Genetic Analysis of Important Traits of Rapeseed Under Normal and Salinity Stress Conditions

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ABSTRACT

Objective: This study aims to evaluate the heritability of salinity tolerance in canola over three years, identifying potential cultivars and crosses that can enhance yield under salinity stress.

Methods: Text

In the first year, 39 rapeseed lines and cultivars were cultivated under normal and salinity irrigation conditions, and grouped based on their salt tolerance. The second year involved crossing five salinity-tolerant cultivars (lines) with three salinity-sensitive cultivars (testers) utilizing a line × tester crossing method. In the third year, the resulting plant materials were grown again under both irrigation conditions to assess performance.

Results: Text

The results indicated significant average heterosis in crosses compared to their parents under both normal and salinity stress conditions. Notably, under normal conditions, all traits except grain yield and 1000-kernel weight exhibited heterosis, while under salinity stress, all traits except pod length and the number of seeds per pod showed similar results. Lines L5 and L2 were identified as strong general combiners for grain yield under normal conditions, while L5 and T2 excelled under salinity stress. The crosses L5 × T3, L5 × T2, and L4 × T2 demonstrated the highest heterosis for grain yield per plant in both conditions, indicating their potential for breeding programs.

Conclusions:

This research provides valuable insights into the heritability of salinity tolerance in canola, highlighting specific lines and crosses that can be utilized in breeding programs to enhance yield under salinity stress, thereby contributing to sustainable agricultural practices in saline-prone areas.

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1 Introduction

Salinity stress is one of the most important environmental stresses that limit agricultural exploitation (Monirifar, 2016). FAO estimates that about 950 million hectares of land around the world are affected by salinity (Flowers and Yeo, 1995, Munns *et al*., 2002). Soils and irrigation water salinity are some of the most important environmental stressors in rapeseed production (Kamkar *et al*., 2004). In Iran, about 24 million hectares of land with different degrees of salinity are affected which are scattered across different climates of the country (Pazira and Sadeghzadeh, 1998). Given that salinity tolerance is a quantitative complicated trait, the choice of breeding method and success for the production of salinity-tolerant cultivars depends on a large extent of understanding the genetic structure of the studied populations (Khattak *et al*., 2001). Understanding the nature of the effects of genes is very important in studying this trait and proposing breeding programs (Muhammad *et al.*, 2014). Thus, recognizing genetic indicators is one of the most important success factors in breeding programs. Also, for accurate estimation of genetic indicators and sufficient knowledge of the genetic structure of the population under study, methods that provide reliable information on gene action should be used (Rozema and Schat, 2013). The salinity tolerance trait is controlled by many genes through complex genetic regulatory networks (Ji *et al*., 2013).

Oilseeds are the second most important source of energy for humans after cereals (Rameeh, 2011). Rapeseed (*Brassica napus* L.) has an important role in oilseed production in Iran and other parts of the world because of its wide adaptation to different diverse climatic conditions (Nemati *et al*., 2012). The Brassica oilseed is the world's third most important source of oils whose production has steadily increased through conventional and modern plant breeding approaches (Bybordi and Tabatabaei, 2009, Sabaghnia et al., 2010b, Sabaghnia *et al*., 2010a). Brassicaceae contains about 3500 species and 350 genera (Amiri-Oghan *et al.*, 2012). Rapeseed seeds contain on average 45-40% of edible oil as well as industrial uses (Nemati *et al*., 2012). Considering the growing population of the world, which will reach 9 billion by 2050, human food security will be the most important challenge facing governments in the future (Yadav, 2010). The main strategy for enhancing production is applying improved varieties based on hybridization (Amiri-Oghana *et al*., 2009). Genetically, the gene composition of some cultivars and genotypes, which appear to be poor in quantitative characteristics, can lead to superior progeny, while the progeny of relatively good cultivars may be undesirable (Farshadfar *et al*., 2013). The production of breeding hybrid varieties requires comprehensive information about the genetic structure of the parents as well as their combining ability in terms of relevant traits (Rameeh, 2012). Estimation of the general combining ability (GCA) and specific combining ability (SCA) of parents can be done using quantitative genetic methods such as diallel crosses, top cross, line × tester, etc. (Nduwumuremyi *et al*., 2013). Line-tester experimental design is a type of top cross method in which several testers are used instead of one tester (Rameeh, 2016). Today, the economic importance of rapeseed as an oil plant has led to higher area cultivation (Singh *et al*., 2008). Given that salinity tolerance is a quantitative complex trait, the choice of breeding method and its success in the production of salinity-tolerant cultivars depends on understanding the genetic structure of the studied populations. Indeed, understanding the nature of the effects of genes in exploring this trait and proposing breeding programs is very important. Thus, one of the most important success factors in breeding programs is the recognition of genetic index and to accurately estimate genetic index and having sufficient knowledge of the genetic structure of the population under study, we must use methods that provide reliable information on gene action (Sofi *et al*., 2006). The purpose of this study was to investigate the inheritance of some important traits of rapeseed such as the number of days to flowering, plant height, number of pods per plant, number of seeds per pod, length of pods, 1000-seed weight, and grain yield in the plant and determining the best common as well as specific combinations plus the amount of heterosis relationship to the superior parent using the line - tester experimental design under

the normal and salinity stress conditions (to evaluate salinity resistance under field conditions).

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2 Materials and Methods

This experiment was performed during three crop years at the Agriculture and Natural Resources Research Centre in Iran in 2017-2020. In the first year, the experiment included the following 39 rapeseed varieties: (1) Talaieh, (2) Sarigol, (3) Zarfam, (4) Zafar, (5) Delgan, (6) Ahmadi, (7) Hyola401, (8) Hyola 60, (9) pp-401015E, (10) T98007, (11) Talaye, (12) SLM046, (13) Geronimo, (14) Modena, (15) Opera, (16) Symbol, (17) KS-11, (18) Colvert, (19) Ks-7, (20) Okapi, (21) Licord, (22) Orient, (23) Option500, (24) H-19, (25) Shiralee, (26) San-14, (27) San-12, (28) SPN178, (29) SPN179, (30) SPN180), (31) SPN182, (32) SPN183, (33) SPN190, (34) SPN184, (35) SPN192, (36) SPN193, (37) RGS003, (38) Dalgon, and (39) SAN56 was carried out in two randomized complete block designs (designs 1 and 2) with three replicates under two irrigation conditions, design 1; 0.631 dSm^{-1} and design 2; 8.7 dSm^{-1}. The seed lines and cultivars investigated in this research were obtained from the Research Seed and Plant Improvement Institute, Oilseed Department, Karaj, Alborz, Iran.

In the second year, five salinity-tolerant cultivars as lines (SPN178, 179, 180, 192, 193) and three salinity-sensitive cultivars as testers (RGS003, DALGON, SAN56), selected from the first-year experiment, were crossed based on the line × tester crossing method.

In the third year, to evaluate hybrids and parents in terms of different traits, 15 F1s along with their parents (A total of 23 genotypes) were sown in two randomized complete block designs (design 1 and design 2) with three replications under two irrigation conditions, normal (NG) and salinity stress (SSG) growth conditions.

The source of water for irrigation experiments (During three years of testing) was two deep wells. The water salinity of the first well is 0.63 dSm^{-1}, which is located in the station. In the NG experiment, the plants were irrigated with normal and natural water (EC = 0.631 dSm^{-1}) which was obtained from this well. The source used water for the SSG experiment was a deep well, which is located about 8 kilometers from the station in which salinity experiments are performed, and its water is transferred to the desired station through piping. The water salinity of this well is (14.7dSm^{-1}), and to reach the desired salinity (8.7 dSm^{-1}), it was mixed with well water (EC= 0.63 dSm^{-1}) in the station and then irrigation was done.

The planting method was the same in all three years of the experiment. Seeds were sown in four 2m rows in each plot with row-to-row intervals of 0.25 m and a density of 80 plants per m^2 for a total area of 2 m^2. The fertilizers were applied at the rates of 100: 90: 50 kg/ha of N: P: K, respectively. The source of the fertilizer and the amount used were respectively: 46% urea fertilizer in the amount of approximately 220 kg/ha, triple super phosphate in the amount of 195 kg/ha, and potassium sulfate in the amount of 100 kg/ha.

In the first year of the experiment at the time of harvesting, two rows on both sides and twenty-five centimeters from both sides of the rows were removed as margins, and all the remaining bushes of the two rows were harvested and their performance was analyzed. In the second year, hybrid seeds formed on separate plants were harvested and similar hybrids were mixed together.

In the third year at the time of harvesting, as in the first year, two rows on both sides and twenty-five centimeters from the top and bottom of the rows were removed as margins, but due to the lack of seeds in some hybrids and the bad vegetation of some hybrids the number of plants in the experimental plots was not equal, therefore, twenty plants were harvested in each plot and the average yield of twenty plants was considered as the seed yield per plant in the calculations.

2-1Trait measurements

2-1-1 Yield and yield components:

In the first year, only the seed yield of the genotypes was measured, and in the third year, in addition to yield, yield components and some physiological traits were also measured. Yield components included plant height (PH), pod length (PL), number of seeds per pod, pods per plant (NSP and NPP), 1000-kernel weight (TKW), and grain yield per plant (GY).

2-1-2 Physiological character

The number of days to 50% flowering and maturity (NDF and NDM) and leaf relative water content (RWC) were quantified. Leaf relative water content measured in fully expanded leaves as described by

(Ravari *et al*., 2016) and calculated according to the followimg Eqn:

$$RWC(\%) = ((FW - DW) \,/\, (TW - DW)) \times 100$$

Where FW, DW, and TW are shoot fresh, dry, and turgid weight, respectively.

2-2 Statistical analyses

All measurements were done on 20 plant samples in each experimental plot. The mean value of the 20 samples was used for data analyses.

Clustering was done using the Euclidean distance method. The cut point was determined based on the maximum difference of the Λ statistic in two consecutive stages. In this way, the value of the statistic when each genotype forms a cluster is equal to zero, and in the last stage when all the figures form a cluster, its index is equal to one and all genotypes are in the same cluster. At the beginning of the grouping, taking into account the hypothetical cut points and the formation of clusters in each stage, the number of clusters was considered as the number of treatments and the number of cultivars or genotypes in each cluster was equal to the number of replications of that treatment. Then multivariate analysis of variance was performed for a completely random design with the number equal to the number of clusters and the statistical value was calculated at that stage and other stages of grouping. Then, the difference of the Λ statistic was calculated for all consecutive steps of multivariate variance. The first point on the graph that had the highest value of Λ statistic based on the following formula (Λ_{k-1} - Λ_k) was selected as the cut-off point and the number of clusters equal to K=4 was selected.

The data of the first year of the experiment including eight salinity tolerance indices for grain yield were subjected to simple analysis of variance based on the criteria of a randomized complete block design based on the following mathematical model: $(X\_ij = \mu + R\_i + T\_j + \varepsilon\_ij)$, where X_{ij} is the numerical value of each observation, μ is the average of the entire population, R_i is total block effects, T_j total treatment effects and ε_{ij} is test error.

In the next step, in order to determine the interaction effects between the type of irrigation water (salinity and normal irrigation) and rapeseed genotypes, combined analysis was performed on the grain yield obtained from two designs (Yp: first environment grain yield (design 1) and Ys: second environment grain yield (design 2) based on the following mathematical model: $(Y\_ijkl = \mu + E\_(i) + (E/R)ij) + T\_(k) + (T \times E)ik + \varepsilon\_(ijkl))$, where Y_{ijkl} is the numerical value of each observation, μ is the average of the entire population, E_i is the effect of environment (type of irrigation), $(E / R)_{ij}$ is the effect of replication in the environment, T_k is the effect of treatment, $(T \times E)_{ij}$ is the interactions between treatment and environment and ε_{ijkl} is the random error.

Also, the data of the third year of the experiment, eight traits measured in the selected parents and their hybrids, including plant height, number of days to flowering, number of days to maturity, length of spikelet per plant, number of seeds per spikelet, number of spikelet per plant, 1,000-kernel weight and yield per plant were subjected to simple line × tester analysis of variance based on the the Line × Tester mathematical model. Also combined variance analysis was done based on the α- lattice design. Pearson's simple linear correlation between yield in stress and non-stress environments and yield indicators was performed

The line × tester model (Kempthorne, 1957) was used to estimate general combining ability (GCA) and specific combining ability effects (SCA). The values of broad(h_B^2)and narrow sense (h_N^2)heritability, gene action, the average degree of dominance, and stress tolerance index (STI) were calculated from the following equations, respectively:

$$h_B^2 = \left(\frac{\mathrm{VA} + \mathrm{VD}}{\mathrm{VA} + \mathrm{VD} + \mathrm{M'e}}\right) \times 100$$

$$h_N^2 = \left(\frac{\mathrm{VA}}{\mathrm{VA} + \mathrm{VD} + \mathrm{M'e}}\right) \times 100$$

$$\text{Gene action} = \frac{MS_{gca}}{MS_{sca}}$$

$$\text{Degree of dominance} = \sqrt{\frac{2\mathrm{VD}}{\mathrm{VA}}}$$

Where MSgca and MSsca are general and specific combining ability mean square, respectively. Additive and dominance variance VA and VD denote, respectively. M_e is the mean of error squares, r is the replication number and M'e reflects Me divided by r, Y_s, and Y_p are the grain yield under stress and non-

stress conditions, $(\bar{Y}_p)^2$ is the mean yield over all genotypes under non-stress conditions.

The eight stress tolerance indices were measured using the following expressions:

$MP = \left(Y_s + Y_p\right)/2$ (Bouslama and Schapaugh Jr, 1984),

$TOL = \left(Y_p - y_s\right)$ (Hossain et al., 1990), $GMP = \sqrt{Y_s \times Y_p}$

(Shen et al., 2006), $SSI = \frac{1-\frac{Y_s}{Y_p}}{\frac{\bar{Y}_s}{\bar{Y}_p}}$ (Fischer and Maurer, 1978),

$YI = \frac{Y_s}{Y_p}$ (Gavuzzi et al., 1997), $YSI = \frac{Y_s}{\bar{Y}_p}$

(Bouslama and Schapaugh Jr, 1984), $STI = \frac{Y_s \times Y_p}{\bar{Y}_P^2}$ (Gavuzzi

et al., 1997), $HM = \frac{2(Y_s \times Y_p)}{(Y_s + Y_p)}$ (Rosielle and Hamblin, 1981).

3 Results and Discussion

First-year results:

Shapiro and Wilks (Shapiro and Wilk, 1965) tests were performed for the normality of the distribution of residuals along with another hypothesis of analysis of variance for simple and combined analysis of variance (Bartlett's method.

The results of a simple analysis of variance for grain yield, in the first year, under both conditions showed significant differences among genotypes (Table 1). Significant effects of genotype in simple ANOVA for YP, YS (Table 1), and of genotype and environment × genotype interaction in combined ANOVA for grain yield (Table 2), non-significant correlation coefficients between Yp and Ys (Table 3), showed the different response of genotypes in the two environments and the existence of genetic variation among varieties. This indicates that selection based on high YP does not always lead to high YS. These findings are concordant with the results of (Van Ginkel *et al*., 1998) and (Bchini *et al*., 2011). The results of analysis of variance of tolerance indices showed that there is a significant difference between the studied cultivars in terms of indices (Table 1). Among the studied indices, four indices, MP, GMP, HM, and STI, had a positive and significant correlation with Yp and Ys and selection based on high values of these mentioned indices will lead to select varieties with high yield in both environments. This means that these indices are suitable for isolating varieties belonging to group A (genotypes with similar good performance in both stress and non-stress environments) from group B (genotypes with good performance only in non-stress environments), C (genotypes with good performance only in stress environments) and D (genotypes with weak performance in both environments) (Fernandez, 1992). This result agrees with the findings of (Bchini et al., 2011), (Izaddoost *et al*., 2013), and (Ravari *et al.*, 2016). (Hefny *et al*., 2013) found these four indicators to be appropriate in a study of sorghum.

Cluster analysis was performed based on Euclidean distance measurement and non-weighted paired group method using arithmetic average (UPGMA). The matrix of the means of Euclidean distance used as input to the cluster analysis was averaged over the three replicates per genotype. The results of cluster analysis (Figure 1) based on the selection indices show that these genotypes are classified into four groups: susceptible (S), mid-susceptible (MS), mid-tolerant (MT), and tolerant (T).

Second Year of the experiment

Based on these results, five salinity-tolerant genotypes (SPN178, 179, 180, 192, 193), and three salinity-sensitive genotypes (RGS003, DALGON, SAN56) were selected for the second-year crosses. In the second year of the experiment, based on the crosses performed (line × tester method) in the field and greenhouse, 15 hybrids were obtained.

Table1. Mean squares of Yp, Ys, and eight tolerance indices for grain yield

SOV	df	Yp	Ys	TOL	MP	GMP	SSI	YI	YSI	STI	HM
R	2	13.7	3.01	4.05	7.34	6.33	0.001	0.006	0.008	0.034	5.48
G	38	165.6**	46.8**	38.0**	96.7**	88.2**	0.01**	0.004**	0.12**	0.48**	80**
E	76	0.309	0.075	0.373	0.099	0.08	0.001	0.0001	0.001	0.001	0.08

**: Significance at the level of α= 0.01, GMP: Geometric Mean Product; HM: Harmonic Mean; MP: Mean Product; STI: Stress Tolerance Index; SSI: Stress Susceptibility Index; TOL: Tolerance; YSI: Yield Stability Index; YI: Yield Index; Y_P: Non-stressed Grain Yield; Y_S: Stressed grain

Table2. Mean squares of 39 rapeseed varieties for grain yield in combined analysis of variance for Yp: first environment grain yield (design 1) and Ys: second environment grain yield (design 2)

Source of variation	df	Mean square
Environment(E)	1	7008.82**
Error 1 (R/E)	4	8.359
Genotype (G)	38	193.53**
Environment × Genotype (E × G)	38	19.01**
Error 2 (R × G/E)	152	0.192

**: Significance at the level of α= 0.01

Table3. Correlation coefficients among Yp, Ys, and salinity tolerance indices

	Yp	Ys	MP	GMP	TOL	STI	SSI	HM	YSI	YI
Yp	1									
Ys	0.19ns	1								
MP	0.64**	0.77**	1							
GMP	0.59**	0.85**	0.98**	1						
TOL	0.78**	-0.82**	-0.77**	-0.69**	1					
STI	0.64**	0.75**	0.87**	0.77**	-0.78**	1				
SSI	0.53**	-0.73**	-0.73**	-0.87**	0.83**	-0.75**	1			
HM	0.67**	0.82**	0.59**	0.81**	-0.7**	0.73**	-0.73**	1		
YSI	-0.64**	0.82**	0.58**	0.64**	-0.79**	0.68**	-0.91**	0.89**	1	
YI	0.09ns	0.99**	0.87**	0.95**	-0.89**	0.96**	-0.82**	0.94**	0.95**	1

ns, *, **: non-significant, significance at the level of α= 0.01, respectively. GMP: Geometric Mean Product; HM: Harmonic Mean; MP: Mean Product; STI: Stress Tolerance Index; SSI: Stress Susceptibility Index; TOL: Tolerance; YSI: Yield Stability Index; YI: Yield Index; Y_P: Non-stressed Grain Yield; Y_S: Stressed grain Yield

3-1 Third-year results

3-1-1 Line × Tester analysis:

The results of simple line × tester analysis of variance revealed that the difference between genotypes for all studied traits was significant at the level of significance α=0.01 and the environment had a different effect on genotypes (Table 4). Combined analysis variance of line × tester based on alpha lattice design indicated that there was a significant difference between environments for all studied traits (Table 5). Thus, the difference in the mean of genotype

traits from one environment to another was significant at the level of significance α=0.01. Also, the effects of normal irrigation and water salinity stress on the studied traits were different. These results are consistent with the results of the initial analysis. Under the two conditions, there was a significant difference between the treatments in most of the studied traits indicating sufficient genetic diversity between genotypes and their reciprocal cross combinations, and therefore line × tester analysis of variance can be performed (Table 4).

Table4. Line × tester analysis for different traits in rapeseed genotypes in normal and salinity stress conditions

Condition irrigation	Mean squares									
	S.O.V	df	PH (cm)	NDF	NDM	PL (cm)	NSP	NPP	TKW	GY (gr per plant)
Normal	R	2	292.71*	106.97 ns	57.43 ns	0.01ns	8.88*	8.88*	0.01	5.77
	Tr	22	516.20**	134.7 **	122.30**	1.43**	20.67**	20.67**	0.19**	59.73**
	P	7	913.21**	178.66**	180.66**	1.74**	28.95**	28.95**	0.28**	103.91**
	P vs C	1	937.41**	280.14*	442.90**	8.66**	65.42**	65.42**	0.10 ns	9.75ns
	Cr	14	287.61**	102.40*	70.22*	0.76**	13.33**	13.33**	0.15**	41.21**
	L	4	637.92**	203.52*	138.7 **	1.22**	24.59**	24.59**	0.14**	93.60**
	T	2	314.29*	114.42	36.36	0.91	19.29**	19.29**	0.16*	37.2**
	L × t	8	105.79	48.84	44.44	0.49	6.21*	6.21*	0.16**	16.02**
	E	44	89.62	41.56	35.63	0.30	2. 66	2.66	0.03	3.56
Stress	R	2	207.13**	10.49	12.01	0.12*	0.93	7.48	0.00	139.66**
	Tr	22	845.11**	162.57**	68.29**	0.54**	10.07**	3331.5**	0.20**	11.53**
	P	7	1097.18**	161.14**	93.52**	0.82**	13.90**	3714.5**	0.33**	15.61**
	P vs C	1	4314.56**	283.46**	84.61*	0.02	1.65	1318.4**	0.08**	5.46
	Cr	14	471.26**	154.64**	54.51**	0.43**	8.75**	3283.9**	0.15**	9.92**
	L	4	586.02**	305.58**	104.13	0.45**	14.30**	5687.2**	0.13**	14.16**
	T	2	1063.36**	20.42*	0.47	1.06**	12.87**	4337.3**	0.34**	24.16**
	L × t	8	265.86**	112.73**	43.22*	0.27**	4.95**	1818.8**	0.11**	4.25
	E	44	37.16	4.87	16.67	0.03	0.58	64.86	0.01	2.73

*and **: significant at 5% and 1% of probability levels, respectively; PH: plant height; NDF: number of days to 50% flowering; NDM: number of days to maturity; PL; pod length; NSP: number of seeds per pod; NPP: number of pods per plant; TKW: 1000-kernel weight; GY: green yield; SOV: Sources of Variation

Table5. Combined variance analysis (normal and salinity stress conditions) based on α-lattice design

		Mean squares							
S.O.V	df	PH (cm)	NDF	NDM	PL (cm)	NSP	NPP	TKW	GY (gr per plant)
E	1	41288.1**	536.12**	7289.9**	34.30**	292.7**	145113.22**	73.15**	3413.8**
R(E)	4	249.92**	58.73*	34.72	0.07	4.91*	7.44	0.00	72.71**
Tr	22	1235.76**	224.93**	153.63**	1.53**	27.23**	3535.98**	0.35**	58.34**
P	7	1907.47**	247.88**	221.05**	2.41**	39.71**	5207.83**	0.55**	96.89**
P vs C	1	4637.09**	563.59**	457.34**	4.80**	43.94**	82.52	0.00	14.90*
C	14	656.95**	188.16**	98.23**	0.86**	19.80**	2946.73**	0.27**	42.04**
L	4	1138.40**	479.94**	220.59**	1.21**	35.34**	6291.71**	0.24**	83.62**
T	2	1264.96**	24.01	14.34	1.74**	30.01**	3741.11**	0.47**	60.21**

L * T	8	264.23**	83.3**	58.01*	0.46**	9.47**	1075.65**	0.24**	16.71**
Tr * E	22	125.56*	73.09**	36.97	0.43**	3.50**	1555.75**	0.04**	13.00**
P * E	7	102.93	91.93**	53.13	0.15	3.14	904.59**	0.06**	22.62**
(P vs C) * E	1	614.88**	0.001	70.17	3.89**	23.1**	1786.40**	0.18**	0.31
C * E	14	101.93	68.89**	26.54	0.33*	2.28	1864.86**	0.03	9.09**
L * E	4	85.54	29.16	22.26	0.46**	3.54	2162.88**	0.03	24.13**
T * E	2	112.74	110.83**	22.48	0.24	2.14	1446.93**	0.04	1.16
L * T * E	8	107.41	78.26**	29.64	0.29	1.69	1820.34**	0.02	3.55
Er	88	63.39	23.22	26.15	0.17	1.62	73.92	0.02	3.15
CV (%)		6.06	6.23	3.78	6.75	6.83	4.29	3.96	11.69

*and **: significant at 5% and 1% of probability levels, respectively; PH: plant height; NDF: number of days to 50% flowering; NDM: number of days to maturity; PL; pod length; NSP: number of seeds per pod; NPP: number of pods per plant; TKW: 1000-kernel weight; GY: green yield

Table 6. Estimation of broad and narrow sense heritability, gene action, and average degree of dominance in normal and salinity stress conditions

Traits	**Broad sense heritability (%)**		**Narrow sense heritability (%)**		**Average degree of dominance**		**Gene action**	
	normal	**stress**	**normal**	**Stress**	**normal**	**stress**	**normal**	**Stress**
PH	90.92	97.79	89.28	84.19	0.19	0.57	27.24	3.10
NDF	86.74	99.07	84.41	78.49	0.23	0.72	18.18	1.91
NDM	100.00	100.00	88.75	88.75	0.50	0.50	3.94	3.94
PL	85.92	97.74	77.06	82.39	0.48	0.61	4.35	2.68
NSP	94.29	98.16	86.67	84.25	0.42	0.57	5.68	3.03
NPP	94.29	99.47	86.67	85.05	0.42	0.58	5.68	2.95
TKW	90.88	98.80	58.74	78.91	1.05	0.71	0.91	1.98
GY	97.75	91.91	89.89	87.43	0.42	0.32	5.72	9.74

The significant difference of parents vs. crosses indicated significant average heterosis under both normal and stress conditions; in the normal condition for all the traits except grain yield and 1000-kernel weight and in the salinity stress condition, for all the traits except, the pod length and the number of seeds per Pod. The mean degree of dominance for all traits under both conditions except, 1000- kernel weight under normal conditions was less than one, suggesting the existence of incomplete dominance (Table 6). Analysis of hybrids into their components based on line × tester analysis revealed that the effect of lines, testers, as well as the effect of line × testers, was significant in terms of the studied traits and the lines had different reactions in combination with different testers.

The ratio of the mean squares of general combining ability (MSgca) to the mean squares of specific combining ability (MSsca) was estimated with estimates greater than one unit indicating that the additive action of genes has a greater role in their genetic control (Table 6). Note that the role of non-additive effects of genes in determining important rapeseed traits has been reported in previous studies (Kang *et al*., 2013, Rameeh, 2010). These results also revealed that the amount of broad heritability of traits ranged from 85.92% for the trait pod length up to 100% for the days to maturity. High values and low range of change broad heritability indicated that genetic variance is more important than environmental variance (Table 6). The lowest amount of narrow sense heritability for the trait 1000-kernel weight was 58.74%, while the highest amount of narrow sense heritability was related to the grain yield (89.89%) under normal irrigation conditions. Heterosis is a common phenomenon in agriculture and an important strategy to increase yield and ensure world food security (Shen *et al*., 2006). The amounts of heterosis compared to the superior parent in both culture conditions, normal and salinity, are outlined in Table 7. Under normal and salinity conditions, crosses, L2 × T3, L2 × T1, and L2 × T2 had the most significant negative heterosis to reduce plant height. As the grain yield is the most important breeding trait of rapeseed, so crosses L5 × T3, L5 × T2, L4 × T2 in both normal and salinity conditions were the compounds with the

highest amounts of heterosis compared to the superior parent in terms of these traits, and had the potential to be used in breeding programs to enhance the yield. The use of heterosis to boost the quantitative and qualitative yield of rapeseed has been previously reported (Amiri-Oghan *et al*., 2012).

The results of general combining ability estimation showed that the difference between the lines in terms of general combining ability is significant in both irrigation conditions (Table 8). Reducing plant height in rapeseed with increasing stock lodging tolerance of cultivars and improving harvest index is one of the important goals of rapeseed breeding., in normal conditions lines; "SPN180" with negative and significant general combining ability values to reduce plant height, also in salinity stress conditions, tester " RGS003" are considered as the desirable cultivar. For important traits such as grain yield, under the normal condition lines, "SPN193" and "SPN179" also in stress salinity condition lines; "SPN193" and Dalgon had high general combining were the best general combiner parent.

Table 7. General combining ability estimates of rapeseed lines × testers for studied traits in normal and salinity stress conditions

Conditions		Tester			Lines				
	Traits	**T1**	**T2**	**T3**	**L1**	**L2**	**L3**	**L4**	**L5**
Normal	PH	-3.89	5.04	-1.16	-4.60	6.18*	-7.60**	-5.60*	11.62**
	NDF	-1.82	3.18	-1.36	-1.47	4.53**	-4.58**	-4.02*	5.53**
	NDM	-0.62	0.74	-0.12	-2.16	1.90	-2.77*	-2.27	5.29**
	PL	-0.14	0.28	-0.15	-031*	-0.25	0.48**	-0.24	0.32*
	NSP	-1.11	1.16	-0.04	-1.38**	0.62	-0.82	-1.04*	2.62**
	NPP	-1.11	1.16	-0.04	-1.38**	-0.62	-0.82	-1.04*	2.62**
	TKW	-0.06	0.12**	-0.06	0.08	0.13**	-0.17**	-0.08	0.04
	GY	-1.76*	1.28	0.48	5.47**	2.95**	-3.26**	-1.09*	3.87**
Salinity	PH	-5.88**	10.17**	-4.28	-1.17	-4.71	-8.06**	-1.95**	6.4**
	NDF	2.53	0.2	-2.73	-2.62**	3.4**	-1.51	-2.95	3.6**
	NDM	-0.62	0.74	-0.12	-2.16	1.90	-2.77	-2.27	5.29**
	PL	-0.29**	0.24**	0.05	-0.15**	-0.09	-0.09	-0.06	0.40**
	NSP	-1.07**	0.60*	0.47	-1.64**	0.02	-0.53**	0.36	1.80**
	NPP	19.58**	8.49**	11.09**	-4.07	17.93**	-10.73**	-33.73**	30.60**
	TKW	-0.01	0.16**	-0.14**	-0.05*	0.18**	-0.16**	0.00	0.03
	GY	-1.46*	0.83	0.63	2.08*	0.65	-0.12	-1.23**	1.78**

*and **: significant at 5% and 1% of probability levels, respectively

Table 8. Specific combining ability estimates of rapeseed crosses for studied traits in normal and stress conditions

Conditions	Line × tester	PH	DNF	DNM	PL	NSP	NPP	TKW	GY
Normal	L1 × T1	1.33	5.22**	3.62**	0.11	-0.89	-0.89	0.09	1.13
	L2 × T1	2.89	4	0.40	-0.22	0.11	0.11*	-0.03	0.25
	L3 × T1	5.00	-4	1.07	0.59**	0.89	0.89	-0.27**	1.51*
	L4 × T1	-11.0**	-6.66**	-4.27**	0.07	-0.89	-0.89	0.24**	-1.59*
	L5 × T1	1.78	-1.44	-0.82	-0.55**	0.78	0.78	-0.03	-1.29*
	L1 × T2	-2.27	-3.44	-3.41**	-0.24	-1.16*	-1.16*	-0.09	-1.99**
	L2 × T2	-2.71	0.33	2.37	0.16	0.18	0.18	-0.06	-0.14
	L3 × T2	-4.60	-4.66	-0.47	-0.40*	-1.04	-1.04	-0.01	-2.56**

	L4 × T2	7.40*	4.67*	3.03*	0.22	2.1**	2.18**	0.15*	3.99**
	L5 × T2	2.18	3.11	-1.52	0.26	-0.16	-0.16	0.01	0.71
	L1 × T3	0.93	-1.77	-0.21	0.13	2.4**	2.04**	0.00	0.87**
	L2 × T3	-0.18	-4.33	-2.77	0.06	-0.29	-0.29	0.09	-0.11*
	L3 × T3	-0.40	8.67	0.60	-0.20	0.16	0.16	0.28**	1.05
	L4 × T3	3.60	2	1.23	-0.28	-1.29*	-1.29*	-0.39**	-2.39**
	L5 × T3	-3.96	-4.6	2.34	0.29	-0.62	-0.62	0.02	0.58
	SE(sca)	5.47	3.72	2.09	0.32	0.94	0.94	0.11	1.09
Salinity	L1 × T1	-102**	0.022	3.62**	0.24**	0.18	-6.53*	0.02	0.19
	L2 × T1	5.89**	-4.42**	0.40	0.15*	0.8**	-9.87**	0.03	0.21
	L3 × T1	3.33	4.9**	1.07	0.02	0.7**	7.13*	-0.15**	-0.21
	L4 × T1	-10.4**	-4.64**	-4.27**	-0.38**	-1.8**	10.4**	0.08**	-0.11
	L5 × T1	11.44	-4.1	0.82	-0.04	0.07	-1.20	0.03	-0.07
	L1 × T2	1.04	-1.31	-3.41**	-0.42**	-0.8**	-24.2**	-0.08**	-1.25*
	L2 × T2	-9.17**	9.2**	2.37	0.16*	0.51	-5.60*	-0.16**	-0.48
	L3 × T2	-2.60*	-7.42**	-0.47	-0.11	-1.2**	28.7**	-0.06*	-0.77
	L4 × T2	9.48**	-0.64*	3.03*	0.36**	1.8**	10.4**	0.21**	1.72**
	L5 × T2	-1.3	0.13	-1.52	0.00	-0.27	-8.60**	0.09**	0.77
	L1 × T3	9.17**	1.28	-0.21	0.18**	0.64*	30.8**	0.07*	1.06
	L2 × T3	-3.2	-4.8**	-2.77	-0.31**	-1.3**	15.4**	0.13**	0.27
	L3 × T3	-0.6	2.5	0.60	0.09	0.53*	-35.2**	0.21**	0.98
	L4 × T3	0.95	5.2**	1.23	0.02	-0.02	-20.8**	-0.29**	-1.61**
	L5 × T3	-12.8**	-4.26**	2.34	0.03	0.20	9.80**	-0.12**	-0.70
	SE(sca)	3.52	1.27	2.09	0.11	0.44	4.65	0.04	0.95

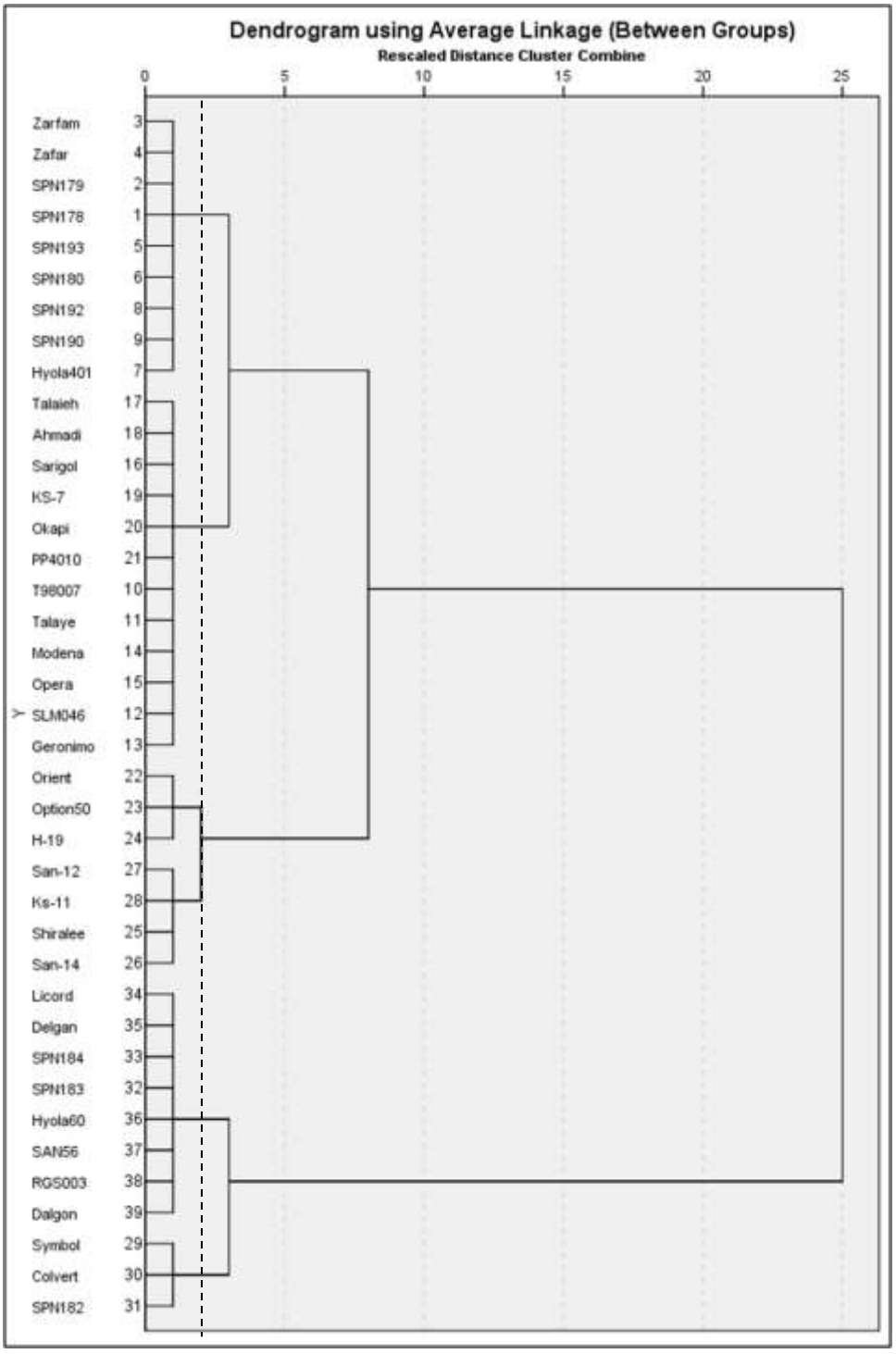
*and **: significant at 5% and 1% of probability levels, respectively; PH: plant height; NDF: number of days to 50% flowering; NDM: number of days to maturity; PL; pod length; NSP: number of seeds per pod; NPP: number of pods per plant; TKW: 1000-kernel weight; GY: green yield

4. Conclusions

Under normal conditions for the trait number of days to 50% flowering, L4 × T1, and for salinity stress, cross L3 × T2 were desirable crosses. For both normal and salinity conditions, cross L4 × T1 was a good combination based on the appearance of the number of days to maturity. For the length of the pod, the combination including L3 × T1 in normal conditions and combination L4 × T2 in the salinity condition were good specific combiners. Based on the appearance of the number of seeds per pod under both conditions the cross containing L4 × T1 was considered a suitable combination for improving this trait. The combination of L1 × T3 under both normal and salinity conditions constituted the best specific combiners in terms of the number of pods per plant. Some previous studies indicated that the 1000-kernel weight trait is more influenced by genetic factors than by other yield components and environmental factors have minor effects on determining this trait (Ding *et al.*, 2012, Shi *et al.*, 2009).

Grain yield has a complex inheritance and is influenced by many environmental factors. Grain yield is a multifaceted trait that is characterized by many other traits with positive and negative effects on yield (Tuncturk and Ciftci, 2007). Since one of the breeding purposes is boosting the yield, the selection of SPN192 and Dalgon parents as transferring parents under normal and salinity conditions can be useful in breeding programs; Under normal and salinity conditions hybrid L4 × T2 with the highest values of specific combining ability in the terms of grain yield trait had the best hybrids and are recommended.

Fig 1. Dendrogram on the basis of the UPGMA method for tolerance indices of grain yield in 39 rapeseed varieties



Declarations

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