

## NON-PARAMETRIC APPROACH TO THE ANALYSIS OF PHENOTYPIC STABILITY OF TWO HALF-SIB MAIZE POPULATIONS

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One of the most important phases in commercial maize breeding programs is the assessment of the value of newly-developed progeny by testing in hybrid combinations. In this study, non-parametric stability measures were applied to analyze the genotype  $\times$  environment interaction and to assess phenotypic stability of two half-sib maize populations, each consisting of 40 genotypes, across 9 variable environments. Non-parametric tests of significance determined the presence of qualitative interaction for grain yield in both observed populations. Results of the stability analysis showed no significant differences between the two progeny groups indicating that the used testers did not bring significant increase in stability in either of the analyzed half-sib populations. Individual genotypes were also compared based on grain yield stability within both progeny groups using the stability parameters  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$  and  $S_i^{(6)}$ . Association between the grain yield and stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$  of the analyzed genotypes was presented graphically enabling the identification of genotypes which can be recommended for further breeding process as the most promising ones. The correlations between grain yield and stability parameters were tested by Spearman's rank correlations. Both progeny groups (HS1 and HS2) showed no significant correlations between the grain yield and stability parameters  $S_i^{(1)}$  and  $S_i^{(2)}$ , but the rank correlations between  $S_i^{(1)}$  and  $S_i^{(2)}$  values were very strong and highly significant. Highly significant negative correlations were

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found between grain yield and stability indices  $S_i^{(3)}$  and  $S_i^{(6)}$  in both progeny groups, and very strong and highly significant correlations were found between  $S_i^{(3)}$  and  $S_i^{(6)}$  values.

*Key words:* genotype by environment interaction, grain yield, stability indices

## INTRODUCTION

Pedigree breeding is one of the most widely used methods in commercial maize breeding programs. When in use, it is essential to comprehensively know the materials, to choose parents with complimentary traits for breeding starts, and to keep precise progeny records which show family relationships (HALLAUER and CARENA, 2009). By using the populations of narrow genetic base, pedigree breeding method produces elite inbred lines in the shortest time frame. The initial material in most commercial breeding programs comprises biparental populations from crossing two elite, most often related, inbred lines, while the lines of the opposite heterotic groups are used as testers. Besides biparental populations, two-parent backcross one (BC1) populations, tri-parental populations and commercial hybrids are the next most common types of populations used in inbred line development (MIKEL, 2006).

One of the most important phases in the breeding process is the assessment of the value of newly-developed progeny by testing in hybrid combinations. There are two major decisions to be made regarding this issue: at which inbreeding level should the tests be performed so as to assess their combining abilities, and which kind of tester should be selected for this purpose. In most cases, the assessment is performed in early generations of inbreeding by testing the progeny in trials on one or more different sites, while elite inbred lines of the opposite heterotic group are used as testers (LEE and TRACY, 2009). During the initial testing phases, the yield of individual progenies is the most important criterion for the following steps of the selection process, and phenotypic stability of the genotypes is usually disregarded at this stage.

According to the theory of quantitative genetics, the expression of quantitative traits is conditioned by the effect of genotype, environment, and their mutual interaction. The genotype  $\times$  environment interaction is present when phenotypic performance of a genotype varies in different environments, i.e. when genotypes react differently depending on the environment (MALOSETTI *et al.*, 2013). The presence of interaction complicates the breeding process and decreases the interdependence of phenotypic and genotypic values, which slows the progress of breeding. One of the ways to eliminate negative consequences of this interaction is to identify stable high-yielding genotypes that positively react to the environment they are grown in.

In general, there are two major approaches for studying the genotype  $\times$  environment interaction. The first one is parametric approach, which implies relating observed genotypic responses to a sample of environmental conditions, and the second one is non-parametric approach, which is based on the genotype rankings across the environments (SEGHERLOO *et al.*, 2008). In the analysis of the interaction, the main role of the experimental design and statistical models is to eliminate the inexplicable variability as much as possible—the noise present in data (ABAY and BJØRNSTAD, 2009). Numerous statistical methods have been developed to examine the interaction. They can be based on the analysis of variance, linear regression, non-linear analysis, multivariate analysis, biplot analysis, or on the methods of non-parametric statistics (BALESTRE *et al.*, 2009). When selecting a model, one should keep in mind that the interaction can occur due to numerous factors and that in fact it represents a reflection of prevalent stress factors at the time the experiment was conducted (drought, diseases, pests). If there are only one or two prevalent factors, then models based on the linear regression are adequate. A more

complex structure of the interaction effect refers to multivariate models that are best adapted to different data sets (BABIĆ *et al.*, 2011). HUEHN (1996) lists some of the advantages of non-parametric models, such as decreased bias in the analysis of interaction caused by extreme values, normal distribution of frequencies is not necessary, homogeneity of variances is not necessary, and addition or deletion of one or more genotypes does not affect the outcome of the analysis (orthogonality of the model). Furthermore, they are easy to use and interpret and more reliable in cases when some of the necessary assumptions for the application of parametric models are violated (TRUBERG and HUEHN, 2000).

According to non-parametric models for the assessment of stability, a genotype will be considered stable if it has a relatively constant ranking in various environments (FLORES *et al.*, 1998). HUEHN (1979) and NASSAR and HUEHN (1987) proposed four non-parametric measures of phenotypic stability:  $S_i^{(1)}$  is the mean of the absolute rank differences of a genotype over the  $n$  environments,  $S_i^{(2)}$  is the variance among the ranks over environments,  $S_i^{(3)}$  is the sum of the absolute deviations for each genotype relative to the mean of ranks, and  $S_i^{(6)}$  is the sum of squares of rank for each genotype relative to the mean of ranks.

Identification of desirable progenies with high yield potential and stability, especially in early generations of inbreeding, is one of the most important steps in commercial maize breeding programs. In relation to this, the rank orders of the genotypes across the environments can be the most essential information. The objective of this study was: i) to analyze the genotype  $\times$  environment interaction on grain yield of two half-sib maize populations and to examine the effect of different testers on grain yield stability, and ii) to identify the individual genotypes that have high yields and stable performances across diverse test environments.

## MATERIAL AND METHODS

### *Genetic material*

The development of two half-sib maize populations started in 2006 with initial crosses of two inbred lines 260341NS (75% Stiff Stalk Synthetic, 25% local) and 462NS (Stiff Stalk Synthetic). In winter nursery 2006/2007,  $F_1$  plants were selfed and about 500  $S_1$  plants were grown in Novi Sad, Serbia during the season of 2007. The earliest 150 individual plants were selfed and after phenotypic selection 50 desirable plants were harvested. In winter nursery 2007/2008, selected plants were grown ear to row and selfed to produce  $S_3$  generation. In the season of 2008, 20 randomly chosen plants from each of the 40  $S_3$  progenies were crossed with two genetically distinct testers. Both testers were lines developed at the Institute of Field and Vegetable Crops, Novi Sad, Serbia. The first tester, 72NS, belongs to Iodent, while the second one, 26NS, belongs to the Lancaster heterotic group. As a result of crossing, two half-sib populations were obtained, each consisting of 40 genotypes whereby each of the forty  $S_3$  progenies was crossed with both testers.

### *Field trials*

The trials were set up as a randomized complete block design with two blocks, on three sites in Serbia (Rimski Šančevi, Srbobran, and Bački Petrovac) in 2009, 2010, and 2011. Combinations of years and locations were treated as 9 environments. The planting was done mechanically, with the distance of 0.75 m between rows and 0.22 m within rows, providing the plant density of 60.606 plants per hectare. The basic plot size was 9.8 m<sup>2</sup> for each genotype (two rows of 6.5 m in length). Conventional cultivation technology was applied, which was adapted to

the ecological conditions of the location. The harvest was done mechanically and grain yield ( $t\ ha^{-1}$ , adjusted on 14% of moisture content) and grain moisture content (%) were recorded for each assessed progeny per basic plot.

### Statistical analyses

Non-parametric testing of the genotype  $\times$  environment interaction was performed according to Bredencamp and de Kroon and Van der Laan (HUEHN, 1996). The method proposed by BREDENCAMP (1974) is based on the linear model for the interaction in which it is a deviation from the additive main effects, and in this case used for the test of non-crossover interaction. The method proposed by DE KROON and VAN DER LAAN (1981) was used to test crossover interaction. This method is based on the transformation of the original data into ranks and the analysis of their values.

Yield stability of both progeny groups was analyzed using the non-parametric method through the stability indices  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$  and  $S_i^{(6)}$  (NASSAR and HUEHN, 1987). The non-parametric stability index  $S_i^{(1)}$  is the mean of genotype rank differences over the studied environments and is calculated using this formula:

$$S_i^{(1)} = 2 \sum_{j=1}^{N-1} \sum_{j'=j+1}^{N-1} \frac{|r_{ij} - r_{ij'}|}{[N(N-1)]}$$

where  $r_{ij}$  is the rank of genotype  $i$  in the environment  $j$ .

$S_i^{(2)}$  is the variance of genotype ranks over the studied environments and is calculated using this formula:

$$S_i^{(2)} = \sum_{j=1}^N \frac{(r_{ij} - \bar{r}_i)^2}{N-1}$$

where  $\bar{r}_i = \sum_{j=1}^N \frac{r_{ij}}{N}$

Based on the non-parametric indices, a genotype  $i$  is considered stable if its ranking is approximately the same across all the environments, and maximum stability is expressed by the genotype with values  $S_i^{(1)}$  and  $S_i^{(2)} = 0$ , i.e. its ranking is identical across all the environments.

It is also possible to test the significance for the values  $S_i^{(1)}$  and  $S_i^{(2)}$  by using the following formula (HUEHN and NASSAR, 1989):

$$Z_i^{(m)} = \frac{[S_i^{(m)} - E\{S_i^{(m)}\}]^2}{\text{var}\{S_i^{(m)}\}}, m = 1, 2$$

where  $Z_i^{(m)}$  has approximate  $\chi^2$  distribution with degree of freedom 1. In order to enable the testing it is necessary to determine the mean value  $E\{S_i^{(m)}\}$  and variance  $\text{var}\{S_i^{(m)}\}$  whereby  $S_i^{(m)} < E\{S_i^{(m)}\}$  for the stable genotype, and  $S_i^{(m)} > E\{S_i^{(m)}\}$  for the unstable genotype.

Parameters  $S_i^{(3)}$  and  $S_i^{(6)}$  combine yield and stability based on yield ranks of genotypes in each environment. These parameters measure stability in units of the mean rank of each genotype and they are calculated as follows:

$$S_i^{(3)} = \sum_{j=1}^N \frac{(r_{ij} - \bar{r}_i)^2}{\bar{r}_i}$$

$$S_i^{(6)} = \sum_{j=1}^N \frac{|r_{ij} - \bar{r}_i|}{\bar{r}_i}$$

Stability indices  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$ , and  $S_i^{(6)}$  were mutually compared by using Spearman's rank correlations. All described analyses were performed within R computing environment (R CORE TEAM, 2016).

## RESULTS AND DISCUSSION

### *Test of significance for genotype × environment interaction*

There are two types of interactions depending on the genotype performance in different environments. An interaction can be non-crossover when genotype rank is constant in all the environments or crossover when genotype rank differs across the environments. Appropriate non-parametric tests of significance for both the non-crossover and crossover type of interaction were applied in order to determine the type of interaction. The null hypothesis is the absence of non-crossover interaction according to BREDENKAMP (1974), and the absence of crossover interaction according to DE KROON and VAN DER LAAN (1981). For two-dimensional data consisting of rows (genotypes) and columns (environments), there are two concepts of crossover interaction: different genotypes rank within the environments  $G \times (E)$  and different environments rank within the genotypes  $E \times (G)$  (TRUBERG and HUEHN, 2000). Based on the results given in Table 1 for both progeny groups, values of non-crossover interaction were not significant according to Bredekamp method, while significant values ( $P < 0.01$ ) were found for  $G \times (E)$  type of crossover interaction and not significant for  $E \times (G)$  according to de Kroon and van der Laan method.

Table 1. Chi-square test statistics for genotype by environment interaction for grain yield

Population	Bredekamp (non-crossover)	de Kroon and van der Laan (crossover)	
		G(E)	E(G)
HS1	127.75 <sup>ns</sup>	102.27**	295.88 <sup>ns</sup>
HS2	126.08 <sup>ns</sup>	105.19**	306.14 <sup>ns</sup>

\*\* $p < 0.01$ ; ns - non significant

### *Stability analysis*

Yield stability of both progeny groups was analyzed through non-parametric stability indices  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$ , and  $S_i^{(6)}$ . Regarding stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$ , the most stable genotype is the one with the value  $S_i^{(1)} = 0$  and the lowest possible variance of rank  $S_i^{(2)}$  in the studied environments. As for  $S_i^{(3)}$  and  $S_i^{(6)}$ , their lowest values indicate maximum stability of a given genotype.

Genotypes within both progeny groups showed different values of stability parameters, i.e. some were more stable than others. However, there were no significant differences in the

stability between the two progeny groups, which was confirmed by Mann-Whitney test (Table 2). Such results show that the used testers did not bring significant increase in stability in either of the analyzed half-sib populations.

Table 2. Mann-Whitney test of differences among two half-sib maize populations for nonparametric stability measures for grain yield

Population	$S_i^{(1)}$	$S_i^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$
HS1:HS2	0.916 <sup>ns</sup>	0.981 <sup>ns</sup>	0.958 <sup>ns</sup>	0.912 <sup>ns</sup>

ns - non significant

Even though there were no available previous studies on the effect of testers on the maize population stability by using non-parametric stability indices, ŽIVANOVIĆ *et al.* (2012) used this method and parameters  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$  and  $S_i^{(6)}$  to analyze the changes in grain yield stability between F<sub>2</sub> maize populations after five recombination cycles. Mann-Whitney test for  $S_i^{(1)}$  and  $S_i^{(2)}$  values did not show significant differences in the stability between the cycles. The same authors found significant differences between the cycles only for the parameters  $S_i^{(3)}$  and  $S_i^{(6)}$  and concluded that the differences were most probably due to differences in the yield *per se*, and not due to stability because the values of those parameters are based simultaneously on the yield and the stability.

Individual genotypes were compared based on grain yield stability within both the first and the second progeny group. Within the first progeny group, genotypes G1, G18, G39, and G40 had the least rank differences, or the highest stability, according to the values of stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$ , while the genotype G4 had the lowest stability (Table 3). Within the second progeny group, genotypes G10, G40, G2, and G26 were the most stable, while the least stable were genotype G28 according to  $S_i^{(1)}$  and genotype G31 according to  $S_i^{(2)}$  (Table 4).

NASSAR and HUEHN (1987) suggested the significance test for  $S_i^{(1)}$  and  $S_i^{(2)}$ . Based on the ranks of corrected  $S_i^{(1)}$  and  $S_i^{(2)}$  data, the  $Z_i^{(1)}$  and  $Z_i^{(2)}$  values are calculated and their sum is compared with the critical values of  $\chi^2$  test. Within the first progeny group, the sums of  $Z_i^{(1)}$  and  $Z_i^{(2)}$  values were 39.53 and 44.28 respectively, which is below the critical value  $\chi^2 = 55.76$  (Table 3). Within the second progeny group, the sums  $Z_i^{(1)} = 38.17$  and  $Z_i^{(2)} = 40.19$  were also below the critical value  $\chi^2 = 55.76$ , and therefore within both groups there were no significant differences in the stability between the progeny (Table 4).

Based on the other two stability parameters  $S_i^{(3)}$  and  $S_i^{(6)}$  within the first progeny group, genotypes G1, G22, G28, and G2 were the most stable, and genotype G8 was the least stable (Table 3). Within the second progeny group, genotypes G2, G10, G35, and G19 had the lowest values of  $S_i^{(3)}$  and  $S_i^{(6)}$  which means they were the most stable, while genotype G33 was the least stable across the studied environments (Table 4). The basic shortcoming of  $S_i^{(3)}$  and  $S_i^{(6)}$  stability parameters is the fact that they are negatively correlated with the yield. In other words, the most stable genotypes are the ones with the lowest  $S_i^{(3)}$  and  $S_i^{(6)}$  values, but they are simultaneously characterized by low grain yields, which makes these parameters less reliable for the identification of high-yielding and stable genotypes (SAGHERLOO *et al.*, 2008).

Table 3. Mean grain yield (t/ha), estimation and test of nonparametric stability measures of the first progeny group (HS1)

Genotype	Yield	$S_i^{(1)}$	$Z_i^{(1)}$	$S_i^{(2)}$	$Z_i^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$
G1	10.1	9.0	3.16	59.8	2.61	3.3	0.7
G2	10.1	14.8	0.38	157.3	0.28	19.7	2.2
G3	11.6	12.4	0.15	110.8	0.24	43.0	4.5
G4	11.0	18.4	4.43	260.8	7.86	84.1	5.6
G5	11.0	16.7	1.95	209.2	2.79	62.3	4.7
G6	11.7	15.3	0.68	172.4	0.74	81.6	6.8
G7	11.4	16.9	2.15	195.9	1.90	84.1	6.0
G8	11.5	16.9	2.15	198.5	2.06	103.8	7.2
G9	10.0	14.7	0.30	153.7	0.20	36.6	2.6
G10	10.7	13.8	0.04	135.0	0.00	35.8	3.0
G11	10.6	18.2	4.05	237.5	5.25	44.2	3.5
G12	10.4	15.6	0.84	174.8	0.83	38.1	3.0
G13	10.5	15.3	0.64	162.9	0.43	38.9	3.0
G14	10.7	14.4	0.19	142.4	0.04	38.0	3.1
G15	11.9	14.4	0.19	142.8	0.04	62.7	5.8
G16	10.9	15.4	0.72	172.5	0.75	46.9	3.7
G17	10.5	15.8	1.06	186.4	1.37	29.1	2.7
G18	10.8	9.3	2.69	72.4	1.79	21.0	2.3
G19	11.6	15.9	1.16	197.1	1.97	68.4	6.0
G20	11.0	15.3	0.68	168.6	0.60	54.6	4.4
G21	11.3	17.3	2.64	210.9	2.92	87.7	5.5
G22	10.3	12.5	0.12	109.8	0.27	16.4	1.6
G23	11.1	14.1	0.09	139.3	0.02	44.8	3.7
G24	11.9	13.2	0.00	120.6	0.08	58.8	5.2
G25	11.0	12.6	0.09	114.4	0.17	34.2	3.2
G26	11.3	14.1	0.09	148.6	0.11	49.0	4.4
G27	11.4	13.9	0.05	138.4	0.01	55.8	4.6
G28	10.6	13.9	0.05	133.3	0.00	17.6	2.1
G29	11.3	14.7	0.33	155.5	0.24	57.5	4.4
G30	11.4	10.7	1.14	79.5	1.40	26.1	3.3
G31	11.7	16.5	1.70	201.0	2.22	102.1	7.3
G32	11.2	12.7	0.06	116.3	0.14	46.7	3.6
G33	11.3	11.4	0.63	101.5	0.49	45.6	4.4
G34	11.9	13.7	0.02	134.7	0.00	64.8	5.2
G35	11.5	14.4	0.19	144.4	0.06	52.5	4.7
G36	10.7	14.3	0.15	144.6	0.06	27.6	2.8
G37	11.3	13.0	0.02	121.4	0.07	52.4	4.0
G38	10.8	15.2	0.57	158.0	0.30	38.6	3.1
G39	11.9	9.6	2.33	66.1	2.18	28.7	3.8
G40	11.2	10.2	1.63	72.4	1.79	23.9	2.5
Sum			39.53		44.28		
		$\chi^2_{Z_i^{(1)}, Z_i^{(2)}}=10.4$			$\chi^2_{\text{sum}}=55.76$		

Table 4. Mean grain yield (t/ha), estimation and test of nonparametric stability measures of the second progeny group (HS2)

Genotype	Yield	$S_i^{(1)}$	$Z_i^{(1)}$	$S_i^{(2)}$	$Z_i^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$
G1	9.1	15.9	1.11	176.3	0.89	23.5	2.3
G2	9.2	10.6	1.24	82.4	1.25	13.5	1.6
G3	10.3	12.2	0.21	131.3	0.00	38.8	3.3
G4	10.8	15.0	0.47	156.7	0.27	70.2	5.6
G5	9.9	11.1	0.83	89.7	0.92	24.4	2.5
G6	10.1	16.2	1.42	187.0	1.40	58.2	4.3
G7	9.8	15.9	1.11	181.3	1.12	45.7	3.5
G8	9.8	16.6	1.76	194.9	1.84	52.8	3.9
G9	9.6	15.1	0.51	157.4	0.28	37.0	3.2
G10	9.6	8.7	3.58	55.9	2.89	15.1	1.9
G11	10.4	14.9	0.41	158.0	0.30	62.8	5.3
G12	10.0	14.4	0.21	147.2	0.09	55.1	4.4
G13	10.4	14.2	0.14	140.8	0.03	56.9	4.8
G14	10.2	16.4	1.59	188.3	1.46	57.6	4.5
G15	9.9	16.8	2.08	201.4	2.24	63.9	4.2
G16	10.0	13.7	0.02	135.3	0.00	36.0	3.0
G17	9.6	15.1	0.51	156.3	0.26	32.1	2.5
G18	9.5	12.3	0.17	118.5	0.11	34.4	2.1
G19	9.8	11.4	0.60	95.4	0.69	21.7	1.9
G20	10.8	13.0	0.02	118.7	0.10	52.1	4.7
G21	10.7	13.4	0.00	125.1	0.03	57.8	5.6
G22	9.9	15.3	0.64	170.3	0.66	50.9	4.1
G23	10.9	11.7	0.43	99.0	0.57	29.6	4.1
G24	10.9	14.9	0.41	172.3	0.74	61.5	5.9
G25	10.5	16.3	1.47	186.4	1.36	85.1	5.8
G26	10.4	11.0	0.91	88.5	0.97	33.5	3.1
G27	10.4	13.2	0.00	137.0	0.01	69.6	4.7
G28	9.8	18.2	3.96	235.3	5.03	68.5	4.6
G29	10.0	11.3	0.71	89.3	0.94	23.1	2.5
G30	10.8	15.6	0.84	176.3	0.89	45.5	4.8
G31	9.9	17.9	3.52	236.9	5.20	75.8	5.1
G32	10.1	14.6	0.28	153.8	0.20	48.7	3.7
G33	10.7	17.3	2.64	213.2	3.09	99.1	7.3
G34	9.9	12.3	0.17	115.9	0.14	35.5	2.8
G35	9.3	13.1	0.01	118.0	0.11	15.8	1.7
G36	9.9	15.7	0.97	170.7	0.68	42.6	3.6
G37	10.3	14.6	0.28	159.9	0.34	67.5	4.9
G38	10.5	15.6	0.88	169.6	0.64	73.5	5.1
G39	10.6	11.8	0.38	96.4	0.66	36.1	3.9
G40	11.0	10.2	1.68	72.3	1.80	28.0	3.9
Sum			38.17		40.19		
		$\chi^2_{Z_i^{(1)}, Z_i^{(2)}}=10.4$			$\chi^2_{\text{sum}}=55.76$		



In commercial maize breeding programs, selection of progeny that will participate in further breeding process is mostly performed on the basis of general combining ability for grain yield. By simultaneous inclusion of stability analysis it is possible to assess the value of individual progenies more precisely and make the selection criterion more objective. One of the ways to enable visual identification of the most desirable genotypes regarding both yields and stability is to graphically display the associations between grain yields and stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$  of the analyzed genotypes. Figure 1(a, b, c, d) shows mean values of grain yield and stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$  which separate the figures into four sectors. The lower right sector of each figure shows the genotypes with yields above the mean, but with low  $S_i^{(1)}$  and  $S_i^{(2)}$  values, which makes them stable. The upper right sector shows the genotypes with yields above the mean, but with high  $S_i^{(1)}$  and  $S_i^{(2)}$  values, characterizing them as unstable. The lower left and upper left sectors show the genotypes with yields below the mean for the given progeny group.

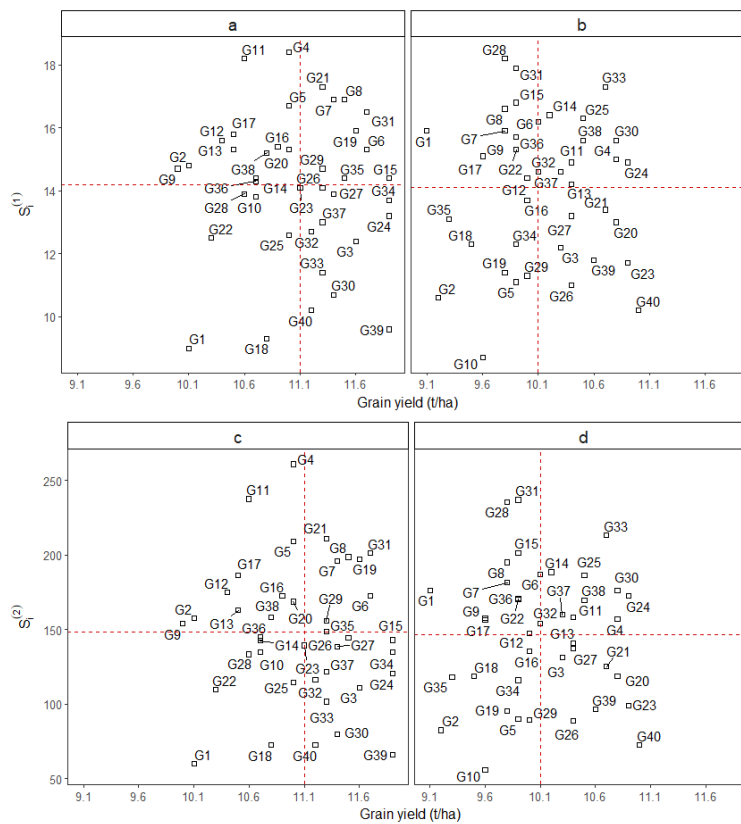


Figure 1. (a,b) Association between grain yield and  $S_i^{(1)}$  value of two maize progeny groups (HS1 and HS2); (c,d) Association between grain yield and  $S_i^{(2)}$  value of two maize progeny groups (HS1 and HS2)

Although the test of significance showed that there were no statistically significant differences between stability of various progenies, the following is observable in the Figure 1: within the first progeny group, lower right sector of the figure 1a includes the genotypes G39, G34, G24, G3, G30, G40, G33, G32, G37 and G27. The second progeny group shows the genotypes G40, G23, G20, G21, G27, G39, G26, and G3 in the lower right sector of Figure 1b. These genotypes stand out as the most stable and with yields above the mean within their progeny groups. Similar distribution of genotypes is shown in Figure 1 c, d which shows the associations between grain yields and  $S_i^{(2)}$  values. Selected genotypes are the ones that will take part in further breeding process consisting of visual phenotypic selection in each generation of inbreeding until  $S_5$  or  $S_6$  generation, after which the progenies are evaluated again in testcross combinations including more testers and environments (LEE and KANNENBERG, 2004). Furthermore, a thorough analysis of the selected genotypes showed that the same progenies within two progeny groups most often displayed a high level of stability with different testers. Genotypes 3, 27, 39 and 40 simultaneously showed high stability and yields (above average) in the crossings to both testers, and therefore these progenies are very promising and should be recommended for accelerated testing, including several testers and locations. Despite the fact that the applied methods give a useful insight on the genotype stability, the stability parameters should not be regarded as the only elements for a definitive conclusion. This especially refers to the genotypes with higher yield and lower stability, like genotypes 15, 6 and 31 from the first group and genotypes 24, 4 and 30 from the second group. These genotypes could also be included in further trials and evaluations, however the focus should remain on their specific adaptations on certain growing regions.

#### ***Correlations between yield and non-parametric stability measures***

The correlations between grain yield and stability parameters were tested by Spearman's rank correlations. Both progeny groups (HS1 and HS2) showed no significant correlations between the grain yield and stability parameters  $S_i^{(1)}$  and  $S_i^{(2)}$ , but the rank correlations between  $S_i^{(1)}$  and  $S_i^{(2)}$  values were very strong and highly significant (0.984 in HS1 and 0.992 in HS2) (Table 5). Highly significant negative correlations were found between grain yield and stability indices  $S_i^{(3)}$  and  $S_i^{(6)}$  in both progeny groups, and very strong and highly significant correlations were found between  $S_i^{(3)}$  and  $S_i^{(6)}$  values (Table 5).

*Table 5. Spearman's coefficients of rank correlation for the mean grain yield and nonparametric stability measures for the first (below diagonal) and second (above diagonal) progeny groups (HS1 and HS2)*

	Yield	$S_i^{(1)}$	$S_i^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$
Yield	-	-0.067	-0.065	-0.653**	-0.821**
$S_i^{(1)}$	-0.0829	-	0.992**	0.601**	0.443**
$S_i^{(2)}$	-0.0246	0.9841**	-	0.606**	0.447**
$S_i^{(3)}$	-0.4281**	0.6962**	0.7298**	-	0.943**
$S_i^{(6)}$	-0.7126**	0.5128**	0.5620**	0.8985**	-

\*\*p<0.01

High values of rank correlation coefficients between  $S_i^{(1)}$  and  $S_i^{(2)}$  values were also found by DELIĆ *et al.* (2009), FLORES *et al.* (1998), SCAPIM *et al.* (2000) and BALALIĆ *et al.* (2011). The presence of high rank correlations between stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$  justifies the selection of one of them for the proper stability assessment. HUEHN (1990) favored  $S_i^{(1)}$  parameter because it is simple to calculate and interpret the results. Due to their strong negative correlations with grain yields, the stability indices  $S_i^{(3)}$  and  $S_i^{(6)}$  are not recommended in plant breeding. Negative correlations between these indices and yields were also found by MOHAMMADI *et al.* (2007), SAGHERLOO *et al.* (2008), and DEGHGANI *et al.* (2016).

Numerous statistical models have been proposed so far, and they can successfully be used in the assessment of genotype adaptability and stability. However, difficulties often arise when selecting the most appropriate one. When choosing the adequate model, the following should be taken into consideration: the number of genotypes and environments in the analysis, differences among the environments, mathematical models which correspond to the given data set, defined concept of stability, and possibility of simple application and interpretation of results (FARSHADFAR *et al.*, 2012). According to SCAPIM *et al.* (2000), non-parametric stability indices are a useful alternative to the parametric methods. Their application does not necessitate normal distribution and homogeneity of variances, or orthogonality of the data. According to most studies, the basic shortcoming of stability indices  $S_i^{(1)}$  and  $S_i^{(2)}$  is their association with the static (biological) concept of stability (FARSHADFAR *et al.*, 2012; NASSAR and HUEHN, 1987; SCAPIM *et al.*, 2000; MOHAMMADI *et al.*, 2009). This concept of stability is generally unacceptable to most plant breeders and agronomists, who give priority to high-yielding genotypes with positive reaction to improved growing technology and favorable environments (BECKER, 1981). A small sample (low number of genotypes) may decrease the reliability of these indices, which should be avoided whenever possible.

#### CONCLUSIONS

Within this research, stability analysis of two half-sib maize populations was assessed based on the results of early progeny testing, which is the most widely used method for inbred line development in commercial maize breeding programs. The results of group stability analysis of two half-sib populations did not show significant differences among them, which leads to the conclusion that the used testers did not affect the increase of stability in either of the analyzed half-sib populations. However, certain progenies showed less yield variation across environments, which characterizes them as more stable than others. The genotypes within the first progeny group are G39, G34, G24, G3, G30, G40, G33, G32, G37 and G27, while the genotypes with desirable traits from the second group are G40, G23, G20, G21, G27, G39, G26, and G3. In addition, genotypes 3, 27, 39 and 40 showed high stability and yields in the crossings to both testers which makes them very promising for further applications. Considering this, by using the yield and yield stability simultaneously as a selection criterion, it can be expected that the selected progenies will provide the basis for development of hybrids with high genetic yield potential and desirable phenotypic stability.

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## NEPARAMETRIJSKA ANALIZA FENOTIPSKU STABILNOSTI DVE POLUSRODNIČKE POPULACIJE KUKURUZA

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### Izvod

U ovom radu, primenom neparametrijskih pokazatelja stabilnosti, analizirana je interakcija genotip-spoljašnja sredina i fenotipska stabilnost dve polusrodničke populacije kukuruza na 9 različitih lokaliteta. Neparametrijskim testovima ustanovljeno je prisustvo unakrsne interakcije za prinos zrna u obe posmatrane populacije. Rezultatima analize stabilnosti ustanovljeno je da ne postoje statistički značajne razlike između dve grupe potomstava, što ukazuje da korišćeni testeri nisu uticali na povećanje stabilnosti ni kod jedne od analiziranih populacija. Parametrima stabilnosti  $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$  i  $S_i^{(6)}$  poređene su razlike u stabilnosti prinosa zrna između pojedinačnih genotipova u obe posmatrane populacije. Takođe, predstavljen je i grafički prikaz odnosa između prinosa zrna i stabilnosti koji omogućava identifikaciju najperspektivnijih genotipova za dalji proces selekcije. Međuzavisnost prinosa zrna i parametara stabilnosti  $S_i^{(1)}$  i  $S_i^{(2)}$  testirana je primenom Spearman-ovih korelacija ranga. U obe grupe potomstava nisu ustanovljene korelacione veze između prinosa zrna i parametara stabilnosti  $S_i^{(1)}$  i  $S_i^{(2)}$ , dok su korelacije ranga između parametara  $S_i^{(1)}$  i  $S_i^{(2)}$  bile veoma jake i visoko značajne. Visoko značajne negativne korelacione veze ustanovljene su između prinosa zrna i parametara stabilnosti  $S_i^{(3)}$  i  $S_i^{(6)}$ , dok su veoma jake i visoko značajne korelacione veze ustanovljene između parametara  $S_i^{(3)}$  i  $S_i^{(6)}$ .

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