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Determining Combining Ability in Sunflower (*Helianthus annuus* L.)

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Abstract: The experiments, involving 25 sunflower hybrids, were conducted in 3 locations in 2000 and 2001. These hybrids were evaluated for 4 important yield components (seed and oil yields, oil content and hull ratio) by combining ability. The experimental design was a randomized complete block design with 3 replications. Based on specific combining ability (SCA) analysis, the hybrid 2453-A x 2644-R was determined as the best hybrid in the experiment. The female lines 2453-A and 0704-A and the male lines R-1001 and 2644-R had higher seed and oil yield performances in this research. However, the 2644-R and 2280-R restorer and HA89-A and BAH-8-A female lines had the highest oil percentages. Among the parental lines, the 2453-A female and 2644-R male had the best GCA (general combining ability) and SCA effects based on the 4 yield traits in the experiment. Additionally, the HA89-A and R-1001 lines were selected also to test combining ability using 2 testers for both female and male lines in future breeding programs.

Key Words: Sunflower, Hybrid breeding, General and special combining ability

Ayçiçeğinde (*Helianthus annuus* L.) Kombinasyon Kabiliyetinin Belirlenmesi

Özet: Deneme 25 ayçiçeği hibridinden oluşmuş ve 2000 ve 2001 yıllarında 3 lokasyonda, tesadüf blokları deneme deseninde 3 tekerrürlü olarak kurulmuştur. Araştırmada ayçiçeğinde 4 önemli verim öğesi olan yağ ve tane verimi, yağ ve kabuk oranında kombinasyon kabiliyeti analizleri yapılmıştır. Araştırmada, en yüksek tane ve yağ verimi 2453-A ve 0704-A ana ve R-1001 ve 2644-R baba hatları, en yüksek yağ oranı da HA-89 ve BAH-8-A ana ve 2644-R ve 2280-R restorer ebeveyn hatlarıyla yapılan melezlerde ölçülmüştür. Yapılan kombinasyon kabiliyeti analizleri sonucunda; 11. (2453-A x 2644-R) melez denemede yer alan hibritler arasında en iyi genotip olarak belirlenmiştir. Ebeveyn hatlarından ise; tek bir tester için 2453-A ve 2644-R, 2 tester için de bu iki hatta ilave olarak HA89-A ve R-1001 hatları, gelecekteki ıslah programlarında kombinasyon kabiliyeti analizleri için seçilmişlerdir.

Introduction

Sunflower is one of the major vegetable oil sources in the world. The preference of Turkish people for vegetable oil, and sunflower being a major crop in rotation in some regions, increase the importance of sunflower in Turkey. Hybrid sunflower seeds are generally used in all sunflower production areas in the country.

Hybrids using lines developed based on heterosis are preferred by farmers due to their high yield performance, quality, and uniformity. The magnitude of heterosis is determined by the combining ability of the parents. Developing inbred lines that have high general combining ability (GCA) and specific combining ability (SCA) for important yield components is a main objective of sunflower hybrid breeding. The second objective is using and crossing these inbred lines to obtain superior hybrids that have high oil and yield potential and resistance to pests and diseases (Miller and Fick, 1997). The GCA of a line means the average value of its performance in hybrids when crossed with other lines. The performance of individual hybrids is used to obtain SCA, and that of the lines crossed to create that hybrid (Fick and Miller, 1997).

Combining ability analysis in sunflower in addition to yield and yield components have also been used by researchers for different characters such as fatty acid composition (Shekar et al., 1998), enzymatic polymorphism (Tersac, 1992), the cadmium level in the achene (Li et al., 1995), stay green trait (Cukadar-Olmedo et al., 1997), salt tolerance (Hussain et al., 1998), resistance to broomrape (Aydin, 1996; Goksoy et

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al., 2001), and somatic embryogenesis (Bolandi et al., 2000). In general, researchers use a Griffing diallel analysis model or a line x tester analysis in evaluating the combining ability of lines and hybrids in terms of various important traits (Petakov, 1992; Javed and Aslam, 1995; Hussain et al., 1998; Joksimovic, 1998; Goksoy et al., 1999). However, due to sunflower inbred lines having branched restorer and cytoplasmic male sterile (CMS) female lines, these methods do not fit efficiently in measuring combining ability in sunflower inbred lines and hybrids. The best method is a factorial mating design or North Carolina Design II, because these combining ability designs in cross pollinated crops are mostly utilized due to the economic importance of F1 hybrids (Beil and Atkins, 1967; Fehr, 1993; Cukadar-Olmedo et al., 1997).

Based on the combining ability analysis of different characters, higher SCA values refer to dominance gene effects and higher GCA effects indicate a greater role of additive gene effects controlling these characters in the plants. If both the GCA and SCA values are not significant, epistatic gene effects play an important role in determining these characters (Fehr, 1993).

The importance of SCA and GCA in seed yield and yield components have been evaluated by many researchers. Although some researchers found that GCA played a more important role in determining yield components (Sindagi et al., 1979; Beser and Pooni, 2001), others indicated that SCA had more effect than GCA on seed yield (Kovacik and Skaloud, 1990, 1992; Shekar et al., 1998; Laureti and Del Gatto, 2001).

Skoric et al. (2000) and Ortegon-Morales and Escobedo-Mendoza (1993) indicated that SCA and GCA are important selection characters and the most useful method to evaluate inbred lines using testers in sunflower breeding programs. They also observed that tester females made a greater genetic contribution in determining important yield components in sunflower. Rojas et al. (2000) found that GCA was of greater importance than SCA in determining seed oil and protein content and that the GCA/SCA ratio was lower for oil and protein quantity but higher for oil content.

The objectives of this study were to evaluate sunflower lines and hybrids for their combining ability in different environments and to identify the best testers for 4 important traits.

Materials and Methods

Twenty-five hybrids were obtained by crossing 5 CMS female lines (BAH4-A, BAH8-A, HA-89-A, 0704-A and 2453-A) with 5 male restorer lines (2644-R, 2284-R, 2280-R, 25711-R and R-1001) developed by the National Sunflower Research Project. These hybrids were produced in a breeding nursery in the 1999 planting season at the Trakya Agricultural Research Institute, Edirne, Turkey.

These 25 hybrids were evaluated utilizing the North Carolina Design II statistical method. This design is a factorial mating design in which each male parent is mated to each female, but none are crossed as female x female or male x male (Fehr, 1993). The experimental design was a randomized complete block design with 3 replications.

Hybrids in the research were planted at 3 locations (Edirne, Kırklareli and Beyazköy-Tekirdağ) in 2000 and 2001. The long-term climatic data for the sunflower vegetation period for these locations in 2000 and in 2001 are given in Table 1. Based on the climatic results, long-term data revealed a noticeable decrease in total precipitation in the vegetation period during 2000 and 2001.

The experiments were conducted in 6-m long, 3-row plots with a 70 x 35-cm plant density. The middle row was harvested and plot size was 3.78 m^2 at harvest. Seed and oil yield (kg ha^{-1}), oil content (%) and hull ratio (%) were measured in this experiment. Combining ability analysis for each character was calculated according to Beil and Atkins (1967). Based on this method, the i female (gi) and j restorer inbred lines (gj) and the general and specific combining ability (gij) were, respectively,

$$gi = (\hat{y}i. - \hat{y}..),\ gj = (\hat{y}j. - \hat{y}..) \text{ and}$$

$$gij = (\hat{y}ij - \hat{y}i. - \hat{y}j. + \hat{y}..).$$

Results and Discussion

GCA of both female and male lines was significant for all traits (Table 1). SCA of hybrids was also significant for all traits. The significance of GCA and SCA for all traits showed the importance of both additive and non-additive (dominance and/or epistasis) gene effects.

Table 1. Long-term climatic data of the sunflower vegetation periods, 2000 and 2001.

Climatic Comp.	April			May			June			July			August			September		
	T*	E	K	T	E	K	T	E	K	T	E	K	T	E	K	T	E	K
Long-Term																		
Max. T. (°C)	34.3	33.5	29.4	33.8	37.1	36.0	34.0	39.3	37.0	37.6	41.5	41.6	37.2	40.8	39.2	34.0	37.8	37.0
Min.T. (°C)	-1.0	-2.3	-2.5	2.7	0.6	1.8	9.2	6.7	5.8	12.6	8.0	9.0	11.0	8.0	3.3	3.7	0.2	3.0
Avg. T. (°C)	11.5	12.7	11.6	16.6	17.8	17.0	28.9	21.9	21.2	23.3	24.4	23.3	23.3	23.9	22.5	19.6	19.6	18.7
Tot. T. (°C)	345	381	348	515	552	527	867	657	636	722	756	722	722	741	691	588	588	561
T. Pre. (mm)	42.9	50.6	43.3	37.6	47.7	53.3	37.8	48.2	48.7	19.4	30.3	28.8	11.8	21.7	22.1	30.7	33.2	29.2
Humd. (%)	76	68	73	75	67	69	71	62	63	67	57	60	67	57	62	72	63	68
2000																		
Max. T. (°C)	29.7	27.6	27.7	26.9	22.6	29.3	29.2	36.8	35.9	38.4	42.2	42.0	32.3	38.8	37.0	33.2	35.8	35.9
Min.T. (°C)	2.3	2.0	0.6	5.2	2.5	0.7	11.7	7.6	5.4	15.8	13.8	12.3	15.5	12.5	11.5	8.5	5.5	2.7
Avg. T. (°C)	14.0	15.5	14.1	16.6	18.4	16.7	20.9	22.7	20.7	24.5	26.8	25.2	24.4	25.2	23.4	20.3	19.7	18.7
Tot. T. (°C)	420	465	423	515	570	519	627	681	622	760	831	781	756	781	724	609	591	562
T. Pre. (mm)	48.5	60.9	44.0	67.0	52.6	34.4	11.8	16.7	13.4	-	0.8	0.6	18.1	30.5	20.8	20.6	42.0	18.4
Humd. (%)	84.8	68.7	75.0	77.2	62.6	66.0	72.2	54.4	59.7	68.7	45.5	47.3	71.7	51.6	57.3	77.1	64.0	66.6
2001																		
Max. T. (°C)	22.0	28.4	25.5	32.0	32.8	32.3	33.0	36.8	36.0	33.6	38.2	36.5	33.7	38.7	36.7	33.9	34.2	33.6
Min.. T. (°C)	2.9	0.6	-1.8	6.6	4.3	1.5	11.8	9.0	7.3	15.9	16.7	14.4	16.5	12.4	15.7	9.3	7.5	8.6
Avg. T. (°C)	12.4	13.0	11.9	16.9	18.3	16.9	21.3	22.6	20.9	25.2	27.3	25.5	25.2	26.5	24.6	21.1	21.2	20.1
Tot. T. (°C)	361	390	356	524	567	525	639	678	626	781	846	789	781	821	762	633	636	602
T. Pre. (mm)	68.6	57.7	51.2	57.2	25.8	19.2	9.2	16.7	5.2	25.6	28.7	3.2	8.6	9.4	5.0	51.1	62.2	60.8
Humd. (%)	76.5	69.6	76.5	67.0	57.9	62.7	61.5	58.2	58.6	75.3	50.1	57.4	67.0	49.3	58.6	71.3	60.2	65.5

* T: Tekirdağ, E: Edirne, K: Kırklareli

The significant SCA effects detected in these traits imply the contribution of non-additive gene effects to the phenotypic variation among the hybrids in the research. Hybrid x environment interactions were not significant, except for hull ratio. Non-significant G x E interactions imply the possibility of evaluating hybrids without diverse environmental conditions influencing these traits (Table 2).

The range of seed yield of the hybrids tested in the experiment was between 1290 and 1644 kg ha^{-1}. The 2453-A x 2644-R hybrid had the highest averaged seed yield (1644 kg ha^{-1}) in the experiment (Table 3). Of all the female parents and restorers tested, 2 female parents, 2453-A and 0704-A, and 2 restorers, R-1001 and 2644-R, had the highest positive GCA effects on seed yield. However, the HA-89-A and 25711-R lines had the highest GCA effects on seed yield and also on all traits among the inbred lines. Although 4 of the crosses with HA-89-A had negative SCA, the HA89-A x R-1001 hybrid had the largest positive SCA value.

A primary problem in plant breeding involves choosing proper parents for testing hybrids, especially when the trait to be improved is quantitatively inherited, such as in the case of yield or oil yield. Hence, in our study, emphasis was focused on parameters that can be utilized to select parental genotypes before actually making specific crosses of interest. Due to the high magnitude of SCA of the HA89-A x R-1001 hybrid, and the R-1001 male also having the highest GCA level among male parents, this restorer was the best combiner for seed yield in the experiment. Two females and 3 males had negative GCA effects. Among the tested hybrids, 8 crosses had positive and 17 crosses had negative SCA values. The male line 25711-R had the highest GCA/SCA

Table 2. Summary of combined variance analysis for 4 sunflower traits.

Sources of Variation	df	Mean Squares			
		Seed Yield	Oil Yield	Oil Content	Hull ratio
Year (Y)	1	693,669.2 **	202,854.3 **	2327.4 **	113.7 **
Location (L)	2	265,063.7 **	56,233.0 **	13.5 **	8.3 *
Y x L	2	11,879.6 **	584.0 **	326.1 **	93.2 **
Reps (L x Y)	12	1783.5 **	617.2 **	86.3 **	18.1 **
Female (F) +	4	3594.9 **	397.3 **	35.1 **	88.1 **
Y x F	4	508.5 ns	71.1 ns	14.1 **	4.1 ns
L x F	8	582.0 ns	120.1 ns	5.5 ns	3.4 ns
L x Y x F	8	901.2 **	206.6 *	12.5 **	2.4 ns
Male (M) +	4	1870.3 **	772.0 **	89.4 **	41.1 **
Y x M	4	675.4 ns	319.5 **	15.8 **	2.7 ns
L x M	8	483.8 ns	104.2 ns	2.6 ns	2.1 ns
L x Y x M	8	806.3 *	161.8 ns	6.9 *	1.5 ns
Female x Male +	16	655.2 *	188.4 **	12.5 **	5.0 **
Y x F x M	16	278.6 ns	69.1 ns	1.8 ns	1.6 ns
L x F x M	32	408.7 ns	105.6 ns	2.0 ns	2.5 **
Y x L x F x M	32	345.4 ns	76.7 ns	2.7 ns	2.8 **
Error	288	340.2	83.7	2.8	2.1

** Significant at the 1% and * significant at the 5% probability level. ns = not significant
+ Female (F) Male (M) and the F x M interaction represent GCAf, GCAm and SCA, respectively.

effect and, in general, restorer lines had higher values than did CMS lines. This means that restorers made a greater contribution to seed yield. The GCA effects on seed yield in this research were similar to those reported by Sindagi et al. (1979) and Laureti and Del Gatto (2001).

Oil yield is the main goal of oil type sunflower production in Turkey. The oil yields of hybrids were between 582 and 742 kg ha^{-1} and the general average was 664 kg ha^{-1} in the experiment. The line 2453-A had the largest positive GCA effect for oil yield among the inbred lines in the experiment (Table 4). This line had the only positive GCA/SCA value, and this means that a greater contribution to oil yield was determined from the GCA of these 2453-A hybrids. However, HA89-A among CMS lines with a lower oil yield performance had the lowest GCA of all. The line 25711-R followed as having the second lowest oil yield capacity.

Although the cross HA89-A x R-1001 utilized the HA89-A female line, this hybrid had the highest SCA effect on oil yield due to its high seed yield capacity and the contribution of the R-1001 male line. This result showed that to obtain hybrids having high SCA effects, at least 1 of the parents should have high a GCA ratio. Crosses 2453-A x 2644-R and HA89-A x R-1001 were the highest in oil yield, followed by the hybrid 2453-A x R-1001.

The 2453-A line had the highest GCA effect, and this genotype should be considered the best combiner to improve oil yield capacity among these lines. Consequently, R-1001, 2644-R, 0704-A and 2284-R had the highest GCA effects for oil yield, in that order. However, more negative SCA effects on oil yield were determined in this study. Similar high negative GCA effects on both oil yield and oil content were obtained by Sindagi et al. (1979), Hussain et al. (1998) and Rojas et al. (2000).

The mean oil percentage of all hybrids over the 2 years was 44.9%. The oil percentages of hybrids ranged between 42.2% and 47.1% (Table 5). The hybrid 2453-A x R-1001 had the highest SCA effect in the experiment and was the best combiner for oil percentage. The hybrids BAH4-A x 25711-R and BAH8-A x 2644-R followed in that order. However, the hybrid BAH8-A x R-

Table 3. GCA effects of inbred lines and SCA effects of crosses on seed yield.

Restorer	Female / Seed Yield (kg ha^{-1})					Restorer Avg./GCA	GCA/SCA
	BAH4-A	BAH8-A	2453-A	0704-A	HA89-A		
2644-R	1440	1438	1644	1526	1374	1484	0.0
SCA	-21	-20	75	0	-34	26	
2284-R	1419	1520	1555	1489	1350	1467	2.9
SCA	-24	53	04	22	-41	08	
2280-R	1452	1382	1444	1513	1313	1421	2.2
SCA	-12	-61	50	-32	-32	-38	
25711-R	1408	1391	1476	1458	1290	1405	4.1
SCA	13	-13	12	-39	-39	-54	
R-1001	1454	1428	1596	1515	1585	1516	0.0
SCA	-38	-61	-04	-42	145	57	
Female	1435	1432	1543	1500	1382	1458	
Avg. /GCA	-24	-27	85	42	-76		
GCA/SCA	1.5	1.3	3.1	-2.3	0.0		

Table 4. GCA effects of inbred lines and SCA effects of crosses on oil yield.

Restorer	Female / Seed Yield (kg ha^{-1})					Restorer Avg./GCA	GCA/SCA
	BAH4-A	BAH8-A	2453-A	0704-A	HA89-A		
2644-R	668	679	742	700	644	687	0.0
SCA	-3	1	26	-1	-23	23	
2284-R	646	696	700	669	630	668	0.0
SCA	-7	36	02	-13	-18	4	
2280-R	669	654	651	707	621	660	-0.2
SCA	24	2	33	33	-19	-03	
25711-R	622	613	634	628	582	616	0.0
SCA	22	5	-11	-2	-14	-48	
R-1001	637	636	740	685	742	688	0.0
SCA	-36	-44	22	-17	74	24	
Female	648	656	693	678	644	664	
Avg./GCA	-15	-8	30	14	-20		
GCA/SCA	0.0	0.0	2.1	0.0	0.0		

1001 had the lowest SCA effect among the hybrids. Additionally, the hybrid 2453-A x 25711-R had the lowest oil content due to the low oil capacity of the 25711-R restorer lines.

The GCA effects assessed the contribution of each genotype to its progeny response. Positive values indicate a contribution toward a large effect on oil percentage, while negative values indicate a contribution toward smaller effects. Among the hybrids, 10 crosses had negative SCA effects and 15 had positive SCA effects. The HA-89-A female and the 2644-R and 2280-R restorer lines had the highest GCA effects on oil percentage. GCA effects contributed more than SCA effects in these lines on determining oil percentages in their hybrids.

Table 5. GCA effects of inbred lines and SCA effects of crosses on seed oil percentage.

Restorer	Female / Oil Content (%)					Restorer Avg./GCA	GCA/SCA
	BAH4-A	BAH8-A	2453-A	0704-A	HA89-A		
2644-R	45.8	47.1	44.5	45.5	46.2	45.8	0.0
SCA	0.4	0.7	-0.6	0.0	-0.5	0.9	
2284-R	44.9	45.6	44.3	44.3	46.1	45.0	0.0
SCA	0.1	0.0	0.0	-0.3	0.2	0.1	
2280-R	45.4	46.9	44.3	46.1	46.6	45.8	0.0
SCA	-0.2	0.5	-0.8	0.6	-0.1	0.9	
25711-R	43.7	43.9	42.2	42.3	44.5	43.3	80.0
SCA	0.7	0.0	-0.4	-0.7	0.3	-1.6	
R-1001	43.3	44.0	45.6	44.6	45.7	44.7	60.0
SCA	-1.0	-1.2	1.7	0.3	0.2	-0.3	
Female	44.6	45.5	44.2	44.6	45.8	44.9	
Avg./GCA	-0.3	0.6	-0.8	-0.4	0.9		
GCA/SCA	0.0	0.0	40.0	20.0	45.0		

Table 6. GCA effects of lines and SCA effects of crosses on the hull ratio of seeds.

Restorer	Female / Hull Ratio (%)					Restorer Avg./GCA	GCA/SCA
	BAH4-A	BAH8-A	2453-A	0704-A	HA89-A		
2644-R	20.6	20.1	22.5	22.7	22.0	21.6	-50.0
SCA	-0.2	-0.4	0.4	-0.2	0.5	-1.0	
2284-R	22.2	22.2	22.9	24.1	22.4	22.8	10.0
SCA	0.2	0.6	-0.3	-0.1	-0.3	0.2	
2280-R	21.7	20.9	23.3	23.6	22.1	22.3	0.0
SCA	0.1	-0.4	0.5	-0.1	-0.1	-0.3	
25711-R	21.8	21.7	23.8	25.2	22.9	23.1	0.0
SCA	-0.5	-0.3	0.2	0.8	-0.2	0.4	
R-1001	23.1	22.8	23.1	24.4	23.5	23.4	70.0
SCA	0.5	0.5	-0.8	-0.4	0.2	0.7	
Female	21.9	21.5	23.1	24.0	22.6	22.6	
Avg./GCA	-0.8	-1.1	0.5	1.4	-0.1		
GCA/SCA	-40.0	0.0	0.0	0.0	-5.0		

A thinner seed hull is another important goal in sunflower breeding. There is a reverse interaction between oil percentage and hull ratio in sunflower (Miller and Fick, 1997). Therefore, the lowest hull ratios and negative GCA and SCA effects would be valuable for our research. The hybrid BAH-8-A x 2644-R had the lowest hull ratio, but the hybrid 2453-A x R-1001 had the lowest SCA effect (Table 6).

The lowest hull ratios and GCA values were measured in hybrids with BAH8-A, BAH4-A and 2644-R as parents in the experiment. The high negative GCA/SCA effects on BAH4-A and 2644-R hybrids demonstrated a greater contribution to genetic variance from GCA effects. Similar lower GCAs in hull ratios were determined by Sindagi et al. (1979).

Conclusion

Seed and oil yield, oil percentage and hull ratio are important traits in sunflower. The lines 2453-A and 0704-A used as female parents and R-1001 and 2644-R used as male parents to produce hybrids had higher seed and oil yield performances and higher GCA effects in this research. The highest oil percentage and GCA effects were produced by the 2644-R and 2280-R restorer lines and the HA89-A and BAH-8-A female lines. Conversely, lower oil percentage and GCA effects were obtained from the BAH-8-A and BAH-4-A female lines and the 2644-R and 2280-R restorer lines.

Based on combining ability analysis, the 2453-A female line was selected as the best combiner for use as a tester line in future sunflower breeding programs due to its high performance in seed and oil yield. The 2644-R restorer line was considered the best tester for female lines due to its high GCA effects for all traits. In order to use 2 testers in our hybrid-breeding program, the R-1001 restorer line and the 0704-A female line could be added due to their high performance and GCA effects. The cross 2453-A x 2644-R was the best hybrid due to its high performance and its having one of the highest SCA effects on the desired yield characters.

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