

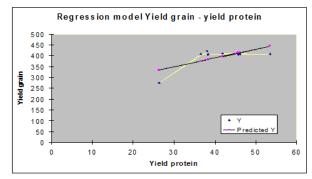
The high significance of the regression models allows predicting the raw protein production.

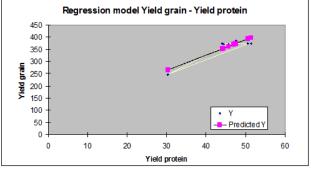
Table 8. Regression models between Grain Yield (Y) and Protein Yield (X)

Vocas of investigation	AGL	JKA	ENOLA		
Years of investigation	Optimal date of sowing	Late date of sowing	Optimal date of sowing	Late date of sowing	
2012	Y = 379.1 + 4.6 X	Y = 233.2 + 5.8 X	Y = 143.3 + 6.8 X	Y = 249.2 + 4.6 X	
2013	Y = 814.8 - 0.56 X	Y = 224.6 + 8.2 X	Y = 123.7 + 9.5 X	Y=173.7 + 7.9 X	
2014	Y= 224.5 + 4.14 X	Y=74.3+6.3 X	Y=98.5 + 5.6 X	Y=28.6 + 6.4 X	









## Optimal date of sowing

Late date of sowing

Figure 5. Regression models according to genotype and date of sowing (MS Excel 2013)

#### 5. Conclusions

A high level of statistical significance was found for the effect of the tested herbicides on the values of raw protein in grain and the grain yield from the studied cultivars under the conditions of the experiment. The years of

investigation had the highest influence on the values of the tested characteristics. Best results were obtained in 2012. The two cultivars formed grain with the highest raw protein content in 2014. Regardless the sowing data the grain of cultivar Enola reached the maximum content of raw protein at the tillering stage of herbicides application, on the contrary of cultivar Aglika. The grain of cultivar

Enola contained raw protein 9.33% more then cultivar Aglika average for the trail.

The auxin type herbicide Lintur 70 VG cased the increasing the raw protein content in maximum degree in comparison the other herbicides. The exceeding to the weed-free control was 16.67% average for the years.

At the late stage of herbicide treatment, the amount of protein decreased in both cultivars. Averaged for the investigated period, the size of protein yield from cultivar Enola was with 18.7 % higher than the yield from cultivar Aglika. Cultivar Aglika has well expressed tolerance to the stages of herbicide application in comparison with cultivar Enola. Aglika's raw protein yield was higher at the stem elongation stage (32 Zadoks) of herbicides applicaton. Cultivar Enola differ from cultivar Aglika with the greatest raw protein yield at the tillering stage (29 Zadoks) of herbicides application.

In all three years of study, highest raw protein yield was obtained from the manually weed-free control variant  $(K_1)$ , while the lowest was from the infested control variant  $(K_2)$ . Averaged for the tested variants of the trial, only after treatment with herbicides Derby super VG and Secator OD the raw protein yield remained at the level of the weed-free control  $(K_1)$ .

There was a strong positive correlation of grain yield with raw protein yield. Averaged for the period of investigation, the date of sowing of cultivar Aglika did not significantly affect the values of the correlation coefficients. In cultivar Enola, the late date of sowing caused sharp increase of the correlation. This tendency was best expressed in harvest year 2014 (r= ,875\*\*). The regression models obtained for the investigated years once again confirmed the high proportional correlation of grain yield with protein

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