COMBINING ABILITY FOR OIL AND PROTEIN KERNEL CONTENTS OF SUNFLOWER INBREDS IN TWO DIFFERENT ENVIRONMENTS

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SUMMARY

Sunflower (*Helianthus annuus* L.) is not only an oil crop but also an important protein crop. The meal remaining after the oil extraction is used for animal feeding as a source of protein. A selection for higher values of oil plus protein content has given positive results for the breeding of both components in other oil crops. Six lines with different sum of protein and oil kernel content were crossed using a Griffing's diallel design. Parents and F1 crosses were evaluated in replicated field trials in two environments with the following objectives: (i) to estimate the general (GCA) and specific (SCA) combining abilities and the reciprocal effects for oil and protein content and their sum, and (ii) to analyse the possible changes of these genetic effects under different environments. The GCA and SCA variances were significant for all of the traits. In general, the GCA effects were more important than the SCA effects, but for oil+protein content the SCA effects were more important than the GCA effects at Novi Sad environment. However, the lower was the GCA/SCA ratio for oil+protein, the higher the GCA/SCA ratio for oil kernel content. Reciprocal effects were more important in the Novi Sad environment.

INTRODUCTION

Sunflower (*Helianthus annuus* L.) is not only an oil crop but also a protein crop. The meal remaining after the oil extraction is used for animal feeding as a source of protein. In the past, breeding work for protein quantity and quality has been scant in sunflower and there are few genetic studies in this field. The breeding strategy adopted has been to increase protein production per hectare maintaining middle or high levels in oil production. To achieve this objective, Jiménez *et al.* (1985) proposed the selection for higher values of oil plus protein content, a strategy, which gave positive results for both components in other oil crops (Grami and Stefansson, 1977).

Selection for the sum of both compounds is more effective, since an increase in one of them will not necessarily be accompanied by a decrease in the other, as usually occurs when selection is limited to either oil or protein alone. Thus total gain is greater when the sum rather than either oil or protein is used as a criterion for selection.

Parents and F1 crosses were evaluated in replicated field trials in two different environments, with the following objectives: (i) to estimate the general (GCA) and specific (SCA) combining abilities and the reciprocal effects for oil and protein kernel content and their sum, and (ii) to analyse the possible changes of theses genetic effects under different environments.

MATERIALS AND METHODS

Six sunflower lines (S7) from different origins and differing in the value of the sum of protein and oil kernel content were chosen to form a 6x6 complete diallel (Table 1). The diallel crosses were made at Instituto de Agricultura Sostenible (CSIC)'s experimental station in Cordoba, Spain, in the 1992 spring season by pollinating 20 emasculated heads of each line with bulk pollen from a minimum of 20 plants from each of the other five lines.

Table 1. Germplasm description of six sunflower inbred lines (1)

Line	PI	Origin	Parent	Hull	Protein (%)		Oil (%)	
				(%)	Seed	Kernel	Seed	Kernel
Co-2-3	265101	URSS	P1	23	34	44	33	42
Co-228-2	431532	Rumania	P2	25	18	22	49	63
Co-289-4	431560	URSS	Р3	28	34	42	35	44
Co-268-3	431547	Turkey	P4	32	30	43	29	42
Co-275-7	340787	URSS	P5	18	19	23	35	64
Co-259-2	431510	USA	P6	21	34	38	38	43

⁽¹⁾ data from 4 years.

The parents, F_1 and F_1 reciprocals, totalling 36 entries, were evaluated in 2 environments in Novi Sad (Yusgoslavia) and Cordoba (Spain) during 1993. The experimental design was randomised complete block with three replications per environment. The experimental unit consisted of three 5.2 m long rows spaced 70 cm apart. Plots were over planted and thinned to one plant per hill, with 25 cm spacing between hills, to give a final plant density of 57 150 plants ha $^{-1}$. Oil content in kernel was determined by a NMR analyser, protein kernel content by Kjeldahl's method multiplying by 6.25 factor to convert nitrogen to crude protein. Both analyses were done on the same kernel subsample.

The protein kernel and oil+protein kernel content where transformed using arcsin transformation (Gomez and Gomez, 1984). Analyses of variance (ANOVA) were completed for all characters using plot mean data. Environments were initially analysed separately and then in combined analysis (data not shown). Due to the large genotype X environment (G X E)

interactions for oil kernel content and the lack of significant genotype effects for the sum of oil and protein kernel content, analyses were the performed at Novi Sad and Cordoba environments separately (data not shown). Environments were considered as random and the genotypes were considered as fixed effects in the ANOVA. The variation among genotypes was partitioned into general combining ability (GCA), specific combining ability (SCA) and reciprocal effects (REC) using Griffing's Method 1 (Griffing, 1956). The model for the combining ability and reciprocal effects of variance was : $\chi_{ijk} = \mu + g_i + g_j + s_{ij} + r_{ij} + e_{ijk}$, where χ_{ijk} is the ij genotype in the kth replication, μ is the overall mean, g_i and g_j are the GCA effects of parents I and I, I, I is the SCA effect, I is the reciprocal effect, and the I is the error effect peculiar to the I is the observation.

RESULTS AND DISCUSSION

Mean oil kernel content over the two environments ranged form 46.2% to 67.3%. The oil kernel contents were higher at Novi Sad (61.9%) than at Cordoba (51.7%). Mean protein kernel content, ranged from 18.2% to 37.7%. Cordoba showed higher protein kernel contents (31.1%) than Novi Sad (22.4%). The mean of the sum of protein and oil kernel content ranged from 78.3% to 87.1%. The values of oil+protein kernel contents were higher at Novi Sad (84.3%) than at Cordoba (82.7%).

The effects entry, GCA and SCA were highly significant (P < 0.001) for all traits at Novi Sad and Cordoba environments (Table 2). There were highly significant REC effects for oil and oil+protein kernel content at Novi Sad, and only for oil kernel content at Cordoba. For protein kernel content the REC were significant (P < 0.05) at Novi Sad and not significant at Cordoba. Therefore, Novi Sad showed itself to be a better environment for the expression of the reciprocal effects of oil and protein kernel content.

Table 2. Analyses of variance of diallel crosses among six sunflower inbred lines for oil, protein (transformed values) and oil+protein (transformed values) kernel content.

			Novi Sad		Cordoba					
			Mean square	S	Mean squares					
Source df		Oil kernel	Protein kernel	Oil+protein kernel	Oil kernel	Protein kernel	Oil+protein kernel			
		%	arcsin (p) ½ §	arcsin (<i>p</i>) ½	%	arcsin (<i>p</i>) ½	arcsin (p) $\frac{1}{2}$			
Entry (G)	35	36.89***	11.86***	3.45***	37.92***	20.17***	4.14***			
GCA	5	133.16***	50.78***	6.76***	123.11***	94.94***	16.21***			
SCA	15	28.65***	6.74***	3.14***	39.53***	12.31***	3.86***			
REC	15	13.03***	4.01*	2.65***	7.91***	3.12	0.40			
Error	70	1.93	1.76	0.94	3.25	1.87	1.33			

§ arcsin transformation, where p is the proportion of character.

For oil kernel content, the GCA/SCA sums of squares ratio was 1.5 at Novi Sad and 1.0 at Cordoba. For protein kernel content this ratio was 2.5 at Novi Sad and 2.6 at Cordoba. The high GCA/SCA ratios indicated that GCA effects were more important than SCA effects. This also suggests there was a predominance of additive gene effects in the inheritance of protein and oil kernel content among the lines studied. The results of oil content are in agreement with those of other authors (Skoric, 1976; Alza and Fernandez–Martinez, 1997), who studied that character. The GCA/SCA ratios obtained for protein kernel content are in disagreement with Bedov (1985), who found a nonadditive genic action governing the inheritance of protein content in sunflower seeds, but are in agreement with the results obtained by Sabbouh *et al.* (1998) for protein content of *Glycine max* (L.) Merr. Seeds.

As expected (Snedecor and Cochran 1967), the variance for the sum of oil and protein, two negatively correlated variables, was less than the sum of their variances. For that reason, it was possible to detect smaller significant differences for the sum than for either oil or protein alone. The GCA/SCA ratios for the sum of oil and protein kernel content were

^{*, **, ***} Significant at the 0.05; 0.01 and 0.001 probability levels, respectively.

different for the two environments 0.7 at Novi Sad and 1.4 at Cordoba. The greater relevance of the SCA effects when calculated on the basis of the sum of oil and protein content, probably resulted as a consequence of the negative correlation between the two variables. This increment was greater in the environment in which the GCA effects for oil kernel content were more important.

The estimates of GCA effects of each parental line and the SCA effects of their crosses are presented in Table 3. The lines P_1 , P_4 and P_6 had the highest CGA values for protein kernel content in both environments. The lines with the lowest protein content showed the lowest GCA effects, suggesting that a high-protein hybrid may be developed if both parent lines have high protein content. This is in agreement with Bedov (1985). The highest GCA values for oil kernel content were found in P_5 and P_2 , while line P_3 showed high oil content, but not significant GCA effects. In general, the sign of the estimates of the additive component for oil was different from those for protein and the sum of oil and protein. For the sum of oil and protein, P_5 showed estimates of GCA effect with an opposite sign between Novi Sad and Cordoba. For both oil and protein content the environment affected the magnitude of genetic effects, but not over their sign. Finally, line P_4 seems to be the most interesting line for its use in breeding programs for a high protein content in sunflower, because it had positive high GCA effects at Novi Sad and Cordoba.

The crosses P2xP4 and P4xP6 from Novi Sad, and P1xP3 from Cordoba, had significant and positive SCA for the sum of oil and protein kernel content, which showed high SCA effects for oil kernel content. However, these hybrids did not have the highest oil+protein kernel content.

In conclusion, the results of this study indicate that for this group of parents, breeding procedures that utilize GCA and SCA effects should be the most effective to improve the protein content.

REFERENCES

ALZA, J.O. and FERNANDEZ-MARTINEZ, J.M. 1997. Genetic analysis of yield and related traits in sunflower (*Helianthus annuus* L.) in dryland and irrigated environments. Euphytica 95: 246-251.

BEDOV, S. 1985. A study of combining ability for oil and protein contents in seed of different sunflower inbreds. p. 675-682. In: Proc. 11th Int. Sunflower Conf., Mar del Plata, Argentina 10-13 Mar. 1985. Int. Sunflower Assoc., Paris, France.

GOMEZ, K.A. and GOMEZ, A.A. 1984. Statistical procedures for agriculture research. 2a Ed. New York, John Wiley & Sons, U.S.A. 680 pp.

GRAMI, B. and STEFANSSON, B.R. 1977. Gene action for protein and oil content in summer rape. Can. J. Plant Sci. 57: 625-631.

GRIFFING, B. 1956. Concept of general and specific combining ability in relation to diallel crossing systems. Austral. Jour. Biol. Sci. 9: 463.493.

JIMENEZ, A.; FERNANDEZ-M., J.; DOMINGUEZ, J.; GIMENO, V., and ALCANTARA, M. 1985. Considerations in breeding for protein yield in sunflower. p. 46-60. *In*: Proc. of the EUCARPIA. Section oil and protein crops. Junio, 10-13, 1985. Cordoba, Espana.

SABBOUH, M.Y.; EDWARDS, L.H. and KHEIM, K.R., 1998. Heterosis and combining ability for protein and oil concentrations in the seeds of soybean (Glycine max (L.) Merr.). SABRAO Journal of Breeding and Genetics 30: 1-17.

SKORIC, D. 1976. Mode of inheritance of oil content in sunflower seed of F1 generation and components of genetic variability. Vol. II, p. 376–388. *In*: Proc. 7th Int. Sunflower Conf., Krasnodar, USSR, 27 June – 3 July 1976. Int. Sunflower Assoc., Paris, France.

SNEDECOR, G.W. and COCHRAN, W.G. 1967. Statiscal methods. 6th. Ed. Iowa State University Press, Ames, Iowa.

Table 3. Estimates of general combining ability (g_i) and specific combining ability (s_u) effects for oil, protein and oil+protein kernel content in Novi Sad and Cordoba environments

	$g_{_{\! \! \! \! \! \! \! \! \! \! \! \! \! \! \! \! \! \! $							s _{//} effects					
	Novi Sad				Cordoba	l					Cordoba	Cordoba	
Parents	Oil kernel	Protein kernel	Oil+Protei kernel	Oil kernel	Protein kernel	Oil+Protei kernel	Crosses	Oil kernel	Protein kernel	Oil+Protei kernel	Oil kernel	Protein kernel	Oil+Protei kernel
	% arcsin (<i>p</i>) ½ § -		%	arc:	sin (<i>p</i>) ½		%	arc	sin (<i>p</i>) ½	%	arcs	in (<i>p</i>) ½	
												-	
							P1 x P1	-4.07***	2.06**	-0.69	-2.45**	2.66***	1.43*
P1	-1.48***	0.49*	-0.64***	-1.40***	0.68***	-0.22	P1 x P2	0.22	-0.11	0.04	-0.44	-0.72	-1.17**
P2	1.77***	-1.34***	-0.14	2.38***	-1.86***	-0.48**	P1 x P3	0.52	0.12	0.50	1.46*	-0.00	1.04*
P3	-0.11	0.18	0.13	-0.52	0.72***	0.50**	P1 x P4	2.17***	-1.03*	0.47	0.59	-0.31	0.03
P4	-1.74***	1.47***	0.36*	-1.07***	1.29***	0.74**	P1 x P5	0.37	-0.53	-0.35	0.88	-1.13*	-0.68
P5	2.90***	-1.52***	0.55***	2.29***	-2.25***	-0.99***	P1 x P6	0.79	-0.51	0.03	-0.04	-0.50	-0.65
P6	-1.34***	0.72***	-0.26	-1.67***	1.42***	0.45*	P2 x P2	-2.63***	0.55	-1.35**	-3.37***	2.10**	-0.01
							P2 x P3	-0.11	-0.07	-0.24	2.47***	-1.33**	0.24
							P2 x P4	1.44**	-0.24	0.81*	0.08	0.25	0.36
							P2 x P5	-0.31	0.12	0.62	-0.78	0.69	0.30
							P2 x P6	1.39**	-0.83	0.12	2.04**	-1.00*	0.29
							P3 x P3	2.65***	-1.69**	0.11	-3.21***	0.49	-1.91**
							P3 x P4	-2.87***	1.56**	-0.39	-2.53***	1.52**	-0.00
							P3 x P5	2.23***	-1.18*	0.43	4.50***	-2.46***	0.42
							P3 x P6	-2.42***	1.27**	-0.41	-2.70***	1.78***	0.21
							P4 x P4	-4.07***	0.72	-2.32***	-3.30***	0.85	-1.42*
							P4 x P5	-0.16	0.61	0.60	1.76**	-0.44	0.71
							P4 x P6	3.50***	-1.62***	0.83*	3.44***	-1.88***	0.32
							P5 x P5	-1.99***	0.40	-1.12*	-5.29***	2.24**	-1.24*
							P5 x P6	-0.14	-0.01	-0.18	-1.03	1.11*	0.50
							P6 x P6	-3.12***	1.70**	-0.39	-1.72	0.49	-0.67

[§] arcsin transformation, where p is the proportion of character *, **, *** Significant at the 0.05; 0.01 and 0.001 probability levels, respectively