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Scientific Paper

Genetic analysis of morpho-physiological characteristics of sunflower under stress and Non-stress drought conditions

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Abstract

Studying the general and specific combinability of the lines under the favorable and limited irrigational conditions is essential to utilize the parents in creating new hybrid varieties. Furthermore, the type of genetic trends can be effective in determining the suitable corrective methods and in forecasting the rate of genetic development stemmed from the choicemade.To achieve the desired goal five CMS lines and four restorer lines (tester) were crossed in a line × tester scheme in order to analyze general and specific combining abilities and gene effects on sunflower for some agronomic and physiologic traits. Hybrids were evaluated in a randomized complete block design, with three replications under optimum and limited water conditions in the Agriculture Research Station, Khoy, Iran during 2017 and 2018 growing seasons. The results of simple analysis of variance in optimum and limited conditions for all traits including 1000 seed weight, %RWC, grain yield, oil yield were significant for all traits. In addition, the results of combined analysis of variance for different traits in optimal conditions showed that the mean square, except for the %RWC traits in the limited conditions for plant height, head diameter, number of seeds per head, 1000 seed weight, proline, catalase enzyme, superoxide dismutase enzyme , Chlorophyll a, chlorophyll b, grain yield and oil yield were significant. results of data variance analysis indicated that the general combinability for lines and tester was significant, as well as the specific combining ability (line×tester) for most traits. Based on the AGK30 and AGK44 combining ability, the RGHK25, RGHK50 and (RGHK50 ×RGHK56) and the (RGHK56 × AGK44) tester were identified as the most suitable line for the hybrid test in both conditions.

Keywords: line, tester,sunflower, General combining ability, specific combining ability.

Análise genética de características morfo-fisiológicas de girassol sob estresse e condições de seca não-estresse

Resumo

Estudar a combinabilidade geral e específica das linhas sob condições de irrigação favoráveis e limitadas é essencial para utilizar os pais na criação de novas variedades híbridas. Além disso, o tipo de tendências genéticas pode ser eficaz na determinação dos métodos corretivos adequados e na previsão da taxa de desenvolvimento genético derivada da escolha. Para alcançar o objetivo desejado, cinco linhas CMS e quatro linhas restauradoras (testador) foram cruzadas em uma linha × Esquema testador para analisar habilidades de combinação gerais e específicas e efeitos gênicos no girassol para algumas características agronômicas e fisiológicas. Os híbridos foram avaliados em um delineamento de blocos completos casualizados, com três repetições sob condições ótimas e limitadas de água nas estações de Agricultura, Khoy, Irã, durante 2017 e 2018. Os resultados da análise simples de variância em condições ótimas e limitadas para todas as características, incluindo o peso de 1000 sementes, % de massa de videira, rendimento de grãos, produção de óleo, foram significativos para todas as características. Além disso, os resultados da análise de variância combinada para diferentes características em condições ótimas mostraram que a média quadrática, exceto para as características de% RWC nas condições limitadas de altura de planta, diâmetro de cabeça, número de sementes por cabeça, peso de 1.000 sementes, prolina enzima catalase, enzima superóxido dismutase, clorofila a,

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clorofila b, rendimento de grãos e produção de óleo foram significativos. Os resultados da análise de variância dos dados indicaram que a combinação geral para linhas e testador foi significativa, bem como a capacidade de combinação específica (linha × testador) para a maioria das características. Com base na capacidade de combinação AGK30 e AGK44, os testadores RGHK25, RGHK50 e (RGHK50 × RGHK56) e (RGHK56 × AGK44) foram identificados como a linha mais adequada para o teste híbrido em ambas as condições.

Palavras-chave: linha, testador, girassol, capacidade geral de combinação, capacidade de combinação específica.

Análisis genético de las características morfofisiológicas del girasol bajo estrés y condiciones de sequía sin estrés

Resumen

El estudio de la combinabilidad general y específica de las líneas en condiciones favorables y limitadas de irrigación es esencial para utilizar a los padres en la creación de nuevas variedades híbridas. Además, el tipo de tendencias genéticas puede ser eficaz para determinar los métodos correctivos adecuados y para pronosticar la tasa de desarrollo genético derivado de la elección. Para lograr el objetivo deseado, se cruzaron cinco líneas CMS y cuatro líneas restauradoras (probador) en un esquema línea x probador para analizar capacidades de combinación generales y específicas y efectos de genes en girasol para algunos rasgos agronómicos y fisiológicos. Los híbridos se evaluaron en un diseño de bloques completos al azar, con tres repeticiones en condiciones de agua óptimas y limitadas en la Estación de Investigación Agrícola, Khoy, Irán durante las temporadas de crecimiento de 2017 y 2018. Los resultados del análisis simple de la varianza en condiciones óptimas y limitadas para todas las características, incluido el peso de 1000 semillas,% RWC, rendimiento de grano, rendimiento de aceite fueron significativos para todas las características. Además, los resultados del análisis combinado de varianza para diferentes características en condiciones óptimas mostraron que el cuadrado medio, excepto el% de características de RWC en las condiciones limitadas para altura de planta, diámetro de la cabeza, número de semillas por cabeza, 1000 semillas de peso, prolina La enzima catalasa, la enzima superóxido dismutasa, la clorofila A, la clorofila B, el rendimiento de grano y el rendimiento de aceite fueron significativos. los resultados del análisis de varianza de los datos indicaron que la combinabilidad general para las líneas y el probador fue significativa, así como la capacidad de combinación específica (línea x probador) para la mayoría de los rasgos. Sobre la base de la capacidad de combinación AGK30 y AGK44, el probador RGHK25, RGHK50 y (RGHK50 × RGHK56) y (RGHK56 × AGK44) se identificaron como la línea más adecuada para la prueba híbrida en ambas condiciones.

Palabras clave: línea, probador, girasol, habilidad de combinación general, habilidad de combinación específica.

Introduction

Sunflower (*Helianthus annuus* L) is an important oilseed crop with high quality of edible oil in the world. Sunflower hybrids are preferred by farmers because of their uniformity, high yield performance, better quality, and resistance against disease. Identification of superior parents for hybridization is an important step in plant breeding. Combining ability of parental lines should be estimated to find the best hybrid combinations. Furthermore, estimation of gene effects could be done by analyzing combining ability values based on F1 mean values (Mijic et al., 2008). Combining ability of inbred lines could be estimated with various methods, such as top cross; line × tester analysis, in which several testers are used, is an extension of this method (Kempthorne.,1957). Ashok et al. (2000) analyzed four male sterile lines and 10 testers and found additive gene effects for seed yield. However, Naroui Rad (2013); Rehman, (2012); Tan (2010) and Asish (2009) analyzed gene effects of inbred lines using line × tester method; they showed that non-additive gene effects were more important for seed yield. Also, observed the after that gene component was more important than the additive component in managing seed yield in sunflower. In addition, reported the importance of dominant genes for seed yield in this crop. In general, most

of the experiments carried out by breeders indicate the importance of non-additive gene effect for seed yield in sunflower. Therefore, production of hybrid varieties in this crop is justified due to the existence of non-additive type of gene action for seed yield. Not only additive gene action was reported to have significant impact on plant height (Bhat et al., 2000) and head diameter (Gvozdenovic et al., 2005), but also non-additive gene effects have been reported to play crucial role in regard with plant height (Gvozdenovic et al. 2005; Gouri Shankar et al., 2007; Andarkhor et al.,2012). Karasu et al. (2010) showed significant general combining ability for plant height, seed number per head, 1000 seed weight and seed yield. Hity (1992) indicated the importance of both additive and non-additive gene action in controlling 1000 seed weight and oil percentage, while Ortis et al. (2005) and Skoric et al. (2007) stressed the preponderance of additive type of gene effects and non-additive gene effects for oil percentage. Though sunflower hybrids are high yielders, the G × E interaction influences their performance (Balu et al. 2007). Sharma et al. (2003) analyzed 60 hybrids in two environments and reported a significant line × tester × environment effects for head diameter, seed yield, days to the inauguration of flowering, plant height and 1000 seed weight. The main objective of the present study was to determine the combining ability in sunflower, using a line × tester method with five lines and four testers.

Material and methods

Experimental site

The Khoy experimental station at 44° and 58 minutes in northern latitude and 38 degrees and 33 minutes in east and elevation from sea level 1103 M. The site has cold and dry climates. Minimum, average and maximum annual temperatures are 30, 12.5 and 42 degrees Celsius, and the average annual rainfall in this region is 292.6 mm.

In order, to determine some of the physical and chemical properties related to the soil of the site before the implementation of the design, the soil composite sample was prepared on average from 0-30 cm depth using the Oger unit from the four parts of the experimental farmhouse. In order to determine the nutrients was sent to Soil Science Laboratory of Soil and Water Research Department of Agricultural and Natural Resources Research Center of West Azarbaijan, the result of which is presented in Table 1.

Table 1. Results of chemical and physical soil analysis to the experimental area.

OC	Total N	Fe available	K available	P available	Soil saturation	pH	Clay	Silt	sand
0.5 -1 0.87	0.1-0.007 0.008	4 -5 4.9	300-400 361	10-15 9.4	42-46 46	7-7.5 7.5	26-28 27	42-45 36	25-27 18

Plant materials used in this study consisted of five CMS lines (AGK2, AGK30, AGK44, AGK110, AGK260) and four male restorer lines (RGHK25, RGHK46, RGHK50, RGHK56), developed in Khoy field station, West Azarbaijan province, Iran. CMS lines were crossed with testers in a line × tester method. The progenies were evaluated during 2017 and 2018 growing seasons, using randomized complete block design with three replications in each year. Each plot consisted of two 5m long rows. The row to row spacing was 65 cm and the plants spaced at 25 cm within the rows. The following traits were measured during growing season: plant height, head diameter, seed weight per head, seed number per head, RWC, % proline, CAT, SOD, Chlorophyll a, Chlorophyll b, oil content, seed yield and 1000 seed weight. Samples of photos taken in conditions of stress and non- stress (Fig. 2 and 3) that three replications were non-strees and three replicates of stress on the ground.

Figure 2. non-strees

Figure 3. Strees

Data for hybrids were subjected to "Line ×Tester" analysis (Singh and Chaudhury, 2001) to estimate general combining ability (GCA), specific combining ability (SCA), and their respective variance components. The estimates of general combining ability and specific combining effects of parents and hybrids were obtained through the following equations: Estimation of GCA effects

Lines: GCA= (Xi../fr)- (X…/fmr)

Testers: GCA= (X.j./mr)- (X…/fmr)

where f= number of CMS lines (female parent)

t = number of testers (male parent)

r = number of replications

Xi = Total number of the F1 resulting from crossing ith lines with all the testers

X.j = Total number of all crosses of jth tester with all the lines

X = Total number of all crosses

Estimation of SCA effects

SCA=Sij-(Xij/r)-(Xi../fr)-(X.j./mr)+(X…/fmr)

where Xij = Total number of F1 resulting from crossing ith lines with jth testers

Estimates of GCA and SCA variances were obtained by expected values of mean squares, assuming lines and testers as fixed and years as random factors. Additive genetic variances for lines and testers was calculated through the following formula:

$$\sigma^2_{GCA} = \frac{1+F}{4}\sigma^2_A$$

where, assuming that the coefficient of inbreeding is unity, σ^2A and σ^2GCA are additive genetic variance of lines and testers. Dominance variance (σ^2D), narrow sense heritability (h^2N) and broad sense heritability (h2B) were obtained using the following formulae:

$$\sigma^2_{SCA} = \frac{(1+F)^2}{2}\sigma^2_D$$

$$h^2_B = \frac{\sigma^2_A + \sigma^2_D}{\sigma^2_A + \sigma^2_D + \sigma^2_e}$$

$$h^2_N = \frac{\sigma^2_A}{\sigma^2_A + \sigma^2_D + \sigma^2_e}$$

where σ^2A represents additive variance, σ^2e error variance, and σ^2e represents the number of replications and number of years.

Plant height

In order to measure plant height, at the physiological maturity stage, the length of ten plants was measured from the ground surface., and then the mean of them was recorded in cm.

Head diameter

The largest diameter per 10 plants was measured at the physiological maturity stage and the mean values were recorded in cm.

Seed number head

To measure the number of seeds per plant, the number of grains per head was manually counted and the mean number of samples was calculated (as a result of the total number of grains filled in the samples harvested).

1000 seed weight

To this end, hundred-seeded samples of each plot were counted and then weighed after being completely dried in the oven. Finally, the average weight was multiplied by 10 and recorded as 1000 seed weight.

Seed yield

Grain yield was estimated in kilograms per unit area based on the dimensions of planting. After delete of half a meter, the effect of the margin on the parties' plots, Ten plants were randomly selected and after thresh and placing in the oven at 48 ° C for 24 hours, so that their moisture content reached 13-14% Accuracy was weighted by precision scale and weighed in kilograms per unit area.

Relative Water Content(RWC)

To determine the relative water content (RWC) in fresh weight laboratory, the samples were distilled in room temperature and in darkness for 24 hours, followed by inflammation. In the next step, the samples were dried in an oven at 70 ° C for 72 hours. The RWC was derived from the following formula:

% RWC= [Fw-dw] / [sw-dw] × 100

Fw: Fresh weight
Dw: Dry weight
Sw: Saturation weight

leaf area index

At the end of flowering from each line, five crops were selected randomly. By measuring the length and width of all leaves, the total leaf area of selected plants was calculated from the relation (fixed coefficient 0.68 × width of leaf × leaf length = leaf area) Rao and Saran (1991)) then, the average leaf area of a plant is calculated and multiplied by the density of plant per square meter, the leaf area index is obtained.

Measurement of chlorophyll

The Arnon (1967) method was used to measure chlorophyll content as follows:

1. The amount of 1 g of vegetable matter was poured into Chinese moss, then it was crushed with liquid nitrogen and then firmly squeezed.

2. 20ml of acetone 80% added to the sample, then placed in a centrifuge machine at a speed of

6000 rpm for 10 minutes. The extracted extracted from the centrifuge was transferred to a glass balloon.

3. A sample of the inside of the balloon was poured into a cuvette spectrophotometer and then read at 653 nm for chlorophyll a and 645 nm for chlorophyll b and 470 for carotenoids separately by spectrophotometer.

4. Finally, using the following formulas, the amount of chlorophyll a and b in mg / g is the fresh weight of the sample.

Chlorophyll a = (19.3 * A663 - 0.86 * A645) V/100W

Chlorophyll b = (19.3 * A645 - 3.6 * A663) V/100W

Measurement of catalase activity

To measure the activity of catalase enzyme by Luhova et al. (2007), a spectrophotometer was used at 240 nm wavelength in 30 seconds. In addition, 20mM sodium phosphate buffer with pH 7 and 20µl hydrogen peroxide 30% were used as an electron receptor. The activity of catalase was expressed in mg / g of protein.

Measurement of Superoxide Dismutase Enzyme Activity: Measurement of Superoxide Dismutase Enzyme Activity Based on Beauchamp and Fridovich (1971). The reaction solution in the final volume of one milliliter was measured to measure the activity of the superoxide dismutase enzyme, which contained 835 µl 50 mM sodium phosphate buffer at pH = 8 and 33 µL of ethylenediamine tetraacetic acid 3 mM, 33 µl nitroblutterazolium 75 mM, 33 Zantin microliter 3 mM, 33 µl diluted solution of zantin oxidase enzyme and 33 µl of enzyme extract. The absorbance changes of the reaction solution to the control were measured by spectrophotometer at 560 nm and the activity of the superoxide dismutase enzyme was expressed in terms of unit per milligram of protein.

Proline measurement method

Bates (1973) method was used to measure proline using the following methods: 0.5gr of herbal material was poured into a tub mortar, followed by adding 10 ml of a sulfosalicylic acid solution of 3%. The resulting solution was then placed in ice. The tubes were centrifuged at 15000 rpm for 10 to 15 minutes at 4 ° C to remove excess material from the solution. Instead of centrifuges, a glass funnel and filter paper can be used to filter the samples. The amount of 2 ml of filtered extrudate was poured into a new tubing and added 2 ml of acid mixture and 2 ml of glacial acetic acid and then mixed well. At the same time, 2 ml of standard solutions of 0, 4, 8, 12, 16 and 20 mg / l of proline were poured into new tubes and added 2 ml of naivinidrin and 2 ml of glacial acetic acid and then mixed well. . The specimens were heated in a hot bath for 1 hour and then placed in an ice bath. The amount of 4 ml of toluene was added to the solution and stirred for 20 seconds with a vortex machine. The proline content of the toluene-phase solution was poured into the cuvette of the spectrophotometer and proline was read at 520 nm.

Statistical Analysis

The MSTATC and SPSS22 software was used to analyze the data.

Results and discussion

The results of variance analysis of different traits in stress and non stress conditions indicated that mean square for all traits was significant.

Mean squares of lines were significant for plant height, head diameter, number of seeds per head, 1000 seed weight, proline, catalase, superoxide dismutase, chlorophyll a, chlorophyll b, grain yield and oil yield. Significance of the mentioned traits indicates that there is a significant difference in the general combining ability of lines for these traits. The general combining ability of the lines indicates the incremental effects of genes According to the table of analysis of variance (Table 3), the mean squares of testers for seed number per head, proline, catalase, superoxide dismutase, chlorophyll a, chlorophyll b were significant both under normal conditions and under stress conditions, indicating the significance of the ability The general combinability of the tester is for these traits. Significance of general combining ability of the testers is indicative of the incremental effects of genes. Also, according to (Table 3), the mean squares of the line × tester for plant height, Head diameter, number of seeds per head, 1000 grain weight, proline, catalase, superoxide dismutase, chlorophyll a, chlorophyll b, grain yield and oil yield were significant in both normal and stress conditions. Significance of these traits showed significant capacity The hybrid hybridity for these traits is. Which indicates the dominant effects of genes.

In the study of general combining ability of

lines (Table 4), the best lines for AGHK2 and AGHK30 plant height were the highest and most suitable for general and negative traits in normal and tropical conditions. Negative and significant admixture for height The plant is considered a favorable correctional trait. Also, the AGK44 line had the most positive and significant general combining ability under normal conditions. In examining other AGHK30 lines traits, the number of seeds per head, 1000 grain weight, grain yield and oil yield had the highest The ability of general combining ability was positive and significant under stress condition but in conditions Normal AGHK2 has the most positive and significant combining ability with a thousand seed weight attribute, which can be selected from the hybrid under normal conditions as the preferred line.. . Also, AGHK44 line for relative water content and superoxide dismutase has the most positive and significant general combining ability. For proline, chlorophyll a, chlorophyll b line, AGHK110 had the most positive and significant general combining ability under stress and normal conditions. Line Above all, it also has a line showing the incremental effects of genes.

In examining the testers for plant height, the RGKH50 and RGKH25 testers had the most general negative and significant general combining ability in normal conditions, which can be corrected for the trait. Also, the RGHK25 titer for head and head diameter traits had the highest positive and significant aggregate Under stress and normal conditions, the RGHK56 tester has the most positive and significant positive effects on stress and normal conditions. For proline traits, the RGHK46 tester has the most positive and significant combining ability under stress conditions. RGHK50 and RGHK56 testers have catalase and superoxide dismutase traits Highest combining ability Positive and meaningful genotypes were in stress. Also, RGHK56 tester has the most positive and significant general combining ability with chlorophyll a in stress conditions Vernal and RGHK25 had the highest positive and significant positive correlation with chlorophyll B in stress conditions and RGHK56 testers under normal conditions. RGHK50 and RGHK25 testers also had the highest positive and significant general combining ability with grain yield and oil yield under normal and stress conditions (Table 5).

In the study of hybrids, the RGHK46 × AGHK260 and RGHK46 × AGHK30 hybrid had the most negative and significant combinability capability for plant height, which can be identified as superior hybrids. RGHK25 × AGHK260 has the most positive and significant positive combining ability for head diameter in stress conditions, which is a good hybrid for selection and RGHK50 × AGHK30 hybrid had the most positive and significant positive combining ability for normal head diameter and for the number of seeds per head, the RGHK50 × AGHK260 hybrid had the highest positive and significant combining ability, which is a good hybrid in stress conditions also The RGHK25 × AGHK30 hybrid has the highest specific combining ability in normal conditions. For the trait 1000 seed weight, RGHK56 × AGHK44 hybrid had the highest positive and significant combining ability under stress and hybrid conditions, RGHK25 × AGHK44 had the highest positive and significant positive specific combining ability in normal conditions. For the relative water content of the hybrid RGHK50 × RGK30 highest positive and significant positive combining ability in stress conditions and RGHK56 × RGK2 hybrids had the highest combining ability in normal conditions. In examining the proline hybrid, RGHK56 × AGHK2 hybrid had the most positive and significant positive combining ability in stress conditions, which is a good hybrid in stress conditions. Also, RGHK46 × AGHK260 hybrid had the most positive and significant positive combining ability for catalase in stress and normal conditions. In the study of RGHK50 × AGHK260 hybrid superoxide dismutase, it has the highest potential for positive and significant positive combining ability under stress and normal conditions. Also, the RGHK46 × AGHK30 hybrid had the highest positive and significant combining ability for chlorophyll a under stress conditions and RGHK46 × AGHK260 in normal conditions. In examining the trait of chlorophyll b, RGHK50 × RGK44 hybrid had the most positive and significant positive combining ability in stress conditions and RGHK46 × AGHK30 hybrids with the highest combining ability in normal conditions. Also, RGHK56 × AGHK44 hybrids had the highest positive and significant combining ability for seed yield and oil yield under stress conditions and RGHK50 × AGHK30 and RGHK25 × RGK44 hybrids respectively for seed yield and oil yield, with the highest degree of combining ability in There were normal conditions that indicate the non-additive

effects of genes, or those of dominant effects.

As it was indicated, dominance effects (line × tester interaction) were significant for plant height, seed number head, %Proline and Chlorophyll b under water sterss. Therefore, these characters were controlled mainly by dominant gene action. (Gaffari, 2006), also, reported the role of both additive and dominant gene action in the inheritance of 1000 seed weight. However, for stem diameter, days to the inauguration of flowering, 1000 seed weight and days to maturity of GCA/SCA ratios were larger than unity showing the importance of additive gene effects (not proved by obtained data). Bhat et al. (2000) also observed that additive type of gene action was of paramount importance for plant height and days to 50% of flowering in sunflower. Furthermore, Karasu et al. (2010) reported significant general combining ability for plant height, 1000 seed weight and seed number per head. On the other hand, Tan (2010), Gvozdenovic et al. (2005) and Gouri Shankar et al. (2007) indicated the significance of non-additive gene effects for plant height in sunflower. Similar to the findings of the present study, although Volotovich (2008) reported the importance of both additive and non-additive gene effects in controlling the oil percentage, other studies have indicated the role of either additive gene action (Ortis et al., 2005) or non-additive gene action. Although Tan (2010) stated that seed yield is controlled by both types of gene action, according to his results, non-additive component was more important than the additive component. Rehman et al. (2012) also indicated the importance of dominant genes in controlling the seed yield in sunflower. Non-additive type of gene action for seed yield justifies the production of hybrid varieties in this crop. However, different results obtained by different researchers may be attributed to different genetic material and the environmental conditions in these studies. Our results are also restricted to the lines and testers under study. Average degree of dominance for most of the traits ranged from incomplete dominance to over-dominance, suggesting the existence of non-additive gene action for these agronomic traits in sunflower (Table 2). Khan et al. (2008) also reported high heritability estimates for 1000 seed weight, seed number per head, oil percent and seed yield in sunflower using line×tester analysis. Senevirathne et al. (2004) and (Skoric et al., 2007) found similar results for other plant material in sunflower.

Average degree of dominance for most of the traits ranged from incomplete dominance to over-dominance, suggesting the existence of non-additive gene action for these agronomic traits in sunflower (Table 2). Gaffari et al. (2011) also reported the preponderance of dominance effects for this trait. As shown in Table 2, broad sense heritability ranged from 0.03 (Plant height) to 6.99 (Seed number head). Most of the traits under investigation showed medium narrow sense heritability. Medium to high narrow sense heritability estimates over two years indicate the efficiency of selection for these traits. Khan et al. (2008) also reported high heritability estimates for 1000 seed weight, seed number head, oil percent and seed yield in sunflower using line×tester analysis. Senevirathne et al. (2004), Sawargaonkar et al. (2008) found similar results for other plant material in sunflower.

Conclusion

In the study of lines, in terms of general combining ability under favorable and limited conditions, the AGHK30 and AGHK44 lines were the best. In the test of the testers in terms of general combining ability, the best testers were RGHK25 and RGHK50 testers. Hybrid RGHK56 × AGHK44 hybrid was considered the best hybrid in optimum and limited conditions in studying hybrid ability for hybridisation. The hybrids were examined for the first time and their hybrids were resistant and have optimum performance under optimum and limited conditions.. It is suggested that this test be tested for stability analysis in different areas and the best hybrid for different regions.

Table 1. Summary of combined analysis of variance for sunflower characters in the line×tester cross under optimum and limited water conditions.

SV	df	Plant height (cm)		Head diameter (cm)		Seed number head		1000 seed weight (gr)	
		NS	S	S	NS	S	NS	S	NS
Year	1	19.20^{ns}	147.40^{ns}	1.50^{ns}	45.53**	11623.00^{ns}	27694.40^{ns}	50.83^{ns}	151.65^{ns}
R/Y	4	1005.11	447.28	4.11	14.12	5851.79	11889.13	23.53	67.41
Line	4	321.90*	2378.02**	11.06**	5.42**	54145.74**	432882.33**	3.19**	31.05**
L×Y	4	103.42^{ns}	125.22^{ns}	2.85**	0.40^{ns}	663.88^{ns}	295.24^{ns}	0.70^{ns}	0.79^{ns}
Tester	3	742.96**	350.49^{ns}	8.57**	2.14^{ns}	6257.80**	110909.47**	2.05^{ns}	11.24**
T × Y	3	93.55^{ns}	21.23^{ns}	2.06**	0.90^{ns}	193.98^{ns}	160.69^{ns}	1.58^{ns}	0.62^{ns}
L × T	12	149.76**	967.37**	3.95**	12.77**	25796.53**	143861.35**	2.99**	10.12**
L×T×Y	12	16.81^{ns}	45.96^{ns}	0.76^{ns}	0.48^{ns}	1062.63^{ns}	461.28^{ns}	1.08^{ns}	0.48^{ns}
Error	76	68.66	133.23	0.42	0.79	7923.11	448.73	1.77	1.45
CV %		6.34	6.71	4.48	7.67	19.44	3.86	4.07	2.82

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 1. Continued. Summary of combined analysis of variance for sunflower characters in the line×tester cross under optimum and limited water conditions

SV	df	% RWC		%Proline		CAT (Unit per mg protein)		SOD (Unit per mg protein)	
		S	NS	S	NS	S	NS	S	NS
Year	1	0.43^{ns}	290.87^{ns}	20.83^{ns}	.033^{ns}	3.35^{ns}	14.58^{ns}	141.41*	7.96^{ns}
R/Y	4	34.10	103.43	89.63	23.98	117.92	132.61	34.78	87.06
Line	4	11.87^{ns}	49.16**	935.24**	219.63**	92.61**	386.26**	824.87**	228.72**
L×Y	4	8.30^{ns}	5.76^{ns}	15.66^{ns}	4.72^{ns}	9.83^{ns}	5.19^{ns}	0.04^{ns}	1.68^{ns}
Tester	3	17.09^{ns}	80.62**	297.83**	9.93*	588.75**	109.76**	207.78**	64.07**
T × Y	3	20.33^{ns}	7.09^{ns}	4.23^{ns}	2.90^{ns}	0.70^{ns}	2.34^{ns}	0.59^{ns}	0.31^{ns}
L × T	12	10.64^{ns}	42.58**	437.31**	113.48**	590.35**	603.15**	1189.34**	349.34**
L×T×Y	12	13.35^{ns}	12.59**	18.06^{ns}	11.64^{ns}	4.73^{ns}	6.77^{ns}	0.34^{ns}	2.34*
Error	76	12.16	11.85	12.44	7.39	8.67	15.05	2.72	1.09
CV %		5.41	4.35	6.06	8.84	3.83	6.65	3.10	3.63

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress. RWC=Relative Water content, CAT=catalase, SOD= superoxide dismutase

Table 1. Continued. Summary of combined analysis of variance for sunflower characters in the line×tester cross under optimum and limited water conditions

SV	df	Chlorophylla (mg g^{-1})		Chlorophyllb (mg g^{-1})		Oil yield (Kg ha^{-1})		Seed yield (Kg ha^{-1})	
		S	NS	S	NS	S	NS	S	NS
Year	1	3.76^{ns}	12.66^{ns}	7.25**	0.07^{ns}	54502.88^{ns}	177356.78^{ns}	183300.83^{ns}	545940.30^{ns}
R/Y	4	3.29	9.41	0.22	1.65	22602.84	71202.17	84758.93	242684.40
Line	4	0.92**	5.64**	8.35**	6.89**	3069.07*	32650.45**	11535.90**	111801.45**
L×Y	4	0.15^{ns}	0.47*	0.02^{ns}	0.25^{ns}	929.18^{ns}	6295.66^{ns}	2525.25^{ns}	2858.55^{ns}
Tester	3	0.47**	4.85**	2.23**	3.69*	2050.69^{ns}	15960.13**	7352.47^{ns}	40494.70**
T × Y	3	0.06^{ns}	0.78**	0.24^{ns}	0.64^{ns}	1344.69^{ns}	3367.11^{ns}	5702.87^{ns}	2258.70^{ns}
L × T	12	1.28^{ns}	2.00**	1.21**	1.89**	3347.31**	9831.77**	10802.83**	36461.45**
L×T×Y	12	0.09^{ns}	0.58**	0.08^{ns}	0.37^{ns}	1116.21^{ns}	1463.52^{ns}	3917.73^{ns}	1742.95^{ns}
Error	76	0.17	0.16	0.09	0.24	2094.63	3156.10	6393.60	5234.71
CV %		7.8	3.42	9.75	7.81	4.54	3.97	4.07	2.82

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 2. Broad and narrow sense heritability estimates on an entry mean basis and degree of dominance of sunflower characters in the line × tester analysis under optimum and limited water conditions.

	Broad sense heritability		**Narrow sense heritability**		**Degree of dominance**	
	S	NS	S	NS	S	NS
Plant height	0.30	0.553	0.077	0.039	2.41	5.13
Seed number head	0.35	0.98	0.013	0.117	6.99	3.83
1000 seed weight	0.14	0.562	-	0.08	-	3.46
%Proline	0.85	0.7	0.05	0.014	5.37	96.98
Chlorophyll a	0.18	0.65	-	0.148	-	2.60
Chlorophyll b	0.74	0.578	0.25	0.136	1.97	2.54
Oil yield	0.14	0.354	-	0.068	-	2.87
Seed yield	0.14	0.56	-	0.08	-	3.46

Table 3. General combining ability for CMS(cytoplasmic male sterility) lines of sunflower in the line × tester analysis.

CMS	Plant height (cm)		Head diameter (cm)		Seed number head		1000 seed weight (gr)	
	S	NS	S	NS	S	NS	S	NS
AGHK2	-4.48**	-8.48**	1.96**	0.48^{ns}	-132.12**	408.58**	-0.66**	2.70**
AGHK30	-9.73**	-26.07**	1.76**	0.07^{ns}	135.47**	-0.92^{ns}	1.18**	-0.92**
AGHK44	9.35**	-1.73^{ns}	0.21*	0.68^{ns}	10.05^{ns}	-110.08**	-0.53*	1.49**
AGHK110	1.93ns	8.85^{ns}	-0.44**	0.43^{ns}	0.38^{ns}	31.83^{ns}	-0.1^{ns}	-3.22**
AGHK260	2.93*	27.43**	0.44**	-1.65^{ns}	-13.78^{ns}	-329.42**	0.11^{ns}	-0.05^{ns}
SE (GCA)	2.39	3.33	0.18	0.48	25.69	6.11	0.38	0.34
SE(gi-gj)	3.38	4.71	0.26	0.68	36.33	8.64	0.54	0.49

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 3. Continued. Summary of combined analysis of variance for sunflower characters in the line×tester cross under optimum and limited water conditions

CMS	% RWC		%Proline		CAT		SOD	
	S	NS	S	NS	S	NS	S	NS
AGHK2	-0.65^{ns}	-4.65**	-18.78**	-9.20**	-6.12**	-12.71**	-7.60**	0.42^{ns}
AGHK30	1.05*	1.88**	14.13**	6.63**	1.64**	3.54**	0.41^{ns}	-1.05**
AGHK44	2.32**	-0.88^{ns}	4.38**	2.55**	-015^{ns}	3.46**	20.12**	10.22**
AGHK110	0.95^{ns}	2.14**	4.95**	-2.37**	4.65**	8.10**	-7.76**	-4.56**
AGHK260	0.32^{ns}	1.50**	5.52**	2.38**	-0.02^{ns}	-2.40**	-5.18**	-5.04**
SE (GCA)	1	0.99	1.01	0.78	0.85	1.11	0.47	0.3
SE(gi-gj)	1.42	1.40	1.44	1.11	1.20	1.58	0.67	0.42

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress. CMS= cytoplasmic male sterility, RWC=Relative Water content, CAT=catalase, SOD= superoxide dismutase

Table 3. Continued. Summary of combined analysis of variance for sunflower characters in the line×tester cross under optimum and limited water conditions

CMS	Chlorophyll a		Chlorophyll b		Oil yield		Seed yield	
	S	NS	S	NS	S	NS	S	NS
AGHK2	-0.36**	-0.85**	-0.76**	-0.93**	-23.25**	73.24**	39.37**	161.80**
AGHK30	0.04^{ns}	-0.25^{ns}	-0.66**	-0.21**	34.78**	-47.01**	70.80**	-55.20**
AGHK44	-0.33**	-0.91**	-0.64**	-0.71**	-12.91^{ns}	68.88**	-32.37*	89.30**
AGHK110	0.61**	1.27**	2.04**	1.79**	-5.82^{ns}	-97.73**	-5.87^{ns}	-193.20**
AGHK260	0.03^{ns}	0.74**	0.03**	0.06^{ns}	7.89^{ns}	2.61^{ns}	6.80^{ns}	-2.70^{ns}
SE (GCA)	0.12	0.11	0.089^{ns}	0.14	13.21	16.21	23.08	20.88
SE(gi-gj)	0.17	0.16	0.12	0.2	18.68	22.93	32.64	29.53

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress. CMS= cytoplasmic male sterility

Table 4. General combining ability for restorer lines of sunflower in the line × tester analysis (average of two years) under optimum and limited water conditions.

Restorer	Plant height (cm)		Head diameter(cm)		Seed number head		1000 seed weight (gr)	
	S	NS	S	NS	S	NS	S	NS
RGHK25	10.43**	-2.25**	1.11**	0.73**	38.45**	145.18**	0.17^{ns}	-0.01^{ns}
RGHK46	6.43**	0.55**	-1.45**	-0.51*	-27.42*	50.78**	-0.45*	-0.32*
RGHK50	9.97**	-7.32**	0.03^{ns}	0.04^{ns}	-15.82^{ns}	-125.42**	0.66**	1.64**
RGHK56	6.90**	9.02**	0.3**	-0.25^{ns}	4.78^{ns}	-70.55**	-0.39*	-1.31**
SE(GCA)	2.13	2.98	0.16	0.43	22.98	5.46	0.34	0.31
SE(gi-gj)	3.02	4.21	0.23	0.61	32.50	7.73	0.48	0.44
SE(gi-gj)	0.17	0.16	0.12	0.2	18.68	22.93	32.64	29.53

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 4. Continued. General combining ability for restorer lines of sunflower in the line × tester analysis (average of two years) under optimum and limited water conditions.

Restorer	% RWC		%Proline		CAT (Unit per mg protein)		SOD (Unit per mg protein)	
	S	NS	S	NS	S	NS	S	NS
RGHK25	-0.86^{ns}	-3.82**	-4.10**	-0.53^{ns}	-4.63**	-0.37^{ns}	-4.44**	-2.29**
RGHK46	0.94*	-1.13*	9.37**	-0.20^{ns}	1.29**	-2.56**	-3.72**	-1.06**
RGHK50	-1.65**	3.92**	-3.23**	-0.93*	11.84**	5.52**	1.24**	-0.94**
RGHK56	1.58**	1.03*	-2.03**	1.67**	-8.50**	-2.59**	6.93**	4.29**
SE(GCA)	0.9	0.88	0.91	0.7	0.76	1	0.42	0.27
SE(gi-gj)	1.27	1.25	1.28	0.99	1.07	1.41	0.6	0.38
SE(gi-gj)	0.17	0.16	0.12	0.2	18.68	22.93	32.64	29.53

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress. . RWC=Relative Water content, CAT=catalase, SOD= superoxide dismutase

Table 4. Continued. General combining ability for restorer lines of sunflower in the line × tester analysis (average of two years) under optimum and limited water conditions.

Restorer	Chlorophyll a (mg g^{-1})		Chlorophyll b (mg g^{-1})		Oil yield (kg ha^{-1})		Seed yield (kg ha^{-1})	
	S	NS	S	NS	S	NS	S	NS
RGHK25	0.28**	0.03**	0.57**	0.3**	4.88^{ns}	-9.63^{ns}	10.43^{ns}	-0.5^{ns}
RGHK46	-0.1^{ns}	-0.99**	-0.74**	-1.05**	-6.23^{ns}	-24.90**	-27.03*	-19.30*
RGHK50	-0.3**	-0.03^{ns}	0.05^{ns}	0.26**	20.13**	67.44**	39.63**	98.30**
RGHK56	0.12*	0.98**	0.13**	0.49**	-18.77**	-32.92**	-23.03*	-78.50**
SE(GCA)	0.1	0.1	0.08	0.12	11.81	14.50	20.64	18.68
SE(gi-gj)	0.15	0.14	0.11	0.17	16.71	20.51	29.19	26.41
SE(gi-gj)	0.17	0.16	0.12	0.2	18.68	22.93	32.64	29.53

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 5. Specific combining ability for 20 hybrids of sunflower in the line × tester analysis (average of two years)

CMS	Restorer	Plant height		Head diameter		Seed number head		1000 weight seed	
		S	NS	S	NS	S	NS	S	NS
AGHK2	RGHK25	9.15**	-1.58^{ns}	-1.81**	1.85**	0.05^{ns}	112.15**	1.97**	-2.75**
AGHK30	RGHK25	6.48**	5.62^{ns}	-0.26^{ns}	0.99**	-16.75^{ns}	324.55**	-0.56^{ns}	0.30^{ns}
AGHK44	RGHK25	-8.12**	-16.48**	2.43**	-2.05**	-22.67^{ns}	-339.25*	0.05^{ns}	3.97**
AGHK110	RGHK25	-7.52**	-20.52**	-0.36^{ns}	-1.79**	39.38^{ns}	-97.45**	-1.46**	-1.52**
AGHK260	RGHK25	4.40^{ns}	-25.67**	2.59**	0.64^{ns}	103.80**	253.65**	-0.99*	2.37**
AGHK2	RGHK46	5.07*	10.87**	-0.63**	0.62^{ns}	-86.33**	-340.95*	0.64^{ns}	-0.72*
AGHK30	RGHK46	-11.87**	4.40^{ns}	-1.02**	-1.53**	-29.93^{ns}	-135.08*	-0.81*	-0.31^{ns}
AGHK44	RGHK46	2.40^{ns}	10.40**	-0.94**	0.27^{ns}	12.47^{ns}	222.38**	1.17**	-1.33**
AGHK110	RGHK46	0.98^{ns}	-24.67**	-1.12**	0.15^{ns}	-89.45**	203.48**	0.53^{ns}	0.66^{ns}
AGHK260	RGHK46	-18.02**	-10.47**	0.74**	-4.97**	-14.25^{ns}	-416.45*	-1.05*	-3.33**
AGHK2	RGHK50	6.05*	-2.60^{ns}	-1.26**	1.46**	-9.85^{ns}	160.75**	-0.23^{ns}	-0.99**
AGHK30	RGHK50	10.98**	37.73**	1.64**	3.36**	113.55**	52.22**	-0.75^{ns}	3.66**
AGHK44	RGHK50	-9.60**	21.08**	0.68**	0.2^{ns}	62.22*	-376.77*	-0.32^{ns}	1.73**
AGHK110	RGHK50	4.07^{ns}	-13.05**	0.28^{ns}	0.56^{ns}	123.58**	402.63**	-1.29**	1.38**
AGHK260	RGHK50	5.47*	-21.52**	-1.65**	-2.12**	201.82**	125.17**	1.76**	-2.55**
AGHK2	RGHK56	0.07^{ns}	-13.48**	0.69**	1.36*	-140.45*	-151.03*	-0.16^{ns}	-0.57^{ns}
AGHK30	RGHK56	-4.93*	30.83**	-0.33^{ns}	-2.85**	-76.62**	-192.52*	-1.19**	-2.01**
AGHK44	RGHK56	2.40^{ns}	7.03*	-0.13^{ns}	1.80**	240.92**	30.22**	2.27**	2.37**
AGHK110	RGHK56	8.47**	3.23^{ns}	1.49**	4.23**	-139.35*	188.42**	-0.78*	-0.12^{ns}
AGHK260	RGHK56	-5.93*	-41.10**	-1.03**	-3.19**	-24.95^{ns}	-26.12**	-0.3^{ns}	-0.24^{ns}
SE(SCA)		4.78	6.66	0.37	0.96	51.39	12.23	0.76	0.69

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 5. Continued. Specific combining ability for 20 hybrids of sunflower in the line × tester analysis (average of two years)

CMS	Restorer	% RWC		%Proline		CAT		SOD	
		S	NS	S	NS	S	NS	S	NS
AGHK2	RGHK25	038^{ns}	-1.71^{ns}	-0.15^{ns}	-0.8^{ns}	-3.96**	-13.78**	-29.71**	-16.84**
AGHK30	RGHK25	1.44^{ns}	3.53**	-0.95^{ns}	0.87^{ns}	-9.63**	-21.63**	13.54**	10.12**
AGHK44	RGHK25	1.02^{ns}	-6.79**	-9.35**	-5.40**	2.92**	6.08**	-22.43**	-13.11**
AGHK110	RGHK25	-2.85**	4.98**	10.45**	5.33**	16.51**	29.33**	38.60**	19.83**
AGHK260	RGHK25	0.25^{ns}	1.93^{ns}	-33.40**	-17.30**	22.21**	14.70**	37.51**	20.25**
AGHK2	RGHK46	-1.21^{ns}	-1.95^{ns}	18.47**	2.70**	-10.69**	-13.60**	-18.40**	-13.40**
AGHK30	RGHK46	-1.55^{ns}	4.40**	12.73**	8.77**	12.68**	12.06**	-12.33**	-6.31**
AGHK44	RGHK46	2.51*	-4.37**	2.20*	5.83**	-24.20**	-13.16**	-6.78**	-0.53^{ns}
AGHK110	RGHK46	-2.37**	3**	7.68**	2.78**	-7.37**	-12.75**	8.45**	2.83**
AGHK260	RGHK46	-3.33**	5.48**	-2.78*	3.12**	26.13**	29.77**	-18.55**	-7.39**
AGHK2	RGHK50	0.62^{ns}	-1.14^{ns}	2.15*	0.52ns	-1^{ns}	9.48**	-2.52**	-2.21**
AGHK30	RGHK50	5.08**	-7.34**	-7.05**	-6.42**	-17.76**	-26.50**	12.62**	6.77**
AGHK44	RGHK50	1.66^{ns}	-1.56^{ns}	10.02**	5.70**	-6.13**	-1.89^{ns}	-29.92**	-14.05**
AGHK110	RGHK50	1.94^{ns}	-2.46*	-15.12**	-6.30**	17.90**	2.39*	8.90**	3.45**
AGHK260	RGHK50	-0.41^{ns}	-2.38*	-11.85**	-5.90**	-21.80**	-9.22**	39.18**	20.43**
AGHK2	RGHK56	-3.19**	6.40**	16.95**	6.50**	10.03**	8.72**	-18.16**	-9.82**
AGHK30	RGHK56	0.08^{ns}	-1.65^{ns}	15.85**	9.62**	-4.75**	13.72**	13.66**	7.81**
AGHK44	RGHK56	1.15^{ns}	-4.456**	0.38ns	-0.38^{ns}	-23.71**	3.06*	14.51**	7.22**
AGHK110	RGHK56	0.31^{ns}	5.92**	6.32**	2.02*	13.03**	-18.40**	-1.89**	1.21**
AGHK260	RGHK56	-1.54^{ns}	0.33^{ns}	-22.55**	-11.25**	15.42**	1.62^{ns}	-26.29**	-16.25**
SE(SCA)		2.01	1.98	2.03	1.57	1.7	2.23	0.95	0.6

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non Stress

Table 5. Continued. Specific combining ability for 20 hybrids of sunflower in the line × tester analysis (average of two years)

CMS	Restorer	Chlorophyll a		Chlorophyll b		Oil yield		Seed yield	
		S	NS	S	NS	S	NS	S	NS
AGHK2	RGHK25	1.49**	0.36**	0.40**	0.04^{ns}	64.13**	-83.33**	118.57**	-165**
AGHK30	RGHK25	-1.03**	-0.39**	0.11^{ns}	0.16^{ns}	-10.67^{ns}	21.35^{ns}	-33.97^{ns}	17.80^{ns}
AGHK44	RGHK25	-0.59**	-0.77**	-0.42**	-0.70**	10.25^{ns}	103.89**	-3.37^{ns}	238.20**
AGHK110	RGHK25	0.13^{ns}	0.80**	0.7**	0.49**	-63.71**	-41.91^{ns}	-87.97**	-91**
AGHK260	RGHK25	-0.51**	0.34**	-1.02**	-0.37**	-30.93**	82.07**	-59.60*	142**
AGHK2	RGHK46	-0.42**	-1.25**	-0.12^{ns}	-1.73**	34.98**	-25.81^{ns}	37.87^{ns}	-43.20*
AGHK30	RGHK46	0.91**	0.75**	0.51**	1.86**	-35.90**	-26.50^{ns}	-48.80*	-18.80^{ns}
AGHK44	RGHK46	0.02^{ns}	0.16**	0.64**	0.23^{ns}	31.85**	-29.76^{ns}	70.53**	-80**
AGHK110	RGHK46	-0.37**	-0.88**	-0.64**	-0.77**	17.12^{ns}	10.07^{ns}	31.53^{ns}	39.50^{ns}
AGHK260	RGHK46	1**	1.67**	0.74**	1.58**	-43.59**	-112.44**	-62.97*	-199.70**
AGHK2	RGHK50	-1.1**	-0.97**	0.12^{ns}	-1.01**	6.37^{ns}	1.89^{ns}	-13.63^{ns}	-59.30**
AGHK30	RGHK50	0.47**	0.18^{ns}	-0.22**	0.19^{ns}	-20.09^{ns}	100.48**	45.03^{ns}	291.50**
AGHK44	RGHK50	-1.08**	-1.34**	1.60**	0.16^{ns}	-19.10^{ns}	69.88**	-18.93^{ns}	104**
AGHK110	RGHK50	0.76**	-0.08^{ns}	-0.53**	-0.03^{ns}	-39.87**	13.98^{ns}	-77.47**	82.80**
AGHK260	RGHK50	0.31*	1.50**	0.44**	0.81**	57.32**	-45.82**	105.87**	-152.80**
AGHK2	RGHK56	0.01^{ns}	-0.08^{ns}	-1.52**	-0.94**	1.64^{ns}	-38.04^{ns}	-9.47^{ns}	-34^{ns}
AGHK30	RGHK56	0.47**	1.52**	0.46**	0.93**	-31.22**	-78.68**	-71.60**	-120.50**
AGHK44	RGHK56	-0.31*	0.05^{ns}	-0.20*	0.01^{ns}	59.15**	102.93**	136.53**	142.30**
AGHK110	RGHK56	0.47**	-0.51**	-0.65**	-0.96**	-38.05**	-33.47^{ns}	-46.80*	-7.30^{ns}
AGHK260	RGHK56	-0.63**	-1.06**	0.40**	0.02^{ns}	10.12^{ns}	9.23^{ns}	-18.13^{ns}	-14.50^{ns}
SE(SCA)		0.24	0.23	0.17	0.28	26.42	43.32	46.16	41.77

*, ** and ns Significant at 5% and 1% probability levels and not significant, respectively. S= Stress NS= non

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