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Original scientific paper

MORPHOLOGICAL CHARACTERISTICS OF THE INTERSPECIES HYBRID BETWEEN SORGHUM AND SUDAN GRASS UNDER INTENSIVE NITROGEN NUTRITION

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The object of this study was a two-year trial (2009 and 2010) regarding variability of morphological characteristics of species belonging to *Sorghum* genus, more specifically interspecies hybrid between sorghum and Sudan grass *Siloking* as affected by different applications of nitrogen. The following morphological characteristics were analysed: plant height, number of leaves, leaf ratio, stem ratio, and number of shoots. Samples were taken from the first cut when the effect of the applied nitrogen doses was the strongest. The results showed that increasing nitrogen quantities significantly affected the tested morphological characteristics, especially the intensity of tillering (increased number of secondary stems), number of the formed leaves, and ratio of leaf weight in the total above-ground biomass. The effect of applied nitrogen depended on the weather conditions, i.e. distribution of precipitation, so that plants reached maximum height when 105 kg N ha^{-1} was applied in the dry year and 180 kg N ha^{-1} in the wet year.

Key words: *Sorghum*, Interspecies hybrid, Genotype, Nitrogen nutrition

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INTRODUCTION

Introduction of a larger number of forage crops into production in the system of green forage conveyer solves the problem of quality voluminous feed lack, and here forage sorghum and Sudan grass play an important role GLAMOČLIJA (2011). Bearing in mind that contemporary breeding programs involving forage sorghum and Sudan grass are aimed at developing F_1 hybrids with profound potential for high, quality and stable yields of fresh biomass, suitable for use as fresh biomass, hay or silage PATAKI (2006)**,** in order to achieve this goal it is extremely important to test the combining abilities of the potential parent components MIHAJLOVIĆ *et al.* (2007). Nowadays, a larger number of hybrids are grown worldwide since they are tolerant to drought in all phenophases. Diversity of interspecies hybrids provides many products used as food and feed, as well as in processing industry MCLAREN (2003). Hybrids between sorghum (*Sorghum bicolor* L.) and Sudan grass (*Sorghum sudanense* L.) are especially important for the production of voluminous feed. These have several advantages over original genetic material, such as higher genetic yield potential, higher leaf weight, more intensive tillering, and faster regeneration after cutting ZEBARINI and THOMAS (2003).

General precondition for high yields implies that all needs of plants are met during entire growing period in the best possible way. Apart from ample food and water supply, this also implies good uptake of nutrients, creation of necessary compounds in the most favourable ratios and quantities, and their use by plants for growth. Cultivated plant species from the family of grasses significantly react to increased nitrogen (N) nutrition, especially when grown on soils of low fertility (IKANOVIĆ *et al.,* 2010). Simultaneously, use of larger nitrogen quantities significantly increases the cost of the production, so that the question of rational application of mineral nutrients arises (BOOKER *et al. 2007*). This issue is also important regarding environmental protection, since remaining quantities of nitrogen are a potential source of contamination for the entire ecosystem, according to MARSALIS *et al.* (2010) and GLAMOČLIJA *et al.* (2011). As far as humans are concerned, most adverse effects of excessive nitrogen application in plant production include accumulation of harmful and toxic substances in plant and animal products which pose a threat to food safety ERISMAN *et al. (*2007). It is important to state that interspecies hybrids have a strong reaction to increased nitrogen nutrition, they use it better from the soil and their above-ground biomass has lower amounts of substances harmful to domestic animals (ERIĆ, 2001; IKANOVIĆ, 2011).

MATERIALS AND METHODS

Two-factor field trial was set up at experimental field Radmilovac on brown forest soil (i.e. eutric cambisol according to FAO soil classification) in 2009 and 2010. The trial was designed as random block system in ten replications on basic plot size 10 m^2 (5 m x 2 m). The object of the study was the interspecies hybrid between sorghum and Sudan grass *Siloking* developed at Institute of Field and Vegetable Crops in Novi Sad. There were three doses of increased nitrogen nutrition applied into soil during seedbed preparation: 105 kg ha^{-1} (N_2), 150 kg ha^{-1} (N_3), 180 kg ha^{-1} (N_4) and control (N_1) – natural soil fertility 60 kg ha^{-1}. Standard sorghum cultivation practices were applied.

Previous crop was winter small grains after which shallow ploughing of post-harvest plant residues was performed in July and deep ploughing in October. Seeds were sown in the third ten-day period of April in both trial years. Plants were cut when panicle started appearing (first cut in

the second ten-day period of July). For the analysis of the morphological characteristics (plant height, number of leaves, leaf ratio, stem ratio, number of shoots) samples of above-ground biomass were taken from the first cut when the effects of the applied mineral fertilizer were maximum. The aim of this study was to test effect of increased nitrogen quantity on the analysed morphological characteristics of the interspecies hybrid *Siloking*.

Weather conditions were unfavourable, especially water regime in the first year since April and May only saw 40 mm precipitation which slowed plant emergence (Table 1). According to POPOVIĆ *et al.* (2011, 2012a, 2012b, 2013) dry periods slowed plant emergence and growth.

Table 1. Precipitation sums (mm) and daily mean temperatures (^{0}C) in the growing period (source: Hydro-meteorological Station in Belgrade)

Year	Parameter	Month						Average
		IV	V	VI	VII	VIII	IX	Sums
2009	Temperature	16	20	21	24	24	20	21
	Precipitation	6	34	153	79	45	45	362
2010	Temperature	14	18	21	24	24	18	20
	Precipitation	41	85	180	41	54	51	452
Ten years	Temperature	15	26	23	25	25	18	21
Average	Precipitation	15	58	102	53	54	49	331

Abundant precipitation in June and later during summer proved to be very beneficial for plant growth in the first year. The second year was more favourable regarding quantity and distribution of precipitation. Temperature conditions in both years were favourable for plant development.

Experimental data were statistically analysed using STATISTICA 8 for Windows (StatSoft). Each of the indicators was processed by descriptive statistics (for annual indicators). In order to reach objective conclusions on the effect of the observed factors on the analysed characteristics of the interspecies hybrid, and possible application of parametric tests (analysis of variance and LSD test), homogeneity of variances was tested by Hartley`s, Cochran`s, Bartlet`s and Levene′s tests. Agreement between empirical distribution of data and model of normal distribution was put to Kolmogor`s -Smirn`s test. Statistical significance of differences between analysed N quantities for each tested morphological characteristic was determined via analysis of variance (ANOVA), Kruskal-Wallis test and median test. Individual comparisons were performed by test of least significant difference and Mann-Whitney test, at the 5% and 1% levels of risk. Relative dependence of characteristics was measured by Pearson`s correlation coefficient, and tested at 5% and 1% levels of significance MALETIĆ (2005). Considering the fact that morphological characteristics of Siloking were analysed through several characteristics, I-distance was used to determine the synthetic rank to show which nitrogen dose gave the best effect of all tested characteristics.

$$D_r^- \sum_{i=1}^{k} \frac{x_{ir} - x_i^-}{\sigma} \prod_{j=1}^{i-1} \left(1 - r_{ij}\right)$$

RESULTS AND DISCUSSION

The analysed morphological characteristics showed high dependence on the applied dosage of nitrogen and weather conditions, i.e. precipitation distribution. In 2009 precipitation distribution was unfavourable and plants reached maximum height when 105 kg N ha^{-1} was applied. Maximum of other observed morphological characteristics (number of leaves, leaf ratio in total biomass and number of shoots) was gained when 180 kg N ha^{-1} was applied (Table 2).

In 2010 precipitation distribution was more favourable and maximum values of the analysed characteristics were gained when 180 kg N ha^{-1} was applied, except for stem ratio (maximum value when 150 kg N ha^{-1} was applied) and leaf weight ratio (maximum in control). Coefficient of variation for all tested characteristics was low (Cv<16%). The largest variability was found in leaf number (9%<Cv<16%) and leaf weight ratio in total above-ground biomass (4%<Cv<12%) (Table 2).

Table 2a. Results of descriptive statistics and critical values of LSD test (2009 year)

Parameter	*Doses of N*	$\bar{x} \pm S\bar{x}$	*Median*	*Interv. of variat.*	*Coeff.of variat. (%)*
Plant height	N105	1.70±0.03	1.70	1.85-1.60	4.80
	N150	1.64±0.03	1.65	1.75-1.45	5.43
	N180	1.64±0.03	1.62	1.85-1.50	6.15
	control	1.67±0.03	1.65	1.85-1.60	4.82
		$LSD_{0.05}$: 0.077		$LSD_{0.01}$: 0.102	
Number of leafs	N105	7.70±0.26	7.50	9.00-7.00	10.69
	N150	7.00±0.21	7.00	8.00-6.00	9.52
	N180	7.80±0.20	8.00	9.00-7.00	8.11
	control	6.40±0.31	6.50	8.00-5.00	15.10
		$LSD_{0.05}$: 0.687		$LSD_{0.01}$: 0.904	
Leafiness	N105	22.98±0.28	22.79	24.78-75.22	3.92
	N150	27.52±0.26	27.63	28.70-25.87	2.94
	N180	28.55±0.90	29.26	33.04-25.21	10.02
	control	22.28±0.30	22.24	24.13-20.76	4.24
		$LSD_{0.05}$: 1.424		$LSD_{0.01}$: 1.875	
Proportion of trees (%)	N105	77.02±0.28	77.21	78.45-75.22	1.17
	N150	72.48±0.26	72.37	74.13-71.30	1.12
	N180	71.45±0.90	70.74	74.79-66.96	4.00
	control	77.67±0.31	77.76	79.24-75.87	1.25
		$LSD_{0.05}$: 1.413		$LSD_{0.01}$: 1.860	
Number of outgrowth	N105	293.67±1.86	295.00	296.00-290.00	1.09
	N150	315.33±8.19	318.00	328.00-300.00	4.50
	N180	421.67±7.26	420.00	435.00-410.00	2.98
	control	230.67±1.76	230.00	234.00-228.00	1.32
		$LSD_{0.05}$: 18.352		$LSD_{0.01}$: 26.700	

Table 2b. Results of descriptive statistics and critical values of LSD test (2010 year)

Parameter	*Doses of N*	$\bar{x} \pm S\bar{x}$	*Median*	*Interv. of variat.*	*Coeff.of variat.*
Plant height	N105	1.63±0.03	1.64	1.75-1.40	6.17
	N150	1.58±0.04	1.60	1.75-1.40	7.37
	N180	1.78±0.03	1.80	1.90-1.60	5.49
	control	1.76±0.04	1.74	2.09-1.59	7.89
		$LSD_{0.05}$: 0.100		$LSD_{0.01}$: 0.132	
Number of leafs	N105	7.50±0.27	7.50	9.00-6.00	11.33
	N150	7.10±0.38	7.00	9.00-5.00	16.86
	N180	8.40±0.37	8.50	10.00-7.00	13.97
	control	7.50±0.22	7.50	9.00-7.00	9.42
		$LSD_{0.05}$: 0.880		$LSD_{0.01}$: 1.158	
Leafiness	N105	26.91±0.36	26.58	28.85-25.30	4.29
	N150	25.88±0.87	24.36	30.34-23.42	10.61
	N180	29.21±1.10	28.41	35.78-24.87	11.91
	control	36.70±1.25	37.42	42.42-29.63	10.79
		$LSD_{0.05}$: 2.654		$LSD_{0.01}$: 3.494	
Proportion of trees (%)	N105	73.09±0.36	73.42	74.70-71.15	1.58
	N150	74.12±0.87	75.64	86.58-69.66	3.71
	N180	70.89±1.12	71.59	75.13-64.20	5.02
	control	63.30±1.25	62.58	70.37-58.58	6.26
		$LSD_{0.05}$: 2.673		$LSD_{0.01}$: 3.519	
Number of outgrowth	N105	299.33±2.40	298.00	304.00-296.00	1.39
	N150	330.00±12.10	325.00	353.00-312.00	6.35
	N180	360.67±23.33	354.00	404.00-324.00	11.20
	control	219.00±5.57	215.00	230.00-212.00	4.40
		$LSD_{0.05}$: 43.996		$LSD_{0.01}$: 64.009	

According to the results of Levene`s, Hartley`s, Cochran`s and Bartlet`s tests, variances were homogeneous for the analysed morphological characteristics: plant height, number of leaves and number of shoots, and heterogeneous for leaf ratio and stem ratio in both years, so that the significance of differences of average values was also tested by parametric model of analysis of variance (ANOVA) and nonparametric Kruskal-Wallis test (Table 3).

The results of parametric (ANOVA) and nonparametric (Kruskal-Wallis) methods of analysis of variance, as well as median test (Table 3) showed that all tested characteristics were statistically significant. Therefore, increased nitrogen doses had a statistically significant effect on the analysed morphological characteristics of this genotype. Our results are in agreement with CAMAKCI (1999) and PATAKI (2006). As stated by IKANOVIĆ *et al.* (2010) and RAKIC *et al.* (2013) nitrogen affects the quality of biomass, as voluminous feed, by increasing the percentage of leaf ratio in total biomass and content of total proteins.

By defining the relative degree of association between the analysed characteristics of this hybrid, based on Pearson`s correlation coefficient it can be concluded that plant height and number of leaves were very significantly positively correlated, while stem ratio and leaf ratio were very significantly negatively correlated. As number of shoots increased, leaf ratio and stem ratio significantly decreased (Table 4).

Table 3. Results of the tests for the analysed characteristics of the interspecies hybrid Siloking

Parameter	Levenes's test		Kolmogor`s Smyran's test	ANOVA		Kruskal-Wallis's test		Test median	
	F	p	p	F	p	H	p	χ^2	p
				2009 year					
Plant height (m)	0.153	0.927	> 0.10	1.090	0.366	2.875	0.411	1.94	0.584
Number of leaf	1.815	0.162	> 0.10	4.292	0.000	14.016	0.000	9.707	0.021
Leafiness (%)	11.758	0.000	<0.001	37.869	0.000	30.332	0.000	40.00	0.000
Proportion of trees (%)	11.696	0.000	<0.001	37.330	0.000	30.003	0.000	40.00	0.000
Number of outgrowth	2.421	0.141	< 0.10	199.77	0.000	10.385	0.016	12.00	0.007
				2010 year					
Plant height (m)	0.172	0.914	> 0.10	7.632	0.000	17.164	0.001	19.798	0.000
Number of leaf	1.341	0.276	> 0.10	3.000	0.043	6.016	0.111	2.400	0.494
Leafiness (%)	2.809	0.053	<0.10	26.037	0.000	23.808	0.000	18.400	0.000
Proportion of trees (%)	2.756	0.056	<0.10	25.680	0.000	23.293	0.000	18.400	0.000
Number of outgrowth	3.197	0.084	< 0.10	20.382	0.000	9.667	0.022	12.000	0.007

Table 4. Pearson's correlation coefficients between the tested traits

Parameter	Plant height	Number of leaf	Leafiness	Proportion of trees	Number of outgrowth
Plant height	-	0.810**	0.044^{ns}	-0.042^{ns}	-0.145^{ns}
Number of leaf		-	-0.006^{ns}	-0.002^{ns}	-0.182^{ns}
Leafiness			-	-0.995**	0.407*
Proportion of trees				-	-0.434*

ns - not significant ; * and ** significant at p<0.05 and p<0.01

Considering leaf ratio in total biomass as a primary feature, and according to mean two-year values of interspecies hybrid characteristics, the results of Ivanovic distance values for ranking utilized quantities of N fertilizer (based on the gained effects of qualitative traits) showed that most exuberant biomass of best quality was reached when 180 kg N ha^{-1} was applied (Table 5).

Table 5. I-distance values for the quality of the interspecies hybrid Siloking

Nitrogen	*I-distance*	*Ranking*
N 105	0,8677	IV
N 150	0,8909	III
N 180	**2,5700**	**I**
Control	1,6874	II

Results of IKANOVIĆ *et al*. (2010) show that forage sorghums positively react to intensive nitrogen nutrition and have a high coefficient of nitrogen utilization. This parameter is of interest to feed producers. Sorghum genotypes are plants with strong root system, good suction force and uptake of nitrogen salts remaining from previous crop. ĆUPINA *et al.* (2002) state that increased plant nutrition with nitrogen is justifiable only under conditions of favourable water regime (irrigation after each cutting). These results showed that decrease of applied nitrogen doses had no harmful effects on yield in relation to values most often used in the production of this forage mass. Recommendations for investing into this method of production could be altered with recommendations for limited irrigation, so that the conditions of the nutrition system are sustainable and profitable. MIRON (2007) points out that the benefit of sorghum lies in the fact that its above-ground biomass dries easily, and it is also suitable for silage. Sorghum biomass is of lower quality than corn biomass. However, interspecies hybrids have higher digestibility value than original species, and can completely replace corn as silage crop.

CONCLUSION

Results of this study show significant, positive and justified effect of using increased nitrogen quantities on the improvement of morphological characteristics of this hybrid. Weather conditions largely affected growing of the tested genotype, especially the quantity and distribution of precipitation. Optimal growth of fresh biomass was gained under conditions of favourable water regime when 150 kg N ha^{-1} was applied.

The plants reached maximum height when 105 kg N ha^{-1} was applied in the dry year, and 180 kg N ha^{-1} in the wet year. The lowest ratio of leaf weight was found in 2009 when 150 kg N ha^{-1} was applied. Increased nutrition with nitrogen significantly affected the intensity of tillering, number of leaves, and ratio of leaf weight in total above-ground biomass, which resulted from higher yield and better quality of fresh biomass.

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MORFOLOŠKE KARAKTERISTIKE INTERSPECIES HIBRIDA SIRKA I SUDANSKE TRAVE U USLOVIMA INTENZIVNE ISHRANE AZOTOM

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Izvod

Predmet ove studije su dvogodišnja istraživanja (2009. i 2010.) varijabilnosti morfoloških osobina vrste roda *Sorghum* interspecies hibrida sirka i sudanske trave *Siloking* zavisno od upotrebljenih količina azota. Analizirane su morfološke osobine: visina biljka, broj listova, udeo lista, udeo stabla, broj izdanaka, a uzorci su uzimani iz prvog otkosa kada je i efekat upotrebljenih azotnih hraniva bio najveći. Rezultati su pokazali da rastuće količine azota značajno utiču na ispitivane morfološke osobine posebno na intenzitet bokorenja (povećanje broja sekundarnih stabala), broj formiranih listova, kao i udeo lisne mase u ukupnoj nadzemnoj biomasi. Efekat upotrebljenog azota zavisio je od vremenskih uslova, odnosno od rasporeda padavina, te su biljke dostigle najveću visinu u varijanti sa primenjenih 105 kg ha^{-1} azota u sušnoj, a u varijanti sa 180 kg ha^{-1} u vlažnoj godini.

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