

COMBINING ABILITY FOR SEED YIELD AND OTHER CHARACTERS IN RAPESEED

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Abstract

Rapeseed (*Brassica napus* L.) is one of the most important oilseed crops of China. The improvement of yield and oil content is presently being emphasized for this crop. The objective of this study was to evaluate combining ability of rapeseed lines to be used in breeding for yield, oil content and other characters. Nine inbred lines used as males were crossed to five recessive genetic male sterile (RGMS) lines used as females in a factorial manner (NCII design) to produce 45 single crosses. The crosses and their parents were tested at Guiyang and Zunyi, Guizhou, China in 2007-2008. The results showed that mean squares for hybrids were significantly different for all characters. The SS ratios of sum of squares due to GCA to sum of squares of hybrids were 0.70, 0.80, 0.88 and 0.82 for seed yield, oil content, days to flowering and days to maturity, respectively, but SCA effects were significant for all characters except for days to maturity. This indicates that both additive and non-additive gene effects were important, but additive gene effects were more predominant for these characters. Males III188, III224, and Q034 gave large positive GCA effects for seed yield. Their respective values were 317.6, 253.1, and 383.5 kg ha⁻¹. Significantly positive GCA effects for oil content were detected for lines III224 and QH303-4A of which the respective GCA effects were 0.66 and 2.31%. The crosses of Qianyou 8A × Q034, QH303-4A × III224, Qianyou 3A × 2365, QH303-4A × 1190, and 24A × III153 gave significant positive SCA effects of 434.6, 429.9, 427.8, 379.4, and 347.0 kg ha⁻¹ for seed yield, respectively. Therefore, they should be considered as candidates for future hybrid breeding program.

Keywords: *Brassica napus* L., combining ability, yield, oil content

Introduction

Rapeseed (*Brassica napus* L.) is one of the most important oilseed crops of China. Its production is increasing rapidly especially during 1990s. The increase of yield in this crop is largely due to the

increase of the annual yield production resulting from the utilization of hybrids and the expansion of planting area of this crop.

In crop breeding for hybrid varieties,

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general and specific combining ability effects are important indicators of the potential of inbred lines in hybrid combinations. General combining ability (GCA) is the average performance of a line in hybrid combinations, and specific combining ability (SCA) is the deviation of certain cross from the average performance of the lines (Sprague and Tatum, 1942). Comstock and Robinson (1948) and Comstock *et al.* (1949) developed three designs known as North Carolina Designs I, II, and III, for uses to determine the importance of GCA and SCA of breeding materials. Among the three designs, NCII design or factorial design has been used more frequently in plants than others to determine combining ability and appropriate parents of a cross among intended lines. The estimates of male and female effects for Design II are equivalent to GCA effect, while the male $\times$ female interaction is equivalent to SCA effect (Hallauer and Miranda, 1988). By using this method, yield, oil content, quality characters related to oil and meal, and other characters in rapeseed were studied for combining ability, heterosis, and heritability (Shen *et al.*, 2002; Tian *et al.*, 2005; Wang *et al.*, 2007).

The relative importance of additive (GCA) and non-additive (SCA) gene actions within a breeding population is important to determine which breeding procedure will efficiently improve the performance of the characters of interest (Dudley and Moll, 1969). Many studies have been made in rapeseed on the inheritance of economic characters, especially seed yield and oil content, but the results were variable. In their materials, Brandle and McVetty (1989, 1990) reported that GCA accounted for 88 and 44% of hybrids sum of squares for seed yield, respectively. Shen *et al.* (2002) found that GCA accounted for 76.65% of the hybrid sums of squares for seed yield.

The objectives of this study were to evaluate performance of rapeseed lines for combining ability effects for seed yield, oil content, days to flowering and days to maturity, and to identify some good lines and crosses for future breeding programs.

Materials and Methods

Plant Materials

Fourteen rapeseed lines which have low erucic acid and glucosinolate contents with different developing backgrounds were used in this study. Among them, five recessive genetic male sterile lines (RGMS) which have 50% sterile and 50% fertile plants were used as females and nine inbred lines were used as males. These male and female lines are shown with basic information in Table 1. These parents were planted in Sept. 2006, and crosses were made in spring, 2007. At the flowering stage, male sterile plants were identified and tagging was made on RGMS lines. Young buds in the inflorescence of plants in female and male parents were covered before blooming with white paper bag. Flowers of male sterile plants covered were pollinated manually with fresh pollen collected from male parents. Five RGMS lines were obtained by crossing male sterile with male fertile plants of the same line manually. Nine male lines were produced by self-pollination. Seeds of each cross or line were harvested, threshed and the bulk of 8-10 plants was made for testing.

Field Experiment

All 45 crosses and 14 parents were evaluated at two locations, Guiyang and Zunyi, Guizhou, China. Both locations grow rapeseed (*Brassica napus* L.) extensively. The experiment was conducted in a randomized complete block design with three replications. Plots consisted of two rows of 5-m in length with 45-cm interrow and 33.3-cm intrarow spacings. Each plot contained 60 plants. Traditional methods of planting for rapeseed practiced in China were used at each location. At Guiyang, all the 60 entries were planted in hills on Sept 26, 2007, and thinned to two plants per hill. The experiment at this location was harvested from May 7 through May 19, 2008. At Zunyi, all 60 entries were planted in seed bed to produce seedlings on Sept 12, 2007. The seedlings were transplanted with two plants per hill on Oct 14,

2007. The harvest was made from May 17 through May 25, 2008.

Data Collection

The following attributes were measured at both locations:

Days to flowering: The number of days from planting until 50% of plants flowered.

Days to maturity: The number of days from planting until 90% of the plants matured.

Seed yield: The weight of seed in kilogram per hectare. The seeds were harvested and seed yield was determined in the following manner:

$$\text{Seed yield } Y = \frac{100 - X}{100 - Y_s} \times F.W. \times \frac{10,000}{A}$$

where Y = yield in kg ha^{-1} , X = moisture content (measured), Y_s = standard moisture content (9%), $F.W.$ = harvested yield in kg plot^{-1} , A = harvested area (m^2).

Percentage of oil: Oil content was determined by Near-infrared reflectance spectroscopy (NIRS).

Statistical Analysis

Data recorded for each character from individual locations were first analysed to determine the homogeneity of variances. They were then used to perform combined analysis of variance. The lines used as parents in this experiment were considered fixed and the locations of the experiments were considered

Table 1. Descriptions of 14 parents, 5 females and 9 males, used in this study

Designation	Prominent character	Origin
Females		
QH303-4A	High oil content with yellow seed-coat	A mutant from an open-pollinated variety Youyan no.6.
24A	Low oil content	Introduced from Sinan county, Guizhou province.
Qianyou 3A	Early flowering and maturity	Introduced from Yunan province.
	Low oil content	
Qianyou 6A	Yellow seed-coat, late flowering	Derived from a hybrid variety named Shuza no. 6 in Shichuan province
Qianyou 8A	High oil content with yellow seed-coat	Derived from a hybrid combination named You 1162 in Guizhou province
Males		
2313	High oil content with yellow seed-coat	A mutant from an open-pollinated variety Youyan no.2.
2365	High oil content with yellow seed-coat	A progeny of cross 95III15 $\times$ III501
III153	High oil content, early flowering	A progeny of cross 8909 $\times$ 9002
III176	Early maturity	A progeny of cross 325 $\times$ 8907
	High oil content, early flowering	
III188		A progeny of cross 210 $\times$ 207
III199	High oil content	A progeny of cross 206 $\times$ 225
III224	High oil content	An inbred line named 2236
Q034	High oil content, late maturity	A progeny of cross 95III20 $\times$ III507
1190	Yellow seed-coat, late flowering	A progeny of cross R $\times$ 8904

a random sample of locations in which rapeseed was grown in this area of Guizhou. The analyses of variance for the experiment were made by using Data Processing System 9.50 whose copyright belonging to Tang Qiyi, China.

In this experiment, the lines used as male and female parents were crossed in accordance with the pattern described for Design II by Comstock and Robinson (1948). The mean squares for male parents and for female parents are independent estimates of general combining ability (GCA) effects. The male $\times$ female interaction mean square is an estimate of specific combining ability (SCA) effect. When hybrids were evaluated in more than one environment, the model was as follows:

$$Y_{ijkl} = m + E_l + R_{(l)k} + G_i + G_j + S_{ij} + (GE)_{il} + (GE)_{jl} + (SE)_{ijl} + e_{ijkl}$$

where

Y_{ijkl} = observed value of the ij^{th} hybrid in the kl^{th} plot,

m = grand mean,

E_l = effect of the l^{th} environment ($l=1, 2$),

$R_{(l)k}$ = effect of k^{th} replication in the l^{th} environment,

G_i = the average effect (GCA) of the i^{th} male parent on its cross,

G_j = the average effect (GCA) of the j^{th} female parent on its cross,

S_{ij} = the deviation average effect of the ij^{th} cross from expected performance based on the parents average effects (SCA),

$(GE)_{il}$, $(GE)_{jl}$, and $(SE)_{ijl}$ = the interactions with environments for the effects defined previously,

e_{ijkl} = the error associated with the $ijkl^{\text{th}}$ observation;

and where

i = 1, 2, ..., m ; $m=9$ (m = number of male),

j = 1, 2, ..., f ; $f=5$ (f = number of female),

k = 1, 2, ..., r ; $r=3$ (r = number of replication),

l = 1, 2, ..., e ; $e=2$ (e = number of environment).

For each character, the GCA estimates (g_i or g_j) for all parental lines and SCA estimates (s_{ij}) for all hybrid genotypes were calculated according to Beil and Atkins (1967) as follows:

$$\begin{aligned} g_i &= (y_i - y_{..}) \\ g_j &= (y_j - y_{..}) \\ s_{ij} &= (y_{ij} - y_i - y_j + y_{..}) \end{aligned}$$

where y_{ij} is the mean of the hybrid of the cross between i^{th} female and the j^{th} male parents, y_i is the mean of all hybrids involving the i^{th} female parent, y_j is the mean of all hybrids involving the j^{th} male parent, and $y_{..}$ is the grand mean of all hybrids.

Standard errors for g_i , g_j and s_{ij} estimates were calculated by using the respective mean squares as follows:

$$\begin{aligned} SE_{GCA} &= MS_{fe} / mre \text{ (for females)} \\ SE_{GCA} &= MS_{me} / fre \text{ (for males)} \\ SE_{SCA} &= MS_{fme} / re \end{aligned}$$

where MS_{fe} , MS_{me} and MS_{fme} are the respective females $\times$ environments, males $\times$ environments and females $\times$ males $\times$ environments mean squares. Two-tailed t tests were used to test the significance of the g_i or g_j and s_{ij} estimates deviated from zero.

To evaluate the relative importance of additive and non-additive genetic effects of each character, SS ratio was calculated as suggested by Pixley and Frey (1991) and Lee *et al.* (2005) as follows:

$$SS \text{ ratio} = (SS_m + SS_f) / SS_{\text{hybrid}}$$

where SS_m , SS_f , SS_{hybrid} are the respective males, females, and hybrids sum of squares. The closer the ratio is to unity, the greater the influence of additive genetic effects on the character.

Results and Discussion

Combining Ability Analysis of Variance

The combined analyses of variance were made after the data for all characters of both locations were analysed and found error variances were homogeneous (Table 2).

Differences among hybrids were significant for all characters (Table 2). The mean squares attributable to male and female parentage of hybrids provide a measure of GCA effects for the two parental groups, respectively. The data indicated that GCA effects were significant ($P < 0.05$) for all characters for males and females, but seed yield.

Partitioning of the hybrid sums of squares into variations due to males (GCA_m), females (GCA_f) and females $\times$ males (SCA) showed that GCA effects accounted for over 70% of the hybrids sum of squares for each character (Table 2). The SS ratio was 0.70 for seed yield, indicating that both additive and non-additive gene effects were important for this character. Similar value of SS ratio for seed yield in rapeseed was found by Shen *et al.* (2002). In this study, the SS ratios of oil content and days to flowering were 0.80 and 0.88, respectively, but both GCA and SCA effects were significant, indicating that additive effects were more important than non-additive gene effects for these two characters. Similar SS ratio for oil content was reported by Brandle and McVetty (1990). However, Shen *et al.* (2002), who used

different genetic materials, reported that genetic effects for oil content were almost entirely additive as they found that SCA effects were not significant. The SS ratio for days to maturity was 0.82 and SCA effects for this character were not significant, indicating that additive effects were predominant for this character.

Estimates of GCA

Estimates of general combining ability for seed yield and other characters of each parent for both parental groups when combined into single cross hybrids are presented for all characters in Table 3. Although GCA effects for seed yield were highly significant, they were mostly negative. Two lines, namely, males III188 and Q034 gave significant positive effects for this character. Their respective GCA effects were 317.6 and 383.5 kg ha⁻¹. Male III224 also gave a large positive effect for seed yield with the GCA effects of 253.1 kg ha⁻¹. This showed that these males were good combiners for seed yield and should be used as parents in a breeding program to improve seed yield. Most female lines gave GCA effects for seed yield but none were significantly different from zero. However, lines

Table 2. Mean squares from combined analyses of variance for combining ability of yield, oil content, days to flowering and days to maturity of rapeseed grown at two environments

Sources	df	Mean squares			
		Yield	OC	DF	DM
Hybrids	44	1046054**	13.75**	59.68**	16.07*
GCA(Males)	8	2585316*	8.28*	142.52**	47.97**
GCA(Females)	4	2867803	104.18**	293.26**	49.14
SCA(Males $\times$ Females)	32	433520**	3.81**	9.78**	3.96
Hybrids $\times$ Env.	44	294168**	1.50**	3.41**	8.89**
Males $\times$ Env.	8	554313**	2.39**	2.54**	6.93**
Females $\times$ Env.	4	654358**	2.97**	1.27**	59.60**
Males $\times$ females $\times$ Env.	32	184109**	1.09	3.90**	3.04*
Pooled error	232	30918	0.81	0.31	2.01
SS ratio		0.70	0.80	0.88	0.82

*, ** significant at 0.05 and 0.01 levels of probability, respectively.

Abbreviations: OC = oil content; DF = days to flowering; DM = days to maturity;

Env. = environment.

Qianyou 6A and 24A were among the best choice for improving seed yield.

There were significant differences among males and females for oil content (Table 2). However, the significance corresponded to negative GCA effects of both male and female parents (Table 3). Significant positive GCA effects of 0.66 and 2.31% were detected for lines III224 and QH303-4A, respectively. These two parents should be good combiners for oil content.

Significant GCA effects were found in 6 males and all 5 females for days to flowering (Table 3). Significant and positive GCA effects were expressed by males 2365, III199, 1190 and females 24A, Qianyou 8A with the GCA effects

values of 2.86, 0.62, 2.89, 1.79, and 2.88 days, respectively. These parents should be good combiners for late flowering. Males III153, III176, III188, females QH303-4A, Qianyou 3A and Qianyou 5A gave significant and negative GCA effects of -1.91, -0.58, -4.08, -1.10, -2.93, and -0.64 days, respectively. These parents should be good combiners for early flowering.

Four male lines were found to show significant GCA effects for days to maturity (Table 3). Among them, lines 2365 and 1190 showed significant and positive GCA effects with the values of 2.00 and 1.03 days; lines 2313 and III176 showed significant and negative GCA effects with the values of -1.67 and -1.80 days. These results indicated that days to maturity

Table 3. Estimates of general combining ability (GCA) effects for four characters of rapeseed lines

Lines	Yield (kg ha ⁻¹)	Oil content (%)	Days to flowering (no.)	Days to maturity (no.)
Males				
2313	129.1	-0.24	-0.48	-1.67**
2365	-404.8**	0.40	2.86**	2.00**
III153	-209.9	-0.06	-1.91**	0.20
III176	-253.8	0.38	-0.58*	-1.80**
III188	317.6*	-1.16**	-4.08**	0.83
III199	-288.1*	-0.18	0.62*	0.56
III224	253.1	0.66*	0.19	-0.64
Q034	383.5**	0.14	0.49	-0.50
1190	73.3	0.06	2.89**	1.03*
LSD 0.05 (m)	266.6	0.55	0.57	0.94
LSD 0.01 (m)	350.7	0.73	0.75	1.24
Females				
QH303-4A	3.2	2.31**	-1.10**	0.28
24A	129.0	-0.39	1.79**	0.45
Qianyou 3A	-393.8**	-1.38**	-2.93**	-1.46
Qianyou 6A	187.7	0.01	-0.64**	-0.33
Qianyou 8A	73.9	-0.55*	2.88**	1.06
LSD 0.05 (f)	215.8	0.46	0.30	2.06
LSD 0.01 (f)	284.0	0.60	0.40	2.71

*, **: significant difference from zero at 0.05 and 0.01 levels of probability, respectively.

of crosses involving lines 2365 and 1190 were relatively longer, and days to maturity of crosses involving lines 2313 and III176 were relatively shorter than other crosses. For female parents, none of them gave significant GCA effects for this character.

For individual male parents, males 2313, III188, III224 and Q034 gave high seed yield in their cross combinations (Table 4). Their high yielding potential was associated with high GCA effects. These lines should be useful in future breeding programs. For female parents, high mean squares of GCA effects for seed yield were associated with negative GCA effects of certain parents, four females gave positive GCA effects but none of them were significant. However, the yield expression shown in Table 4 suggested that four females with positive GCA effects were quite good combiners.

The early maturity related to negative GCA effects of days to maturity. Both males 2313 and III176 gave significant and negative GCA effects, indicating that these two lines should be useful in future breeding programs for earliness.

SCA Effects

Specific combining ability effects were significant for all characters except for days to maturity (Table 2). The estimates of these

values are shown in Table 5 for seed yield, oil content, days to flowering and days to maturity. Significant SCA effects for seed yield were found in 6 crosses. Among them, 5 crosses gave positive SCA effects. The highest positive SCA effect was found in the cross of Qianyou 8A $\times$ Q034 with the value of 434.6 kg ha⁻¹, followed by crosses QH303-4A $\times$ III224, Qianyou 3A $\times$ 2365, QH303-4A $\times$ 1190 and 24A $\times$ III153, with the SCA effects of 429.9, 427.8, 379.4, and 347.0 kg ha⁻¹, respectively. These results indicate that these crosses should be considered in the production of hybrids for high seed yield.

Among 45 crosses, significant SCA effects were found in 9 crosses for oil content. Among them, 3 crosses gave positive SCA effects. The highest positive SCA effect was found in the cross of 24A $\times$ III153 with the value of 1.45%, followed by crosses Qianyou 6A $\times$ III176 and Qianyou 6A $\times$ 1190 which gave the SCA effects of 1.09 and 1.02%, respectively.

Significant SCA effects were found in 7 out of 45 crosses for days to flowering. Among them, 2 crosses gave positive SCA effects while 5 crosses gave negative SCA effects. The highest positive value of SCA effect was found in the cross of QH303-4A $\times$ III188 with the value of 1.63 days. Significant negative SCA effects of days to maturity were found in 3 out of

Table 4. Means for yield and oil content of high yielding crosses average over two locations

Characters	Yield (kg ha ⁻¹)	Oil content (%)	Days to flowering (no.)	Days to maturity (no.)
Qianyou 8A $\times$ Q034	3,090	39.44	176	241
Qianyou 6A $\times$ Q034	3,052	39.96	175	241
QH303-4A $\times$ III224	2,884	43.38	172	241
24A $\times$ III188	2,789	37.82	171	241
24A $\times$ 2313	2,786	39.35	175	241
Qianyou 8A $\times$ III188	2,782	38.62	172	244
Qianyou 6A $\times$ III224	2,711	40.72	174	240
QH303-4A $\times$ 1190	2,653	42.63	176	243
24A $\times$ Q034	2,620	39.46	177	242
Qianyou 6A $\times$ 2313	2,572	40.05	173	240

45 crosses. The lowest negative value of SCA effect was found in the crosses of Qianyou 3A $\times$ III224 and Qianyou 6A $\times$ III199 with the value of -1.64 days. These crosses should be considered in breeding for early maturity.

Conclusions

Results from the analysis of variance for combining ability demonstrated that both GCA and SCA effects were important for most characters of rapeseed included in this study. Relative importance of GCA and SCA effects for each character as examining by the SS ratio

of sum of squares due to GCA effects to hybrids sum of squares showed that, for all characters, GCA effects were more important than SCA effects. However, SCA effects were significant in the expression of seed yield, oil content and days to flowering, indicating that non-additive gene effects also contributed to the variation observed for these characters. Therefore, the performance of single cross hybrids may be adequately predicted on the basis of GCA effects, and the best hybrids should be obtained from crosses between parents having high GCA effects.

From the present study, it may be

Table 5. Estimates of specific combining ability (SCA) effects for four characters of rapeseed crosses

Characters	Crosses									
	Females	Males								
		2313	2365	III153	III176	III188	III199	III224	Q034	1190
Yield	QH303-4A	181.9	18.5	-80.1	-84.2	-217.6	-289.6	429.9*	-338.0	379.4*
	24A	330.2	-408.6*	347.0*	171.5	144.8	-61.3	-116.6	-90.1	-316.8
	Qianyou 3 A	-263.5	427.8*	4.2	-29.4	151.5	209.0	-171.1	-290.1	-38.3
	Qianyou 6 A	57.3	121.9	-130.9	-67.1	-238.5	121.5	72.3	283.6	-220.2
	Qianyou 8 A	-305.8	-159.6	-140.2	9.1	159.8	20.4	-214.5	434.6*	196.0
		LSD0.05=343.3			LSD0.01=451.9					
Oil content	QH303-4A	0.24	-0.91*	-0.58	0.05	-0.04	0.40	0.74	-0.49	0.59
	24A	0.32	0.21	1.45**	-0.87*	-0.29	0.47	0.21	0.04	-1.54**
	Qianyou 3 A	-0.58	0.28	0.51	-0.44	0.21	0.41	-0.26	0.13	-0.25
	Qianyou 6 A	0.62	0.51	-1.48**	1.09*	-0.55	-1.73**	0.38	0.14	1.02*
	Qianyou 8 A	-0.60	-0.09	0.11	0.17	0.67	0.46	-1.07*	0.18	0.18
		LSD0.05=0.83			LSD0.01=1.10					
Days to flowering	QH303-4A	-0.63	0.20	0.80	-0.37	1.63*	-0.07	-0.97	-0.93	0.33
	24A	0.64	-1.69*	0.24	0.74	0.24	1.04	-1.36	1.34	-1.22
	Qianyou 3 A	-0.97	0.70	0.97	0.13	-1.70*	-0.23	0.70	-1.10	1.50
	Qianyou 6 A	0.74	0.90	-3.00**	-0.33	-0.16	-2.03*	1.07	1.60*	1.20
	Qianyou 8 A	0.22	-0.11	0.99	-0.18	-0.01	1.29	0.55	-0.91	-1.81*
		LSD0.05=1.58			LSD0.01=2.08					
Days to maturity	QH303-4A	-0.18	0.32	0.29	-1.38	0.49	0.09	0.29	-0.01	0.12
	24A	0.99	0.15	0.12	-0.55	-1.51*	-0.08	0.45	0.15	0.29
	Qianyou 3 A	-0.77	0.23	0.36	0.53	0.56	1.16	-1.64*	0.23	-0.64
	Qianyou 6 A	0.26	-0.74	0.40	0.56	0.10	-1.64*	-0.10	0.43	0.73
	Qianyou 8 A	-0.29	0.04	-1.16	0.84	0.37	0.47	1.01	-0.79	-0.49
		LSD0.05=1.40			LSD0.01=1.84					

*, ** : significant difference from zero at 0.05 and 0.01 levels of probability, respectively.

suggested that males Q034, III188 and III224 should be good combiners and used extensively in a breeding program aimed at developing breeding materials to improve yield as they gave high GCA effects. Males Q034 and III224 were one of the parents in the cross combinations with high SCA effects for seed yield. III224 also gave high GCA effect for oil content. Males 2313 and III176 should be used in future breeding programs for earliness as they gave significantly negative GCA effects for days to maturity.

The highest yielding hybrids with considerable portions of both GCA and SCA effects should be the best choice for rapeseed breeders. In this study, top eight single cross hybrids (Table 4) were found to give positive GCA effects for both parents and positive SCA effects. Therefore, these crosses should be taken as candidate hybrids in future rapeseed breeding programs, especially crosses with high oil content.

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