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Non-parametric Measures for Yield Stability in Grass Pea (*Lathyrus sativus* L.) Advanced Lines in Semi Warm Regions

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ABSTRACT

Multi-environment trials play a significant role in selecting the best cultivars to be used at different locations. The objective of this study was to identify grain and forage yields stability of grass pea advanced lines across different locations. The 14 advanced lines of grass pea, developed by the International Center for Agricultural Research in Dry Areas (ICARDA), were tested at three different research stations in semi-warm regions of Iran for three consecutive years. Ten non-parametric measures of stability were used to identify stable lines across nine environments. Three non-parametric tests (Bredenkamp, Hildebrand and De Kroon and Van der Laan) for Genotype-Environment (GE) interaction were highly significant, recommending differential responses of the lines to the test environments. Mean yields had a significant positive correlation with $S_i^{(6)}$, NP2, NP3, NP4, Fox-rank and Kang's rank-sum statistics. The results of correlation analysis and principal components analysis indicated that only non-parametric superiority measure could be useful for simultaneous selection of high yielding and stable lines. According to cluster analysis by forage and grain mean yields and non-parametric statistics, the line L3 with the highest forage and grain yields and Fox-rank as well as the lowest values of other non-parametric statistics could be introduced as high yielding stable cultivar for rain-fed conditions of semi-warm areas.

Keywords: Fox-rank, GE interaction, Rain-fed conditions, Interaction effect, Grass pea.

INTRODUCTION

Grass pea (*Lathyrus sativus* L.) is the most important grain legume cultivated as one of the cheapest sources of dietary lysine-rich protein for the people of low income countries and also as forage for farm animals. Grass pea is a major crop in parts of Asia, northern Africa, southern Europe (France and Spain), and, to a lesser extent, in the Middle East countries such as Iran (Milczak *et al.,* 2001). The important chemical composition of grass pea seeds includes protein content and total dietary fiber content (Kasprzak and Pzedzicki, 2008). Grass pea seeds are also a rich source of a number of proactive non-nutritional components of food such as oligosaccharides, tannins and phytoestrogens. In general, three main characteristics of this grain legume consist of its massiveness, drought tolerance, and adaptability to a wide range of soil types, including the marginal lands (Ahmadi *et al.,* 2012b).

The development of genotypes, which can be adapted to a wide range of diversified environments, is the final objective of plant

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breeders in a crop improvement program. Major goal of plant breeding programs is to increase stability and stabilize crop yield over a range of environments (Segherloo *et al.,* 2008). Genotype-Environment (GE) interaction effects are of major importance to the plant breeding developing improved varieties or genotypes. These interaction effects are a major problem when comparing the performance of genotypes across environments (Kang, 1990). When genotypic performance in different environments is extremely different, GE becomes a major challenge to selection and genetic improvement programs (Zobel and Talbert, 1984). Therefore, a more stable genotype as compared to others, should give relatively more stable yield across the environments. There are two major GE stability approaches; the first one is a parametric approach which relies on distributional assumptions and involves relating the observed genotypic responses, and the second one is non-parametric approach, which defines environments and phenotypes relative to biotic and abiotic factors and any needs assumptions (Huehn, 1990). The non-parametric statistics cluster genotypes according to their similarity of response to a range of environments (Lin *et al.,* 1986). The non-parametric measures for stability based on ranks provide a valuable alternative to existing parametric measures based on absolute data (Akcura and Kaya, 2008) and, also, these do not require any assumption about the normality and independence of observation as well as homogeneity of error variances. In addition, when sample size is very small, non-parametric methods are the obvious choice, unless the nature of the population is exactly known (Huehn, 1990). Several non-parametric statistics have been expanded by biometricians to define and interpret the responses of genotypes to environmental variation. Huehn (1979) and Nassar and Huehn (1987) suggested four non-parametric statistics, namely $S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$ and $S_i^{(6)}$ based on the classification of the genotypes in each environment, and described stable genotypes as those whose position in relation to the others remained unaltered in the set of environments assessed. Kang (1988) proposed a method based on yield performance and Shukla's stability variance (Shukla, 1972) for selecting high yielding and stable genotypes. Fox *et al*. (1990) using the stratify ranking of the cultivars suggested another non-parametric measure for general adaptability. In this method, integration of stability of performance with yield is necessary for selecting high-yielding, stable genotypes. Thennarasu (1995) introduced four non-parametric statistics (NP1, NP2, NP3 and NP4) based on ranks of adjusted means of the genotypes in each environment, and described stable genotypes as those whose position in relation to the others remained unaltered in the set of the studied environments .

Parametric and non-parametric methods for estimating GE interactions and phenotypic stability are widely used in plant breeding, although these methods have been used to evaluate stability and test environments in many crops like *Linum* (Adugna and Labuschangne, 2003), *Chenopodium* (Bhargava *et al.,* 2007), wheat (Mohammadi *et al.,* 2007; Mohammadi and Amri, 2008; Ahmadi *et al.,* 2012a), maize (Scapim *et al.,* 2010), safflower (Jamshidmoghaddam and Pourdad, 2013) and chickpea (Segherloo *et al.,* 2008). Unfortunately, relatively few reports provide information on the phenotypic stability studies in forage crops. In Iran, the information about the GE interaction for forage crops such as grass pea is very limited. Thus, the objectives of the present study were to: (i) evaluate the adaptation and stability of some advanced lines of grass pea selected from the ICARDA, (ii) identify advanced lines that have both high mean yield and stable yield performance across different environments for semi warm regions of Iran, and (iii) study the relationships among non-parametric stability methods.

MATERIALS AND METHODS

Experimental Sites, Design and Plant Materials

The present study was conducted across nine environments, including three semi warm sites in Kermanshah, Gachsaran, and Lorestan during 2005/2006, 2006/2007, and 2007/2008 growing seasons under rain-fed conditions. The three different climate locations are located in the semi-warm regions of Iran. The detailed description of these test locations is shown in Table 1. In each year and location, 14 advanced lines of grass pea were tested. These lines were developed by the International Center for Agricultural Research in Dry Areas (ICARDA). The names, lines code and origin of these lines are given in Table 2. In each environment (year×location), the experimental design was a randomized complete block design with three replications. Each plot had four rows of 4.5 m length with spacing of 25 cm between rows. The seeding rate was 150 seeds per m^2 in all the environments. Crop management practices, such as weeds control, were carried out by hand during crop growth and development. In all experiments, for each line, the central four rows were harvested for grain yield measurement in order to exclude border effects. Forage (at 50% flowering stage) and grain yields at physiological maturity (kg ha^{-1}) were obtained by converting the yields obtained from the plots to hectares.

Statistical Analysis

In order to test the significance of GE interaction, three non-parametric statistical methods consisting of Bredenkamp (1974), Hildebrand (1980) and De Kroon and Van der Laan (1981) were used. The methods of Hildebrand and Bredenkamp are based on the usual linear model for interaction. The method of De Kroon and Van der Laan defines the interaction according to the crossover interaction model. Huehn and Leon (1995) indicated that the Hildebrand method depends on the concept of interactions as deviations from additively, and the De Kroon and Van der Laan method depends on a crossover interaction concept. Thus, the De Kroon and Van der Laan method is suggested if the crossover interaction concept is intended and non-parametric methods must be applied because the assumptions for the parametric methods are not valid. If the usual interaction concept and non-parametric methods must be applied, the Hildebrand method is suggested. On the other hand, the null hypothesis for Bredenkamp is not crossover type for genotype by environment interactions. For non-parametric measures, Nassar and Huehn (1987) suggested four non-parametric stability statistics including $S_i^{(1)}$, $Si^{(2)}$, $S_i^{(3)}$ and $S_i^{(6)}$. These parameters, based on yield ranks of lines in each environment, are estimated as follows:

$$S_i^{(1)} = 2\sum_{j}^{n-1} \frac{\sum_{j'=j+1}^{n} \left| r_{ij} - r_{ij'} \right|}{\left[N(n-1) \right]} \bar{X}, \quad (1)$$

$$S_i^{(2)} = \frac{\sum_{j=1}^{n} (r_{ij} - \bar{r}_{i.})^2}{(N-1)}, \quad (2)$$

$$S_i^{(3)} = \frac{\sum_{j=1}^{n} (r_{ij} - \bar{r}_{i.})^2}{\bar{r}_{i.}}, \quad (3)$$

$$S_i^{(6)} = \frac{\sum_{j=1}^{n} \left| r_{ij} - \bar{r}_{i.} \right|}{\bar{r}_{i.}} \quad (4)$$

Where, r_{ij}, $\bar{r}_{ij}$ and N is the rank of the *i*th genotype in j^{th} environment, the mean rank across all environments for each genotype and number of environments, respectively. The lowest value for each of these statistics revealed high stability for a certain genotype. Also, the significance tests for the $S_i^{(1)}$ and $S_i^{(2)}$ statistics were developed in a manner that was similar to that proposed by Nassar and Huhn (1987). The rank of a genotype in a specific environment cannot be based on the phenotypic values, because the stability has to be measured

Table 1. Agro-climatic characteristics of environments in yield stability experiments for 14 grass pea advanced lines.

Environments		GY^a (kg h^{-1})	FY^b (kg h^{-1})	Coordinate		Altitude	Rainfall	Soil condition	
Location	Year			Latitude	Longitude	(m)	(mm)	Texture	Type [c]
Kermanshah	2006	1126.14	5824.28	34°19" N	47°70" E	1,322	505	Silt-loam	Cambisols
	2007	1296.50	7152.50				551		
	2008	407.21	2612.83				159.2		
Gachsaran	2006	2017.52	6052.14	30°10" N	50°50" E	669.5	560.7	Silt-loam	Regosols
	2007	1689.00	7629.52				511.2		
	2008	678.619	3122.71				184.5		
Lorestan	2006	2071.92	5457.23	23°26" N	48°17" E	1,147.7	438.3	Silt-loam	Regosols
	2007	2228.71	6505.23				557.8		
	2008	857.57	3223.38				251		

[a] Grain Yield , [b] Forage Yield, respectively.[c] Type according to FAO system of soil classification.

Table 2. Line code, name, and origin of 14 grass pea advance lines evaluated in nine environments of Iran.

Line code	Name	Origin	Line code	Name	Origin
L1	Sel 515	ICARDA	L8	Sel 1307	ICARDA
L2	Sel 1326	ICARDA	L9	Sel 554	ICARDA
L3	Sel 474	ICARDA	L10	Sel 1332	ICARDA
L4	Sel 1329	ICARDA	L11	Sel 678	ICARDA
L5	Sel 686	ICARDA	L12	Sel 736	ICARDA
L6	Sel 459	ICARDA	L13	Sel 1327	ICARDA
L7	Sel 669	ICARDA	L14	Sel 1321	ICARDA

independently of the genotypic effect. Therefore, the rank of the i^{th} genotype in the jth environment is determined on the basis of the corrected phenotypic values, namely, ($X^*_{ij}= X_{ij}-\bar{x}_{i.}+\bar{x}_{..}$), where, X_{ij} is the performance of the ith genotype in the jth environment, $\bar{x}_{i.}$ is the mean performance of the i^{th} genotype and $\bar{x}_{..}$ is the overall mean in across environments. Accordingly, Thennarasu (1995) proposed four non-parametric stability parameters based on adjusted ranks of genotypes within each test environment. The formulas to compute these methods are shown below:

$$NP1 = \frac{1}{N}\sum_{j=1}^{n}\left|r^*_{ij} - M^*_{di}\right| \text{ , (5)}$$

$$NP2 = \frac{1}{N}\left[\sum_{j=1}^{n}\left|r^*_{ij} - M^*_{di}\right| / M_{di}\right], \quad (6)$$

$$NP3 = \frac{\sqrt{\frac{\sum(r^*_{ij} - \bar{r}^*_{i.})^2}{N}}}{\bar{r}_{i.}} \quad (7)$$

$$NP4 = \frac{2}{N(N-1)}\left[\sum_{j=1}^{n-1}\sum_{[j'=j+1]}^{n}\left|r^*_{ij} - r^*_{ij'}\right| / \bar{r}_{i.}\right] \quad (8)$$

Where, r^*_{ij} is the rank of i^{th} genotype in the j^{th} environment based on adjusted data, $\bar{r}^*_{ij}$ and M^*_{di} are mean and median ranks, respectively, for adjusted values, while $\bar{r}_{i.}$ and M_{di} are the mean and median ranks of the i^{th} genotype in the j^{th} environment, respectively. Also, $\bar{r}^*_{ij}$ and M^*_{di} are the mean and median ranks obtained from the original data (un-adjusted). Coupled with this, Fox *et al*. (1990) suggested another non-parametric superiority measure for general adaptability. This classified ranking method consists of scoring the number of environments in which each genotype ranked in the top, middle, and bottom thirds of trial entries. The genotype that occurred mostly in the top third (high top value) was considered a widely adapted cultivar. Kang's rank-sum (Kang, 1988) is another non-parametric stability measure that utilizes both yield and Shukla' stability variance (Shukla, 1972) as selection criteria. This parameter gives a weight of one to both yield and stability statistics to identify high-yielding and stable genotypes. The genotype with the highest yield and lower stability variance is assigned a rank of one and all genotypes are ranked in this way. The ranks of yield and stability variance were added for each genotype and the genotypes with the lowest rank-sum are the most desirable. Spearman's rank correlation was calculated to measure the relationship among the statistics using SAS software (1987). To better understand the relationships among the non-parametric statistics, a Principal Component Analysis (PCA) based on ranks of stability parameters were performed by STATISTICA software (2007). To cluster the lines, a hierarchical cluster analysis based on mean yield and stability measures was performed. The Euclidean distance was used as a dissimilarity measure required in Ward's clustering method (Ward, 1963), and the discriminant analysis test was used to estimate the optimal number of clusters.

RESULTS

The results of significance test for GE interaction with different non-parametric statistical measures are given in Table 3. The χ^2 value was used to test the effects of G, E, and GE interaction effect. As shown in Table 3, Bredenkamp (1974), Hildebrand (1980), and De Kroon and Van der Laan (1981) statistics were significant.

Non-parametric Stability Analysis

The results of the stability analysis for grain and forage yields based on $S_i^{(1)}$ to $S_i^{(6)}$ and NP1 to NP4 non-parametric statistics as well as rank-sum and general adaptability are shown in Table 4. Taking mean yield as a first parameter for assessing the lines, L1, L3, and L9 gave the highest mean grain and forage yields, while L10, L13, L2, L4, and

Table 3. The significance test of genotype × environment interaction for 14 grass pea advanced lines evaluated in nine environments of Iran.

Statistics	*df*	Forage yield χ^2	Grain yield χ^2
De Kroon and Van der Laan	104	1190.606**	1151.977**
Hildebrand	104	3633.941**	3636.027**
Bredenkamp	104	1152.008**	1151.975**

**: Significant at the 0.01 probability level.

Table 4. Mean values and non-parametric stability statistics for forage and grain yields and tests of non-parametric stability (Z_1 and Z_2) for 14 advanced lines of grass pea evaluated in nine environments of Iran.

Yield	Code	Mean (kg h^{-1})	$S_i^{(1)a}$	$Z^{(1)}$	$S_i^{(2)}$	$Z^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$	NP1[b]	NP2	NP3	NP4	Rank-sum[c]	Fox-rank[d]
Stability measures based on forage yield	G1	5459.70	4.33	0.13	13.78	0.20	13.05	2.89	3.33	0.42	0.48	0.51	11	22.22
	G2	5094.85	4.67	0.00	15.28	0.03	18.97	4.72	3.00	0.50	0.54	0.72	14	22.22
	G3	6176.85	1.89	10.58	2.78	5.96	1.79	0.93	4.00	0.31	0.36	0.15	11	88.89
	G4	4825.96	5.17	0.38	18.61	0.18	25.28	5.28	3.89	0.65	0.79	0.88	25	22.22
	G5	5669.19	5.11	0.31	18.25	0.13	16.22	3.56	3.33	0.33	0.47	0.57	12	55.56
	G6	5256.78	3.44	2.00	8.86	1.79	9.97	2.69	2.11	0.30	0.37	0.48	9	22.22
	G7	5347.59	3.50	1.82	10.28	1.17	10.88	3.09	2.67	0.30	0.40	0.46	12	44.44
	G8	4789.78	4.78	0.03	16.03	0.00	19.56	4.64	4.33	0.62	0.79	0.73	27	33.33
	G9	5687.85	5.28	0.56	21.03	0.75	17.81	3.76	4.33	0.36	0.51	0.56	13	55.56
	G10	4775.63	2.44	6.74	4.86	4.26	10.00	4.29	1.89	0.63	0.67	0.63	16	0.00
	G11	5395.89	4.00	0.58	11.36	0.79	13.19	3.35	2.89	0.41	0.52	0.58	13	33.33
	G12	5485.44	5.44	0.90	22.94	1.47	20.91	4.35	3.78	0.31	0.49	0.62	16	44.44
	G13	4838.96	3.33	2.39	8.44	2.00	14.14	4.47	2.44	0.61	0.66	0.70	14	11.11
	G14	5208.63	4.89	0.08	17.50	0.05	18.26	3.91	3.33	0.48	0.53	0.64	17	33.33
Stability measures based on grain yield	G1	1381.74	5.06	0.24	18.28	0.14	19.64	3.94	3.11	0.44	0.51	0.68	11	33.33
	G2	1305.33	3.67	1.33	9.94	1.31	16.65	5.40	3.22	1.07	0.76	0.77	16	0.00
	G3	1548.11	3.39	2.19	8.36	2.04	6.14	1.94	3.22	0.27	0.38	0.31	14	77.78
	G4	1309.67	3.89	0.79	10.53	1.08	13.07	3.66	3.00	0.43	0.58	0.60	20	11.11
	G5	1453.81	5.11	0.31	19.78	0.41	16.75	3.34	3.11	0.28	0.40	0.54	11	55.56
	G6	1376.26	3.72	1.18	10.11	1.24	9.97	3.10	2.56	0.37	0.41	0.46	9	44.44
	G7	1412.52	5.28	0.56	20.19	0.51	19.65	4.05	3.33	0.37	0.48	0.64	10	44.44
	G8	1329.48	6.44	4.53	30.19	6.39	33.45	6.12	4.89	0.81	0.73	0.89	24	44.44
	G9	1479.37	4.72	0.01	15.75	0.01	13.50	3.07	3.33	0.33	0.44	0.51	13	55.56
	G10	1269.48	3.56	1.65	9.28	1.60	13.63	3.92	2.56	0.51	0.59	0.65	21	11.11
	G11	1352.07	3.61	1.49	9.25	1.61	11.10	3.10	2.22	0.37	0.42	0.54	10	22.22
	G12	1391.59	5.17	0.38	18.75	0.21	18.75	4.25	4.22	0.60	0.58	0.65	15	44.44
	G13	1275.56	5.61	1.31	22.75	1.39	30.33	6.33	4.00	1.00	0.74	0.94	25	33.33
	G14	1362.22	4.17	0.32	12.11	0.56	14.06	3.65	3.00	0.38	0.54	0.60	11	22.22

[a] Si: Huehn's (1979) non-parametric stability indices; [b] NP: Thennarasu's (1995) non-parametric stability indices; [c] Kang's (1988) stability index, [d] Fox *et al*. (1990) stability index.

Test statistics: $E(S_i^{(1)})$= 4.64 *and* $Var(S_i^{(1)})$= 0.72, $E(S_i^{(2)})$= 16.25 *and* $Var(S_i^{(2)})$= 30.45.

χ^2 sum= 23.68 and χ^2 Z_1, Z_2= 3.84; Grand mean for grain= 1347.80 (kg h^{-1}), and Grand mean for forage= 5286.65

L8 had the lowest mean grain yield across environments. The significance tests of $S_i^{(1)}$ and $S_i^{(2)}$ (Z_1 and Z_2, respectively) were derived from Nassar and Huehn (1987). For each line, Z_1 and Z_2 values were estimated based on the ranks of adjusted and summed data over lines to obtain Z values (Z_1 and Z_2 sums were 16.28 and 18.47, respectively). Since both of these statistics were less than the critical value of $\chi^2_{(0.05, df=14)} = 23.68$, no significant differences in rank stability were found among the 14 advanced lines grown in nine environments. However, the individual Z-values for some lines were significant because they showed large Z-values, in comparison with the critical value $\chi^2_{(0.05, df=1)} = 3.84$.

Non-parametric Measures of Stability for Grain Yield

Huehn's (1979) and Thennarasu's (1995) superiority index (Fox *et al*., 1990) and Kang's (1988) non-parametric statistics of stability for grain yield of 14 advanced lines are presented in Table 4. According to the $S_i^{(1)}$ and $S_i^{(2)}$, lines L3, L11, and L10 with the lowest value were identified as the stable lines for grain yield, while, the unstable lines were L8, L7 and L13. Based on $S_i^{(3)}$, lines L3, L7 and L9 were stable. On the other hand, lines L8, L13, and L7 with highest values were identified as the unstable lines. According to the $S_i^{(6)}$, lines L3, L9, and L6 had the lowest value and L13, L8, and L2 had relatively higher values of this statistic, indicating higher and lower stability, respectively. According to Thennarasu's (1995) stability statistics (NP1, NP2, NP3 and NP4), lines with minimum values are considered more stable. Based on NP1, the lines L11, L10, and L6 with lower values were identified as stable in comparison to other lines. However, the lines L8, L12, and L13 had the highest values of NP1. According to the values of NP2, lines L3, L5, and L9 had the lowest value, while lines L2, L13, and L8 had the highest values and were considered as unstable in comparison to other lines. Lines L3, L5, and L6 had the lowest NP3 values and, therefore, were the most stable lines. However, lines L1, L2, and L8 with maximum values were identified as unstable lines. Also, according to the NP4, line L3, followed by L6 and L9, had the lowest value and, therefore, were the most stable. But, lines L1, L2, and L8 had the highest NP4 and were the unstable lines. The highest value of non-parametric superiority index (Fox *et al.,* 1990) was shown by L3, followed by L5 and L9. These lines were adapted lines, because they ranked in the top third of lines in a high percentage of environments (77.78 and 55.56%, respectively). Also, based on this method, L2, L4, and L10 were identified as unstable lines. Kang's rank-sum stability parameter (Kang, 1988) indicated that lines L6, L7, and L11 with the lowest value were the stable lines and L13, L8, and L10 with highest values were identified as the unstable lines.

Non-parametric Measures of Stability for Forage Yield

The lowest value for each statistic of stability used indicates maximum stability for a certain line. Accordingly, the $S_i^{(1)}$ and $S_i^{(2)}$ of the tested lines showed that L3, L10, and L13 had the lowest value. The unstable lines based on these parameters were L12, L9, and L4. As for the $S_i^{(3)}$, line L3 followed by L6 and L10 with lowest values were the most stable lines, while the lines L4, L12, and L8 were identified as more unstable than the other lines. Based on the $S_i^{(6)}$, lines L3, L6, and L1 were stable, whereas, lines L4, L2, and L8 were the least stable ones (Table 4). Moreover, the results of Thennarasu's (1995) non-parametric stability statistics are shown in Table 4. According to the first stability statistic (*NP1*), lines L10, L6, and L13 were stable in comparison with the other lines. Lines L7, L6, and L3 with the lowest value of NP2 were stable and L8, L9 and L3 with highest value were identified as unstable lines. The results of NP2, NP3 and NP4 were similar to each other and nominated L3, L6 and L7 as stable lines, so that these lines had the desirable mean yield performance. However, based on NP3 and NP4 parameters, the lines L10 and L8 were unstable. According to non-parametric superiority measure (Fox *et al.,* 1990), L3, L9, and L5 occurred mostly in the top third, thus, these lines were stable. The unstable lines of this method were L2, L4, and L10, because these lines occurred mostly in the bottom third of the ranks. Using the Kang's rank-sum stability parameter (Kang, 1988), lines L6, L3, and L1 were identified as the most desirable lines.

Relationships among Mean Yields and Non-parametric Measures

The results of the Spearman's coefficients of rank correlation among mean yields and the ten non-parametric stability statistics are shown in Table 5. Mean yields had a significant positive correlation with $S_i^{(6)}$, NP2, NP3, NP4, Fox-rank (Fox *et al.,* 1990) and Kang's rank-sum (Kang, 1988) and insignificant negative correlation with $S_i^{(1)}$, $S_i^{(2)}$ and NP1. Correlations between Kang's rank-sum (Kang, 1988) index with $S_i^{(3)}$, $S_i^{(6)}$, NP2, NP3 and NP4 estimated based on forage yield were positive and significant. Also, correlation between Kang's rank-sum in terms of grain yield with $S_i^{(6)}$, *NP2*, *NP3,* and *NP4* was positive and significant. The Fox-rank had positive correlation with NP2 and NP3, and negative significant correlation with NP1. The stability statistics $S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$ and $S_i^{(6)}$ obtained for grain yield had a significant positive correlation with each other and $S_i^{(1)}$, $S_i^{(2)}$ had a significant positive correlation with NP1, while $S_i^{(3)}$ and $S_i^{(6)}$ had significant positive correlations with NP1, NP2, NP3, and NP4. On the other hand, in terms of forage yield, the $S_i^{(1)}$ and $S_i^{(2)}$ measures were positively and significantly correlated with each other and with NP1. Also, the $S_i^{(3)}$ and $S_i^{(6)}$ measures had significant positive correlation with each other, and with NP3 and NP4. The high correlation between mean yield and stability parameters was expected, as the values of these statistics were higher for high yielding genotypes.

In order to obtain information on the relationships, similarities, and differences among the non-parametric stability statistics,

Table 5. Spearman's coefficients of rank correlation among mean grain (upper main diameter) and forage (down main diameter) yields and 10 non-parametric stability statistics of 14 grass pea advanced lines evaluated in nine environments of Iran.

Parameter	$S_i^{(1)a}$	$S_i^{(2)}$	$S_i^{(3)}$	$S_i^{(6)}$	NP1[b]	NP2	NP3	NP4	Mean	Rank -sum[c]	Fox-rank[d]
$S_i^{(1)}$		0.99**	0.86**	0.62*	0.72**	0.31	0.31	0.49	-0.06	0.18	-0.28
$S_i^{(2)}$	1.00**		0.87**	0.61*	0.69**	0.29	0.30	0.50	-0.05	0.21	-0.28
$S_i^{(3)}$	0.84**	0.83**		0.85**	0.67**	0.59*	0.57*	0.81**	0.19	0.35	0.02
$S_i^{(6)}$	0.44	0.44	0.82**		0.54*	0.90**	0.86**	0.95**	0.58*	0.57*	0.40
NP1	0.59*	0.59*	0.51	0.16		0.33	0.37	0.43	-0.21	0.46	-0.43
NP2	0.09	0.09	0.47	0.76**	0.04		0.96**	0.92**	0.77**	0.65*	0.64*
NP3	0.21	0.21	0.61*	0.87**	0.07	0.93**		0.89**	0.77**	0.74**	0.64*
NP4	0.35	0.35	0.767*	0.95**	0.11	0.86**	0.93**		0.66**	0.61*	0.51
Mean	-0.18	-0.18	0.24	0.65*	-0.37	0.75**	0.79**	0.73**		0.55*	0.84**
Rank-sum	0.42	0.42	0.76**	0.87**	0.26	0.74**	0.88**	0.88**	0.64*		0.26
Fox-rank	-0.33	-0.33	-0.01	0.39	-0.61*	0.64*	0.55*	0.49	0.79**	0.26	

[a] S_i: Huehn's (1979) and Nassar and Huehn (1987) non-parametric stability indices; [b] NP: Thennarasu's (1995) non-parametric stability indices; [c] Kang's (1988) stability index, [d] Fox *et al.* (1990) stability index.

Principal Component Analysis (PCA) based on the rank correlation matrix was performed. The main advantage of using PCA over cluster analysis is that each statistic can be assigned to one group only. The relationships among different non-parametric statistics are graphically displayed in a biplot graph (Figure 1). The first two PCAs justified 86.63 and 87.87% of the total variation for ranks of stability statistics and mean grain and forage yields, respectively. The PCA_1 versus PCA_2 were used to produce the biplot illustrated in Figure 1. According to both biplots, the $S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$ and NP1 statistics were negatively associated with mean yields and were placed in group I. The grouping of these stability statistics related to the concept of static stability and not to genotypic mean yield. Group II consisted of Kang's rank-sum, $S_i^{(6)}$, NP2, NP3 and NP4 statistics. Group III included Fox-rank stability parameter and mean yields. The clustering of mean yields into this group indicates that mean yields had the main influence on the ranking across environments.

Clustering Lines Based on Mean Yields and Non-parametric Statistics

In order to group grass pea advanced lines tested in terms of high yielding and stability, cluster analysis based on mean yields and non-parametric statistics was performed (Figures 2 and 3). Cluster analysis based on forage and grain mean yields and their related non-parametric statistics separated the 14 advanced lines into three main groups. According to mean of grain yield and its related non-parametric statistics, the group I included the low yielding lines L13 and L8. Also, these lines with higher value of non-parametric statistics were identified as unstable lines. The lines L2, L10 and L4, which had low yields and higher values of non-parametric statistics, clustered in group II. The other lines were clustered in two subgroups as group III, such that the first subgroup included L3 (high yielding), and L6, L11, and L14 (moderate yielding). The line L3 was identified as the stable line by Fox-rank and Huehn's (1979) and Thennarasu's (1995) stability indices. The other lines in this subgroup had a relatively

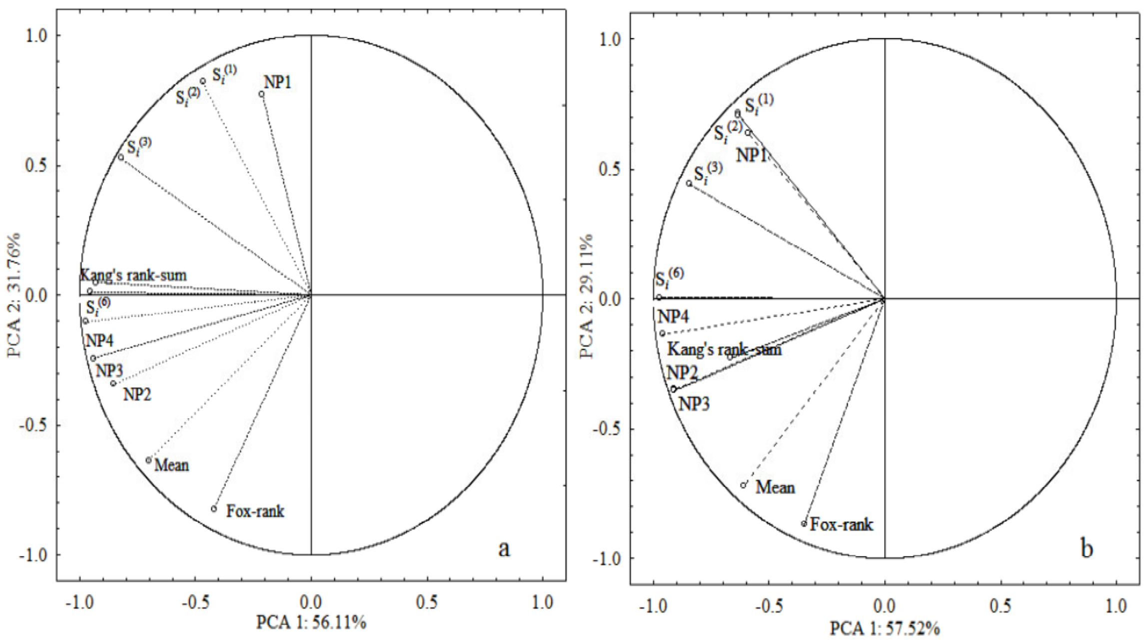

Figure 1. Biplot of PCA1 *versus* PCA2 for grouping mean of: (a) Forage yield and (b) Grain yield with non-parametric statistics of stability in14 grass pea advanced lines evaluated in nine environments of Iran.

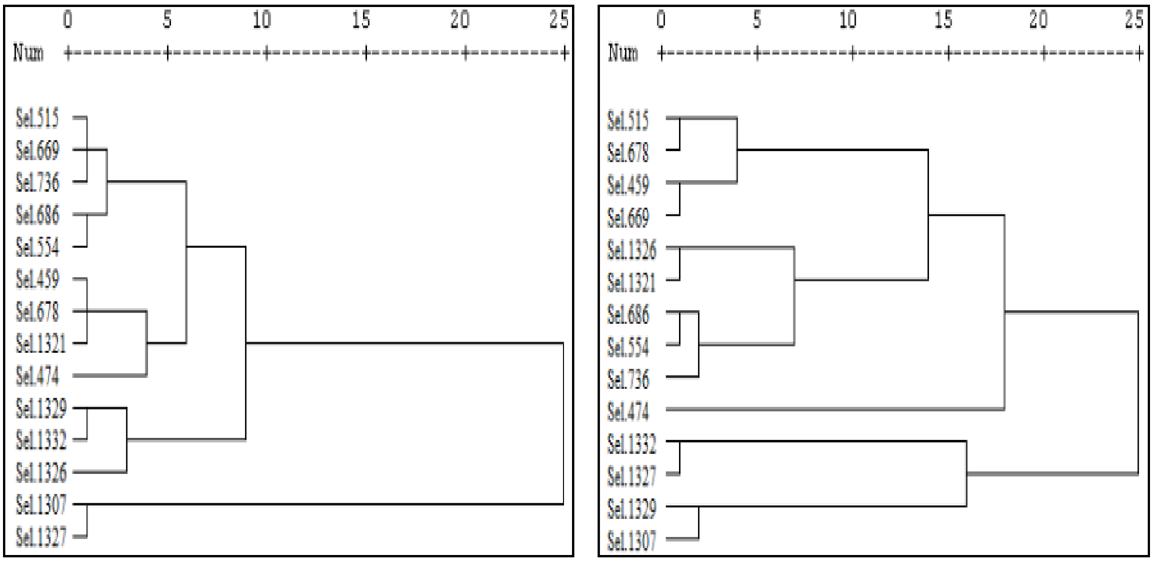

Figure. 2 Dendrogram showing hierarchical classification of 14 grass pea advanced lines based on non-weighted values of 10 non-parametric stability statistics and mean of grain (right) and forage (left) yields.

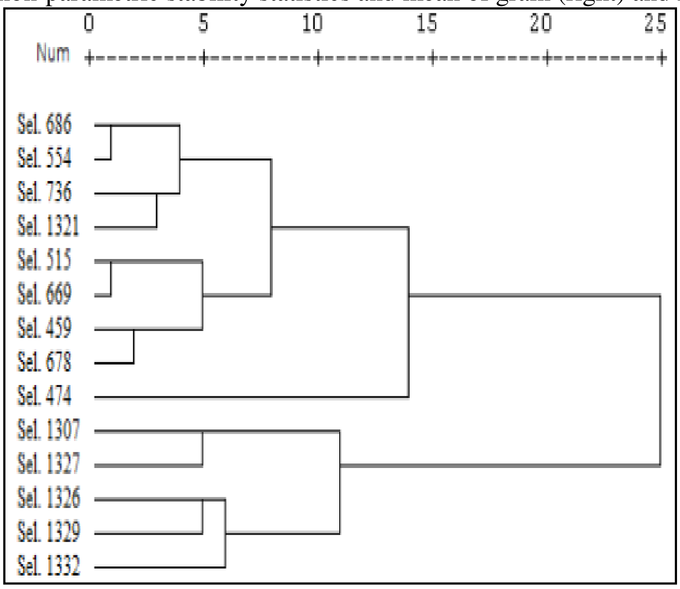

Figure 3. Dendrogram showing hierarchical classification of 14 grass pea advanced lines based on non-weighted values of 10 non-parametric stability statistics with both grain and forage mean yields.

moderate value of non-parametric statistics. The second subgroup from group III comprised the lines L1, L7, L12, L5, and L9. The lines L5 and L7 had a high mean yield and the L5 was identified as stable by Thennarasu's stability parameters (Figure 2). Based on the mean of forage yield and its non-parametric statistics, the group I consisted of lines L8, L4, L13, and L10. These lines had the lowest forage yield and the highest non-parametric values, therefore, these lines were recognized as the unstable lines. The line L3, singly placed in the second group with the highest forage yield, was also identified as the stable line by Kang's rank-sum, Fox-rank and Huehn's and Thennarasu's stability indices. The other lines, which had moderate forage yields, clustered in group III and included lines L1, L11, L6, L7, L2, L14, L9, L12 and L5, of which lines L6 and L7 were identified as stable by Kang's rank-sum, Huehn's and Thennarasu's stability indices. Since the results of non-parametric statistics based on grain and forage yields were different, to identify stable lines with both the highest grain and forage yields, the cluster analysis was performed based on mean yields and non-parametric statistics (Figure 3). Based on the obtained dendrogram, the group I comprised the low yielding lines L4, L10, L2, L13, and L8 that had a relatively higher value of non-parametric statistics and were identified as unstable lines. The other lines, which had high and moderate yields, clustered in group II. The group II constituted two subgroups, and the first subgroup included the line L3. This high yielding line was introduced as stable by all of the stability indices. Most of the lines, which had moderate yields clustered in second subgroup. For all lines in this subgroup, with a few exceptions, the ranks of non-parametric measures were in agreement with that of overall ranks.

DISCUSSION

A significant genotype×environment interaction effect often limits researcher's ability to select high yielding and stable genotypes in breeding programs (Kang *et al*., 1991). In the present study, the stability of lines was assessed using 10 non-parametric measures of stability viz. $S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$ and $S_i^{(6)}$ (Huehn, 1979), NP1, NP2, NP3 and NP4 (Thennarasu, 1995) as well as Kang's rank-sum and Fox-rank. The results of non-parametric tests for the GE interaction effect indicated the same level of significance for Bredenkamp (1974), Hildebrand (1980) and De Kroon and Van der Laan methods (Table 3). Similar results were reported by Huehn and Leon (1995), Segherloo *et al*. (2008) and Mohammadi *et al*. (2007). We found that the three non-parametric statistics of Huehn (1979) ($S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$) and *NP1* statistic of Thennarasu (1990) clustered together. For both mean yields, the $S_i^{(1)}$, $S_i^{(2)}$ and NP1 were positively and significantly correlated, revealing that the three measures were similar under different environmental conditions. Therefore, only one of these parameters would be sufficient to select stable genotypes in a breeding program (Mohammadi *et al.,* 2007). Sagherloo *et al.* (2008) also found a significant positive association among these statistics in chick pea (*Cicer arietinum* L.). Mohammadi *et al.* (2007) reported significant positive correlations between $S_i^{(1)}$ and $S_i^{(2)}$ in durum wheat. Nassar and Huehn (1987) reported that $S_i^{(1)}$ and $S_i^{(2)}$ were associated with the static concept of stability. The stability statistics of $S_i^{(1)}$, $S_i^{(2)}$ and *NP1* indicate static concepts of stability, and are not correlated with mean yield. Consequently, these stability statistics could be used as parallel methods to select genotypes with high stability and moderate yield. According to non-parametric statistics calculated based on ranks in terms of both mean yields, $S_i^{(6)}$, NP2, NP3, and NP4 statistics were positively correlated with mean yields, and thus are recommended for use in line selection. According to our study, the highly positive correlation between Fox-rank and Kang's rank-sum with mean yields showed that these indices were the best to identify high yielding lines (Table 5). Also, considering biplot of principal component analysis, the two first PCAs axes recognized TOP (Fox *et al.,* 1990) and mean yields as a one group (group III) from the other statistics. Flores *et al*. (1998) suggested that the TOP parameter was associated with mean yield and the dynamic concept of stability. The group I stability statistics represent a static concept of stability, and were correlated neither positively nor negatively with mean yield. Therefore, the group I statistics could be used as compromise approaches that select lines with moderate yield and high stability. Clustering of $S_i^{(6)}$, NP2, NP3, NP4 and Kang's rank-sum statistics in the same group indicated that these statistics were similar under different environmental conditions. In addition, like the group II, these statistics identify lines that are stable based on the static or biological concept of stability, but dissimilar group II, they were also strongly positively correlated with high yield. Therefore, only one of these statistics would be adequate for selecting the stable lines in a breeding program (Sabaghnia *et al.,* 2006; Mohammadi *et al.,* 2007). Several parametric and nonparametric statistics of stability have been presented and compared in the literature (Lin *et al.,* 1986; Flores *et al.,* 1998). To make recommendations, it is essential to assess the relationship among these statistics and compare their powers for different stability models.

Overall, both yield and stability of performance should be considered simultaneously to advantage the useful effect of GE interaction and to make the selection of the lines more precise and refined. In conclusion, our results revealed that, among the lines tested at different environments, the line L3, namely, Sel. 474, could be introduced as the cultivar with high forage and grain yields as well as the most stable line for rain-fed conditions of semi-warm areas.

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REFERENCES

1. Adugna, W. and Labuschagne, M. 2003. Parametric and Nonparametric Measures of Phenotypic Stability in Linseed (*Linum Usitatissimum* L.). *Euphytica,* **129**: 211-218.
2. Ahmadi, A., Mohammadi, A. and Najafi Mirak, T. 2012a. Targeting Promising Bread Wheat (*Triticum aestivum* L.) Lines for Cold Climate Growing Environments Using AMMI and SREG GGE Biplot Analyses. *J. Agr. Sci. Tech.,* **14**: 645-657
3. Ahmadi, J., Vaezi, B., Shaabani, A. and Khademi, K. 2012b. Multi-environment Yield Trials of Grass Pea (*Lathyrus sativus* L.) in Iran Using AMMI and SREG GGE. *J. Agr. Sci. Tech.,* **14**: 1075-1085.
4. Akcura, M. and Kaya, Y. 2008. Nonparametric Stability Methods for Interpreting Genotype by Environment Interaction of Bread Wheat Genotypes (*Triticum aestivum* L.). *Genet. Mol. Biol.,* **31**: 906-913.
5. Bhargava, A., Shukla, S. and Ohri, D. 2007. Evaluation of Forage Yield and Leaf Quality Traits in *Chenopodium* Spp. in Multiyear Trials. *Euphytica,* **153**: 199-213.
6. Bredenkamp, J. 1974. Nonparametric Prufung von Wechsewirkungen. *Psychol. Beitr.,* **16**: 398-416.
7. De Kroon, J. and Van der Laan, P. 1981. Distribution-free Test Procedures in Two-way Layouts: A Concept of Rank Interaction. *Stat Neeri.,* **35**: 189-213.
8. Eberhart, S. and Russell, W. 1966. Stability Parameters for Comparing Varieties. *Crop. Sci.,* **6**: 36-40.
9. Flores, F., Moreno, M. T. and Cubero, J. I. 1998. A Comparison of Univariate and Multivariate Methods to Analyze Environments. *Field. Crop. Res.,* **56**: 271-286.
10. Fox, P., Skovmand, B., Thompson, B., Braun, H. J. and Cormier, R. 1990. Yield and Adaptation of Hexaploid Spring Triticale. *Euphytica,* **47**: 57-64.
11. Hildebrand, H. 1980. Asymptotosch Verteilungsfreie Rangtests in Linearen Modellen. Med. *Inform. Stak.,* **17**: 344-349.
12. Huehn, M. 1979. Beitrage Zur Erfassung der Phanotypischen Stabilitat. *Edv. Med. Biol.,* **10**: 112-117.
13. Huehn, M. 1990. Nonparametric Measures of Phenotypic Stability. Part 1. Theory. *Euphytica,* **47**: 189-194.
14. Huehn, M. 1996. Non-parametric Analysis of Genotype×Environment Interactions by Ranks. In: "*Genotype by Environment Interaction*", (Eds.): Kang, M. S. and Gauch H. G. CRC Press, Boca Raton, FL, USA, PP. 213-228
15. Huhn, M. and Leon, J. 1995. Nonparametric Analysis of Cultivar Performance Trials: Experimental Results and Comparison of

Different Procedures Based on Ranks. *Agron. J.,* **87**: 627-632.
16. Jamshidmoghaddam, M. and Pourdad, S.S. 2013. Genotype×Environment Interactions for Seed Yield in Rainfed Winter Safflower (*Carthamus tinctorius* L.) Multi-Environment Trials in Iran. *Euphytica,* **190**: 357-369.
17. Kang, M. S. 1990. *Genotype-by-Environment Interaction and Plant Breeding*. Louisana State University Agricultural Center, Baton Rouge, LA, USA.
18. Kang, M. 1988. A Rank-Sum Method for Selecting High-Yielding, Stable Corn Genotypes. *Cereal. Res. Commun.,* **16**: 113-115.
19. Kang, M., Gorman, D. and Pham, H. 1991. Application of a Stability Statistic to International Maize Yield Trials. *Theor. Appl. Genet.,* **81**: 162-165.
20. Kasprzak, M. and Rzedzicki, Z. 2008. Application of Everlasting Pea Wholemeal in Extrusion-cooking Technology. *Int. Agrophys.,* **22**: 339-347.
21. Lin, C. S., Binns, M. R. and Lefkovitch, L. P. 1986. Stability Analysis: Where Do We Stand? *Crop. Sci.,* 26: 894-900.
22. Milczak, M., Pedzinski, M., Mnichowska, H., Szwed-Urbas, K. and Rybinski, W. 2001. Creative Breeding of Grasspea (*Lathyrus sativus* L.) in Poland. *Lathyrus Lathyrism Newsletter,* **2**: 85-88.
23. Mohammadi, R., Abdulahi, A., Haghparast, R. and Armion, M. 2007. Interpreting Genotype×Environment Interactions for Durum Wheat Grain Yields using Nonparametric Methods. *Euphytica,* **157**: 239-251.
24. Mohammadi, R. and Amri, A. 2008. Comparison of Parametric and Non-Parametric Methods for Selecting Stable and Adapted Durum Wheat Genotypes in Variable Environments. *Euphytica,* **159**: 419-432.
25. Nassar, R. and Huehn, M. 1987. Studies on Estimation of Phenotypic Stability: Tests of Significance for Nonparametric Measures of Phenotypic Stability. *Biometric.,* **43**: 43-53.
26. Sabaghnia, N., Dehghani, H. and Sabaghpour, S. H. 2006. Nonparametric Methods for Interpreting Genotype×Environment Interaction of Lentil Genotypes. *Crop. Sci.,* **46**: 1100-1106.
27. SAS Institute. 1987. *SAS/STAT User's Guide: Version. 9.1*. SAS Inst Inc., Cary, NC, USA.
28. Scapim, C. A., Pacheco, C. A. P., do Amaral Júnior, A. T., Vieira, R. A., Pinto, R. J. B. and Conrado, T. V. 2010. Correlations between the Stability and Adaptability Statistics of Popcorn Cultivars. *Euphytica,* **174**: 209-218.
29. Segherloo, A. E., Sabaghpour, S. H., Dehghani, H. and Kamrani, M. 2008. Non-parametric Measures of Phenotypic Stability in Chickpea Genotypes (*Cicer arietinum* L.). *Euphytica,* **162**: 221-229.
30. Shukla, G. 1972. Some Statistical Aspects of Partitioning Genotype Environmental Components of Variability. *Hered.,* **29**: 237-245.
31. STATISTICA Statistical Software. 2007. *STATISTICA Data Analysis Software System: Version 8*. Sta Stof Inc., North Melbourne, Australia.
32. Tai, G. C. 1971. Genotypic Stability Analysis and Its Application to Potato Regional Trials. *Crop. Sci.,* **11**: 184-190.
33. Thennarasu, K. 1995. On Certain Non-parametric Procedures for Studying Genotype Environment Interactions and Yield Stability. PhD., PJ School IARI, New Delhi, India.
34. Ward, J. H. 1963. Hierarchical Grouping to Optimize an Objective Function. *J. Am. Stat. Assoc.*, **58**: 236-244.
35. Zobel, B. and Talbert, J. 1984. *Applied Forest Tree Improvement*. Wiley**,** New York City, New York, United States.

تجزیه ناپارامتری پایداری عملکرد لاین‌های پیشرفته خلر (*Lathyrus sativa* L.) در مناطق نیمه گرمسیری

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چکیده

آزمایش‌های چند محیطی نقش مهمی را در انتخاب بهترین ارقام جهت استفاده در مناطق متفاوت نشان می‌دهند. هدف از این مطالعه بررسی پایداری عملکرد دانه و علوفه لاین‌های پیشرفته خلر در مکان‌های مختلف بود. ۱۴ لاین پیشرفته خلر انتخاب شده از مرکز بین المللی ICARDA در سه مکان مختلف در نواحی نیمه گرمسیری ایران طی سه سال زراعی متوالی ارزیابی شدند. به منظور شناسایی لاین‌های پایدار در مجموع نه محیط (ترکیب سال و مکان) از ۱۰ آماره ناپارامتری پایداری استفاده شد. آزمون‌های ناپارامتری (Bredenkamp, Hildebrand and De Kroon and Van der Laan) اثر متقابل ژنوتیپ در محیط بسیار معنی‌داری را نشان دادند، که بیانگر پاسخ متفاوت لاین‌ها به محیط-های متفاوت می‌باشد. عملکرد دانه همبستگی بین عملکرد دانه و آماره‌های $S_i^{(6)}$، NP2، NP3، NP4، مجموع رتبه کنگ (Kang) و رتبه Fox مثبت و معنی‌دار بود. نتایج تجزیه همبستگی و تجزیه به مؤلفه‌های اصلی نشان دادن که آماره‌های ناپارامتری می‌توانند برای انتخاب همزمان لاین‌های برخوردار از عملکرد بالا و پایدار مفید واقع شوند. بر اساس تجزیه خوشه‌ای برای میانگین عملکردهای دانه و علوفه و آماره‌های ناپارامتری لاین شماره ۳ با دارا بودن بالاترین عملکرد دانه و علوفه و رتبه Fox و همچنین کمترین مقادیر دیگر آماره‌های پارامتری می‌تواند به عنوان رقمی پر محصول و پایدار جهت کشت در شرایط دیم برای مناطق نیمه گرمسیری معرفی شود.