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Original scientific paper

ASSESSMENT OF SOME PARAMETERS PRODUCTIVITY AND QUALITY OF POPULATIONS *Phleum pratense* L. GROWN IN CONDITIONS OF SERBIA

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Phleum pratense (L.) is the important forage grass species in Serbia. The study was conducted on 20 autochthonous population of *Phleum pratense* (L.), originating from Western Serbia. Plant height, protein content and crude cellulose content had a high direct impact on yield. Populations of PP16, PP20, PP2 and PP9 had the significantly higher yield of the raw matter compared to all the tested population and the highest values for the other evaluated parameters, also. The average yield of crude biomass was positive statistically highly significantly correlated with plant height (r=0.87**), and positively significantly correlated with a content of crude proteins and positive non significant correlated with crude cellulose (r=0.42 ^{ns}). Based on obtained values and by the appropriate choice of selection methods, we conclude that we have excellent genotypes, PP16, PP20, PP2 and PP9 for a successful selection process in order to obtain new high yielding varieties of *Phleum pratense*.

Key words: Timothy grass, population, yield components, chemical composition, correlation

INTRODUCTION

Phleum pratense (L.) a cool-season hexaploid perennial, known as Timothy grass is a Eurasian origin plant. It belongs to the family of the grass – *Poaceae*. Grass family has economic and ecologic importance. Grasses are represented in almost all ecosystems and are an important

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part of the natural food chain. Genus *Phleum* includes about 15 species, native to Europe, Asia, North Africa, North and South America where the *Phleum alpinum* species is represented. However, there is evidence that timothy is an allopolyploid (STEWART *et al.*, 2011). Timothy has excellent dry matter quality. In the last two decades, *Phleum pratense* (L.) has become one of the leading cultivated crops for production of animal feed, even in Serbia (VUČKOVIĆ, 1999, JANKOVIĆ *et al.*, 2017a; 2017b), because of favorable environmental condition, productivity and quality of biomass. For the successful cultivation it suits fresh and moist soil. It is widely used, can be met on mesophilic meadows of *Arrhenatheretalia phylum*, common on grasslands, roadsides, arable land edges, starting from the low-lying areas, to the high-Alps at a height of 2650 m above sea level (ALIBEGOVIĆ – GRBIĆ *et al.*, 2005). It increases its productivity by increasing of altitude. It does not show special sensitivity in relation with pH reaction of soil and it can be successfully grown on acid soils. *Phleum pratense* has the largest resistance to low temperatures compared to the other blades-grass. It has a high freezing resistance (LEMEŽINE *et al.* 2004). It is sensitive on strong and prolonged drought and high temperatures, because of its root system poor suction power. Timothy formed five times shorter roots in dry conditions compared to the watter conditions (JARGIELLO and HARROT, 1996).

Phleum pratense (L.) achieves excellent results in areas higher than 600 m above sea level, in Serbia. The aim of this study was to determine the productivity and variability of population germplasm of *Phleum pratense* (L.) for 20 autochthonous populations from Western Serbia area, in order to select genotypes for further crossbreeding and getting new high yielding varieties of *Phleum pretense L.*

MATERIALS AND METHODS

Plant materials

Genetic resources of 20 autochthonous populations of *Phleum pratense* (L.) were collected from Western Serbia area, namely the Kolubara district, during 2008. Most of the autochthonous populations *Phleum pratense* (L.) from the territory of Kolubara district are in the highland area and differ in length of the growing season. The traits were conducted from 2008 to 2010 years. At the start of the study, in 2008, 20 autochthonous population were collected from different landscapes of the Kolubara district territory landscapes (slopes, plateaus, waterlogged, shaded terrain, etc.). Geographic distribution and origin of 20 populations used in this work during the years of research is as follows: PP1, Bobova (453 msl, the edge of the forest); PP2, Gornje Leskovice (747 msl, the edge of the forest); PP3, Donje Leskovice (747 msl, the edge of the forest); PP4, Krčmar (650 msl, the edge of the forest); PP5, Bratačić (351 msl, field near the road); PP6, Gornji Lajkovac (525 msl, field near the road); PP7, Golubac (373 msl, field near the road); PP8, Lopatanj (392 msl, field near the road); PP9, Planinica (559 m.n.v., the edge of the forest); PP10, Pričević (298 msl, meadow near the road); PP11, Lelić (329 msl, field near the road); PP12, Struganik (575 msl, the edge of the forest); PP13, Sushica (535 msl, field near the road); PP14, Suvodanje (543 msl, meadow); PP15, Komirić (283 msl, meadow); PP16, Rožanj (973 msl, meadow near to forest); PP17, Gunjaci (395 msl, meadow); PP18, Skadar (530 msl, the edge of the forest); PP19, Popučke (161 msl, meadow); PP20, Carina (742 msl, meadow, near the forests). The size of the experimental unit was 10 m^2. Sowing seeds of collected autochthonous populations of *Phleum pratense* (L.) were carried out at the site Rađevo, Village (233 msl). The Municipality of Valjevo, in three replicate, on 13.10.2008. Type of land on which a trial was conducted is slightly acidic (pH 5.9%), leached soil, with medium fertility and humus

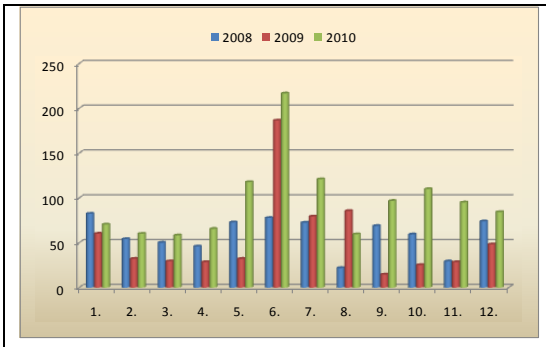
content of 3.59%. Ten plants from each replicate were taken for morphological analysis (height of the plants). After harvest has been made following analysis of the quality: the content of crude fiber and raw protein content (% of dry matter). The contents of crude fiber was done by the modified Hannerg-Stohman's method while the raw protein content (% of dry matter) was done according to the method of Kjeldahl. The yield of fresh matter per plant was also determined.

Statistical analysis

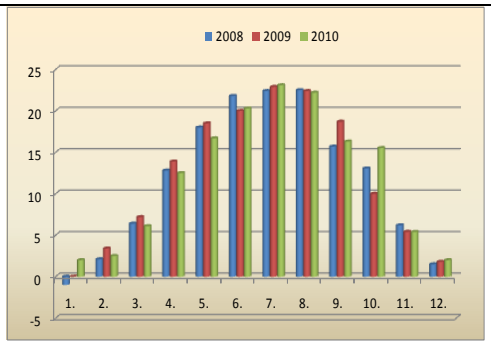
Data reported for the yield of Timothy grass was assessed by analyses of variance (ANOVA) and Fisher's LSD test was used for any significant differences at the P< 0.05 levels between the means. All the analyses were conducted using software package statistics 12.0 (StatSoft Inc. USA). Testing of the significance of the difference between the calculated average values of the investigated factors (year and genotype) were done by using a model of two-factor analysis of variance. The relative dependence was defined by correlation analysis method (Pearson's correlation coefficients). Obtained coefficients were tested by the t-test for the significance level of 0.05% and 0.01%. The coefficient of variation (CV) is the ratio between the standard deviation and the arithmetic mean and is expressed in percentages and calculated using the formula: CV=SD/$\bar{x}$ 100.

Meteorological conditions

Temperature and precipitation were recorded throughout the entire experiment by a meteorological station (Valjevo, Serbia) which is located next to the experimental field. In first year, the average temperature was 11.79ºC and total precipitation was 709.8 mm. During the second year average temperatures was 12.02ºC while the total precipitation was 651.1 mm. During the third year of the average temperature amounted to 12.05ºC while the total precipitation was 1156.2 mm (Figure 1).



a.



b.

Figure 1. Total precipitation (mm), a, and monthly air temperature average (°C), b, Valjevo

In agro-climatic conditions of the area, production is conducting mainly in natural water regime (POPOVIĆ *et al.* 2012; 2014; 2015; 2016a; STANISAVLJEVIĆ *et. al.* 2015; STRBANOVIĆ *et. al.* 2015; KRESOVIĆ *et al.* 2016). Best year of successful growth and development of plants in experiment, was the second production year.

RESULTS AND DISCUSSION

The description of agronomically important and useful characteristics is an important prerequisite for effective and efficient utilization of germplasm collections in breeding programs (MOHAMMADI and TALEBI, 2015). Mitigation of global climate change impact on the agricultural production is the major priorities in future research (PEJIĆ *et al.*, 2016). Timoty grass as a drought tolerant plant is interesting for growing in semi arid climate of Serbia region. Table 1. shows the descriptive statistics of the *Phleum pratense* (*L.*) population studied traits for years of experiment.

Analysis of variance showed significance for all the traits. Plant height, protein and crude cellulose content had a high direct impact on the yield of crude biomass per plant. The highest yield of the crude matter per plant was determined for populations PP16, PP20, PP2 and PP9 and statistically significantly higher in comparison to all other tested population. Populations of PP16, PP20, PP2 and PP9 also had the largest values for the other tested parameters (Table 1, Figure 2).

The most attention in the breeding is given to a yield of crude and dry biomass, because it is a characteristic that records large variations, which are depend upon a great number of genes, as well as on the agro-ecological conditions, the genotype and the genotype-environment interaction.

Table. 1. Productivity parameters for Phleum pratense (L.) population in Western Serbia

Population	2009	2010	$\bar{x}$	SD	CV	2009	2010	$\bar{x}$	SD	CV
	Plant height, cm					**Yield of crude biomass per plant**, g				
PP1	57.85	74.38	66.12	8.27	17.68	48.75	67.65	58.20	9.45	22.96
PP2	79.59	96.23	87.91	8.32	13.38	76.00	84.08	80.04	4.04	7.14
PP3	72.27	89.59	80.93	8.66	15.13	67.36	78.70	73.03	5.67	10.98
PP4	75.79	89.35	82.57	6.78	11.61	68.88	76.88	72.88	4.00	7.76
PP5	45.81	60.66	53.24	7.43	19.72	31.43	35.41	33.42	1.99	8.42
PP6	71.56	87.10	79.33	7.77	13.85	37.31	39.97	38.64	1.33	4.87
PP7	46.87	61.82	54.35	7.47	19.45	38.72	40.81	39.77	1.05	3.72
PP8	44.03	63.45	53.74	9.71	22.55	28.24	32.08	30.16	1.92	9.00
PP9	71.70	86.18	78.94	7.24	12.97	75.72	83.59	79.66	3.94	6.99
PP10	32.46	47.52	39.99	7.53	26.62	22.80	28.23	25.52	2.72	15.05
PP11	41.94	53.22	47.58	5.64	16.77	29.26	34.44	31.85	2.59	11.50
PP12	68.92	85.34	77.13	8.21	15.05	30.59	40.63	35.61	5.02	19.94
PP13	52.92	68.88	60.90	7.98	18.53	24.90	35.20	30.05	5.15	23.23
PP14	64.65	80.22	72.44	7.78	15.20	29.36	35.44	32.40	3.04	13.27
PP15	30.91	45.40	38.16	7.25	26.85	12.54	14.50	13.52	0.98	10.25
PP16	92.04	108.20	100.1	8.08	11.41	89.87	100.06	94.97	5.10	7.59
PP17	49.43	64.25	56.84	7.41	18.44	31.32	36.84	34.08	2.76	11.45
PP18	64.71	79.05	71.88	7.17	14.11	51.32	64.08	57.70	6.38	15.64
PP19	23.26	35.98	29.62	6.36	30.36	11.85	13.84	12.85	1.00	10.95
PP20	81.45	97.98	89.72	8.27	13.03	74.80	92.57	83.69	8.88	15.02
Average	58.41	73.74	66.08	7.67	16.41	44.05	51.75	47.90	3.85	11.37
CV	32.13	26.37	28.88	-	-	53.14	51.12	51.89	-	-

Table. 1cont. Productivity parameters for Phleum pratense (L.) population in Western Serbia

Population	2009	2010	$\bar{x}$	SD	CV	2009	2010	$\bar{x}$	SD	CV
	Crude cellulose , %					**Crude protein**, %				
PP1	26.25	25.59	25.92	0.47	1.80	14.00	14.04	14.02	0.03	0.20
PP2	26.60	25.48	26.04	0.79	3.04	14.30	14.22	14.26	0.06	0.40
PP3	26.00	26.02	26.01	0.01	0.04	14.10	14.24	14.17	0.10	0.70
PP4	26.05	26.07	26.06	0.01	0.05	14.10	14.30	14.20	0.14	0.99
PP5	26.00	25.80	25.90	0.14	0.55	13.50	14.30	13.90	0.57	4.07
PP6	26.02	26.00	26.01	0.01	0.04	14.05	14.09	14.07	0.03	0.20
PP7	25.80	26.24	26.02	0.31	1.19	14.00	14.02	14.01	0.01	0.10
PP8	25.00	26.24	25.62	0.88	3.42	13.50	14.10	13.80	0.42	3.07
PP9	26.01	26.09	26.05	0.06	0.22	14.10	14.46	14.28	0.25	1.78
PP10	25.20	25.80	25.50	0.42	1.66	13.60	13.70	13.65	0.07	0.52
PP11	25.10	25.88	25.49	0.55	2.16	13.80	14.04	13.92	0.17	1.22
PP12	25.60	26.24	25.92	0.45	1.75	14.10	14.26	14.18	0.11	0.80
PP13	24.90	26.98	25.94	1.47	5.67	14.10	14.32	14.21	0.16	1.10
PP14	25.96	26.00	25.98	0.03	0.11	14.15	14.33	14.24	0.13	0.89
PP15	24.30	24.66	24.48	0.25	1.04	13.20	14.20	13.70	0.71	5.16
PP16	26.02	26.12	26.07	0.07	0.27	14.20	14.60	14.40	0.28	1.96
PP17	25.79	26.01	25.90	0.16	0.60	13.50	14.42	13.96	0.65	4.66
PP18	26.00	25.88	25.94	0.08	0.33	14.01	14.13	14.07	0.08	0.60
PP19	25.03	25.09	25.06	0.04	0.16	13.40	13.64	13.52	0.17	1.25
PP20	26.05	26.07	26.06	0.01	0.05	14.20	14.52	14.36	0.23	1.58
Average	25.68	25.91	25.79	0.16	0.77	13.88	14.20	14.05	0.21	0.21
CV	2.26	1.81	1.71	-	-	2.09	1.70	1.72	-	-

Tested parameters	**LSD**	**Plant height**	**Yield of crude biomass**	**Crude protein**	**Crude cellulose**
Genotype	0.5	3.808	1.208	0.235	0.359
	0.1	5.047	1.602	0.312	0.476
Year	0.5	1.207	0.382	0.075	0.113
	0.1	1.601	0.506	0.098	0.150
Genotype x Year	0.5	5.401	1.709	0.333	0.507
	0.1	7.158	2.265	0.441	0.673

The objective of each breeding process is the creation of genotype with increased yield of crude and dry matter with the improvement of quality. Variability of the yield of crude biomass per plant, between the years, was in interval 3.72%<CV<23.23%.

Variability of all genotypes in yield of crude biomass per plant, during the year, was extremely high (51.12% <CV<53.14%), Table 1, Figure 2b. Variability of plant height, between the years, recorded large fluctuations. The coefficient of variation was ranged 11.41% < CV < 30.36%, Table 1, Figure 2a. The variability of plant height for examined genotypes during the year was high, which indicates the heterogeneity of the samples (26.37% < CV < 32.13%).

Variability of crude celulose, between the years, was extremely small and ranged between 0.04%<CV<5.67%, Table 1, Figure 3a. The variability of crude celulose per plant for examined genotypes during the year was small, which indicates the homogeneity of samples (1.81%<CV<2.26%). Variability of crude proteins content, over the years, was small and ranged 12.10%<CV <5.16%. The variability of crude proteins per plant for examined genotypes, during the year, was small, which indicates the homogeneity of samples (1.70 % < CV<2.09 %), Table 1, Figure 3b.

The highest yield of dry biomass was detected in the population PP16 – 94.97 g per plant, and the lowest in PP19 – 12.85 g per plant (Table 1).

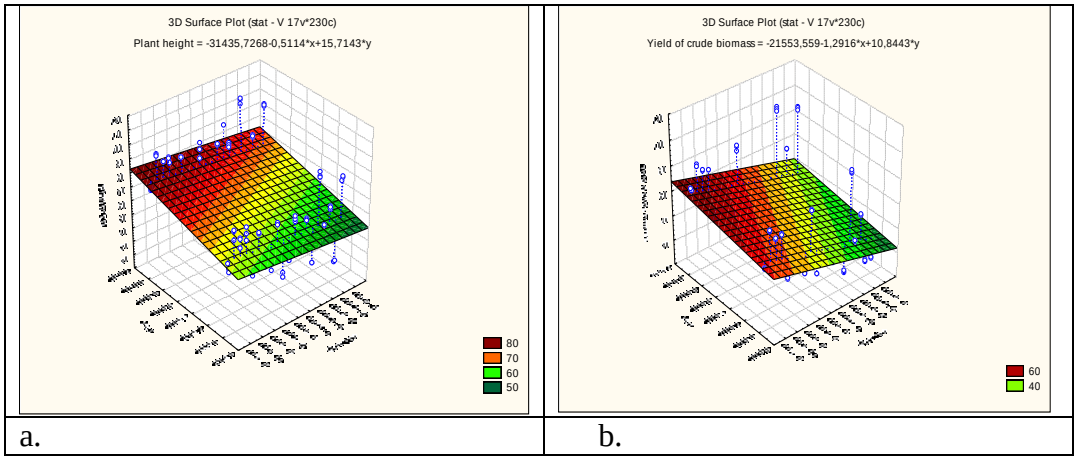


Figure 2. Plant height, a., and yield of crude biomass, b., for *Phleum pratense* (L.) population

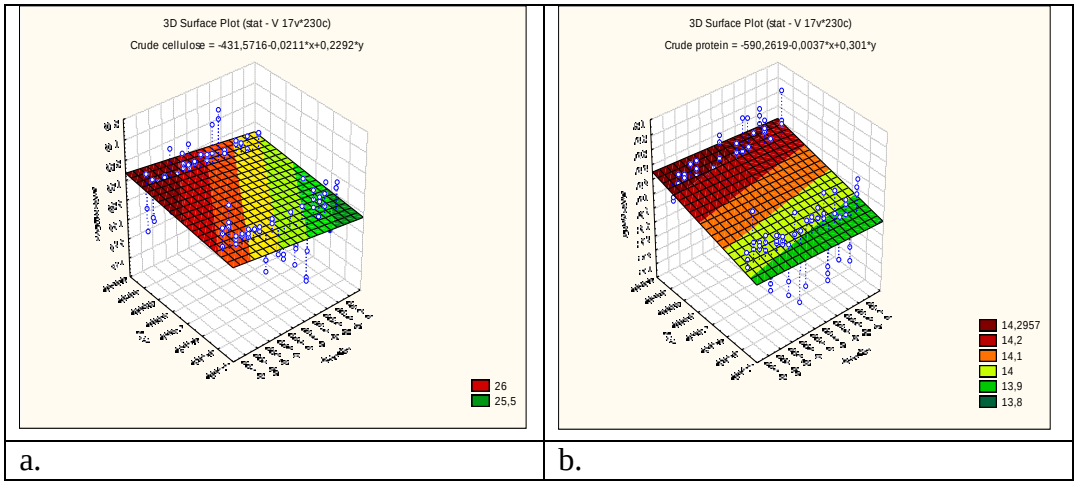


Figure 3. Crude cellulose, a., and crude protein, b., for *Phleum pratense* (L.) population

Plant height is an important parameter for the breeding process of *Phleum pratense* (L.). Competitive ability depends on the height of the plants, which is very important, considering that

the *Phleum pratense* (L.), and other perennial grasses, mainly used in mixtures with other types of grasses and legumes, and very rare as a clean crop.

The maximum height was observed in the population PP16 (100.12 cm), then in PP20 (83.72 cm) and PP2 (87.91 cm). The lowest population was PP19 (29.62 cm). The largest intra-population variability was obtained for the dry matter yield per plant (CV=51.89%), while the smallest variable feature was crude fiber content (CV = 1.71%); (Table 1).

Correlations of studied features

The results of the relative dependence of examined indicators of *Phleum pratense* (L.) population were presented by Pearson's correlation coefficient and shown in Table 2. It can be concluded that the yield of raw biomass has a statistically significant correlation with all tested parameters. The yield of crude biomass has a positive and highly statistically significant correlation with plant height (r=0.78**) and are positive significantly correlated with crude proteins, P <0.05 (r=0.53*) and positive nonsignificant correlated with crude cellulose (r=0.42 ^{ns}). The height of plants was in a positive statistically highly significant correlation with the crude proteins and the crude cellulose content (r=0.72** and r=0.57*), Table 2. According to our research there are also research by Jancic et al. (2008). In silage made of legumes JANČÍK *et al.* (2008) reported that in the present study CP was highly correlated (P < 0.05) with ADF, NDF with cellulose.

Table 2. The correlation of yield and yield and quality components

Parameters	Yield of crude biomass	Plant height	Crude protein	Crude cellulose
Yield of crude biomass	1.00	0.78**	0.53*	0.42^{ns}
Plant height	0.78**	1.00	0.72**	0.57*
Crude protein	0.53**	0.72**	1.00	0.51*
Crude cellulose	0.42 ^{ns}	0.57*	0.51*	1.00

ns- non significant* and ** significant at level of p<0.05 and p< 0.01

Based on the obtained value and also by the appropriate choice of selection methods, we can see that excellent genotypes (PP16, PP20, PP2 and PP9) are available to us for a successful selection process in order to obtain new varieties of *Phleum pratense* (L.). The efficiency of selection and breeding depends on the present genetic variation in the initial breeding material. The efficiency of selection and breeding depends on present genetic variation in the initial breeding material. The first and essential step in the selection is the examination of starting material in order to determine its variability and the basic characteristic. The main objectives in the breeding grass are directed on obtaining varieties of high productivity and good quality, long-lived and tolerant to limiting environmental conditions. Depending on the specific intended purpose of future varieties are chosen selection criteria and characteristic on which selection is done. Breeding of perennial grasses, in our conditions, is primarily directed towards increasing yields or maintaining stable yields with quality improvement. Selection in a given direction is done for creating the varieties with different maturing and which are intended for different forms

of exploitation (TOMIĆ and SOKOLOVIĆ, 2007; VUČKOVIĆ *et al*., 2007; BABIĆ *et al.* 2015; POPOVIĆ *et al.*, 2016b; VASILEVA and VASILEV, 2012, 2017). Timothy grass belongs to the later-maturing group of grasses (VUČKOVIĆ, 1999) and also in a high grasses group and it keeps good quality for a long period. That is why it is recommended to be used in mixtures.

CONCLUSIONS

Based on the obtained results it can be concluded that the plant height, protein content and crude fiber content of Timothy grass had a high direct impact on yield. Populations of PP16, PP20, PP2 and PP9 had the significantly higher yield of the raw matter compared to all the tested population and the highest values for the other evaluated parameters, also. The average yield of raw biomass was positive and statistically highly significantly correlated with plant height (r=0.87**), and positively significantly correlated with a content of crude proteins P <0.05 (r=0.53*) and positive non significant correlated with crude cellulose ($r=0.42^{ns}$). Based on obtained values and by the appropriate choice of selection methods, we conclude that we have excellent genotypes, PP16, PP20, PP2 and PP9 for a successful selection process in order to obtain new high yielding varieties of *Phleum pratense*.

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OCENA NEKIH PARAMETARA PRODUKTIVNOSTI I KVALITETA POPULACIJA *Phleum pratense* L. PROIZVEDENE U USLOVIMA SRBIJE

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SAŽETAK

Phleum pratense (L.) je najvažnija vrsta krmnih trava u Srbiji. Istraživanje je sprovedeno na 20 autohtonih populacija *Phleum pratense* (L.), poreklom iz zapadne Srbije. Visina biljke, sadržaj belančevina i sadržaj sirovih vlakana imali su visoko značajan uticaj na prinos. Populacije PP16, PP20, PP2 i PP9 imale su znatno veći prinos sirove biomase u odnosu na sve testirane populacije i najviše vrednosti za ostale ispitivane parametre. Prosečni prinos sirove biomase bio je u pozitivnoj statistički značajnoj korelaciji sa visinom biljaka (r =0,87**), i u pozitivnoj korelaciji sa sadržajem sirovih proteina i sirove celuloze. Na osnovu dobijenih vrednosti, zaključujemo da imamo izvrsne genotipove, PP16, PP20, PP2 i PP9, za uspešan postupak selekcije, odgovarajućim odabirom metoda selekcije, kako bi se dobile nove visoke sorte *Phleum pratense.*

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