Analysis of combining ability for characters of male sterile lines in rapeseed (Brassica napus L.)

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Materials and Methods

Plant Materials
Plant materials used in this study consisted of ten recessive genetic male sterile lines (RGMS) with low erucic acid and glucosinolate contents and varied oil contents. These lines are IIAB, Qianyou 3AB, Qianyou 5AB, Qianyou 7AB, Qianyou 8AB, You 2894AB, Qianyou 6AB, QH303-4AB, You 157AB and You 2341AB. These parents were planted in Sept. 2006 and crossed in a half diallel (Griffing, method 2) in spring 2007. At flowering, both male sterile and male fertile plants were identified and tagged. Flower buds of tagged plants were covered with white paper bags after bloomed flowers were cut away. Flowers of male sterile plants covered were pollinated with fresh pollen of male plants manually within three days after blooming and immature buds at the top of inflorescence were cut away. For each cross, plants with male sterile were pollinated by male fertile plants in all combinations including selfing. The pollinated flowers were covered back after pollination, and the paper bags were removed after 15 days. Four to five plants were pollinated for each cross. According to Griffing’s method 2, forty five F₁ crosses and ten parents were obtained. Seeds were obtained by bulking of 4-5 plants in each cross.

Field Experiment
Experiment was carried out in a randomized complete block design with three replications at Guiyang, Guizhou, China, during Sept., 2007 to May, 2008. Plots consisted of two rows of 5-m in length with 45-cm inter-row and 33.3-cm intra-row spacings. All the 45 crosses and 10 parents were planted in hills on 27 Sept., 2007, and thinned to two plants per hill within 45 days after planting. Each plot contained 60 plants. The experiment was harvested from May 7 through May 19, 2008.

Data Collection
After blooming, ten plants, five male sterile and five male fertile, were selected randomly for each plot and tagged. At maturity, these ten tagged plants were measured for plant height, branches per plant, pods per plant and seeds per pod. Means of these characters were used for analysis. One thousand seed weight was measured by using a bulk of seed from each plot. Days to flowering, days to maturity, and yield were based on plot observation. Oil content was analyzed by using open pollinated bulked seeds.

Statistical Analysis
Diallel analysis method of Griffing’s approach Method II Model I (Griffing, B., 1956) was used to analyze for the combining abilities of the 10 male sterile lines and their crosses.

To evaluate the relative importance of additive and non-additive genetic effects for each character, the ratio of M₂gca/M₂sca was used.

The analysis of data for GCA and SCA effects were made by using DPS 9.50 data processing system that copyright belonging to Tang Qiyi, China.

Results

Combining Ability Analysis of Variance
The results for analyses of variance for general combining ability (GCA) and specific combining ability (SCA) are presented in Table 1. The analyses indicated that both GCA and SCA variance were highly significant (\( P < 0.01 \)) for all characters except SCA variance of seeds per pod, which suggested that both additive and non-additive gene effects were important for the expression of these characters. The relative importance of GCA and SCA was judged from the ratio of mean squares GCA to SCA which
helped to indicate the predominant presence of either additive and non-additive effects. The ratios showed that additive gene action was predominant for all characters. These ratios for seed yield (1.48), pods per plant (1.92), seeds per pod (2.25) and plant height (2.44) were quite low, but very high for certain characters such as oil content (20.70).

Table 1. Mean squares for general combining ability (GCA) effects and specific combining ability (SCA) effects of nine characters of diallel crosses of rapeseed involving 10 parents.

<table>
<thead>
<tr>
<th>Sources of Variation</th>
<th>df</th>
<th>Mean squares</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Yield P/P S/P TSW B/P PH DF DM OC</td>
</tr>
<tr>
<td>GCA</td>
<td>9</td>
<td>155,548 ** 3,514 ** 3.24 ** 0.20 ** 1.78 ** 145.98 ** 16.96 ** 27.67 ** 18.42 **</td>
</tr>
<tr>
<td>SCA</td>
<td>45</td>
<td>105,029 ** 1,833 ** 1.44 * 0.06 ** 0.23 ** 59.84 ** 2.50 ** 2.62 ** 0.89 **</td>
</tr>
<tr>
<td>Error</td>
<td>108</td>
<td>28,037 830 0.68 0.02 0.11 12.44 0.44 0.64 0.31</td>
</tr>
<tr>
<td>MSgca/MSsca</td>
<td></td>
<td>1.48 1.92 2.25 3.33 7.74 2.44 6.78 10.56 20.70</td>
</tr>
</tbody>
</table>

GCA effects

Highly significant GCA effects for seed yield were found for three RGMS lines. They were lines Qianyou 8AB and You 2894AB with positive GCA effects of 230.94 and 127.65 kg ha⁻¹, and Qianyou 3AB with negative GCA effects, respectively. Although other lines were not significant, positive GCA effect were found in lines Qianyou 6AB and You 157AB. The GCA effects of the parents were found to associate with mean of crosses. These results indicate that Qianyou 8AB and You 2894AB and their crosses should give good yield performance, and, in contrast, Qianyou 3AB and its crosses should give poor yield performance.

The highly significant GCA effects for oil content (OC) were found for all RGMS lines except Qianyou 6AB. Among them, lines You 2894AB, QH303-4AB, You 157AB and You 2341AB gave significantly positive GCA effects of 0.99, 1.62, 1.20 and 1.53% while lines IIAB, Qianyou 3AB, Qianyou 5AB, Qianyou 7AB and Qianyou 8AB gave significantly negative GCA effects. The results showed that lines You 2894AB, QH303-4AB, You 157AB, You 2341AB and their crosses should give good oil content.

Among the 10 RGMS lines, the significantly negative GCA effects for pods per plant (P/P) were found for Qianyou 5AB and positive GCA effects for lines You 2894AB, You 157AB and You 2341AB, while no significant GCA effects were found for all other lines. The results indicate that You 2894AB, You 157AB, You 2341AB and their crosses gave more pods per plant.

Lines Qianyou 3AB and Qianyou 8AB gave significantly positive GCA effects for seeds per pod (S/P) at P < 0.01, while lines Qianyou 7AB and You 2341AB gave significantly negative GCA effects for the same character at P < 0.05 and P < 0.01, respectively. Other lines were not significant for GCA effects for seeds per pod.

For 1,000-seed weight, lines IIAB and Qianyou 5AB showed significantly positive GCA effects at P < 0.01, while lines Qianyou 3AB and Qianyou 8AB gave significantly negative GCA effects at P < 0.01 and P < 0.05, respectively. No significant GCA effects were found for other lines. The results indicate that lines IIAB, Qianyou 5AB and their crosses gave good performance for 1,000-seed weight.

For branches per plant (B/P), significant GCA effects were found for all lines except lines Qianyou 7AB, You 2894AB and Qianyou 6AB. Among them, lines Qianyou 3AB, QH303-4AB, You 157AB and You 2341AB gave positive GCA effects, while lines IIAB, Qianyou 5AB and Qianyou 8AB gave negative GCA effects. This indicates that lines Qianyou 3AB, QH303-4AB, You 157AB, You 2341AB and their crosses had more primary branches than others.

Significantly positive GCA effects for plant height were found for lines Qianyou 8AB, You 2894AB and You 2341AB, while significantly negative GCA effect was found for Qianyou 3AB. The results showed that Qianyou 3AB should be good for breeding for short varieties, and lines Qianyou 8AB, You 2894AB and You 2341AB were good for tall varieties.

Lines IIAB, Qianyou 8AB and You 2341AB expressed significantly positive GCA effects for days to flowering (DF), while lines Qianyou 3AB, QH303-4AB and You 157AB gave significantly negative GCA effects for this character. Similar results were found for days to maturity (DM). Lines IIAB, Qianyou
8AB, QH303-4AB and You 2341AB gave significantly positive GCA effects, while lines Qianyou 3AB, Qianyou 5AB and You 157AB gave significantly negative GCA effects. Significantly positive GCA effects for these two characters indicate that these lines were good for late flowering and maturity. On the other hand, significantly negative GCA effects were suitable for breeding for early flowering and maturity.

SCA effects
Estimates of SCA effects for seed yield were positively significant in seven crosses. The big magnitude of SCA values indicate that lines used in these crosses were of more diverse in seed yield than other crosses, and may be developed to be candidate hybrid varieties. Crosses between lines Qianyou 8A × You 2894B, You 2894A × Qianyou 6B, Qianyou 3A × Qianyou 8B and Qianyou 6A × QH303-4B which gave significant SCA effects were also high yielders among other crosses.

Significantly positive SCA effects for oil content were found in six crosses. The cross of Qianyou 7A × Qianyou 6B showed the highest positive SCA effect for this character; however, the mean oil content was not the highest. However, this combination could be used for line improvement or in case that the level of seed yield is acceptable.

Significantly positive SCA effects were found for pods per plant in four crosses. Crosses between lines IIA × You 2894B, Qianyou 3A × Qianyou 8B, Qianyou 8A × QH303-4B and You 2894A × Qianyou 6B gave highly significant SCA effects. These crosses also gave high means of pods per plant. This character is associated with seed yield; therefore it was usually found in high yield lines and crosses.

Significantly positive SCA effects of seeds per pod were found in five crosses. The highest SCA effect was found in the cross between lines IIA × QH303-4B which also showed the highest seeds per pod.

Estimates effects of SCA were found significant for seed size (TSW). Among 11 crosses, 7 showed positive values. The highest SCA effect was found in the cross between lines Qianyou 6A × You 157B which seed size was also significantly larger than others.

Significantly positive SCA effects for branches per plant were found in two crosses. The highest SCA effect was found in the cross between lines You 2894A × Qianyou 6B which also showed high branches per plants.

For plant height, ten crosses gave significantly positive SCA effects and three crosses gave significantly negative SCA effects. The highest positive value was found in the cross of lines Qianyou 5A × You 2341B which also gave high plant height. The maximum value of negative SCA effect was found in the cross between lines You 2894A × QH303-4B which also gave relatively lower plant height. The results indicate that crosses with significantly positive and negative SCA effects were good for developing respective tall and short hybrids.

For days to flowering, eleven crosses gave significantly positive SCA effects, four crosses gave significantly negative SCA effects. The highest positive value was found in the cross of lines QH303-4A × You 157B. The maximum value of negative was found in the cross between lines IIA × Qianyou 8B. For days to maturity, significantly positive SCA effects were detected in nine crosses, while significantly negative SCA effects were found in eight crosses. The highest positive value was found in the cross of lines Qianyou 3A × QH303-4B. The maximum value of negative SCA effect was found in the cross between lines Qianyou 7A × You 2894B. The results indicate that crosses with significantly positive SCA effects were good for developing late hybrids, while crosses with significantly negative SCA effects were good for developing early hybrids.

Discussion
The analyses of variance for GCA and SCA indicated that both GCA and SCA were important for all characters studied in this experiment. However, the ratios of MSgca/MSsca indicate that additive gene effects are more important than non-additive gene effects. The small ratios of MSgca to MSsca for seed yield and pods per plant may indicate the high heterosis of these characters because heterosis relates proportionally to non-additive gene actions. For other characters, especially oil content, the ratio of MSgca/MSsca was very high (20.70). The high magnitude of mean square GCA relative to SCA indicates that the diversity within the materials studied is not high and line improvement for these characters is effective.

The analyses of GCA did not show that any single line was a high general combiner for all the characters simultaneously. Outstanding lines such as You 2894AB gave significant GCA estimates for seed yield, oil content, pods per plant and plant height should be favourable for any breeding program. Moreover, most of the crosses involving You 2894AB were good yielders. Therefore, it was the best choice for breeding of rapeseed hybrids. However, Qianyou 8AB was a good combiner for seed yield.
It gave significant GCA effects and some of its crosses expressed high yield. Though Qianyou 3AB was not a good combiner for seed yield, it was good for short plant height, early flowering and early maturity, and its cross sometimes also could result in high yield as the cross lines Qianyou 3A × Qianyou 8B. It can be used in breeding program for short plant height and early maturity.

Analysis of specific combining ability in this study revealed that a number of crosses showed significant SCA effects for each character, but none showed the best SCA effects simultaneously. Cross lines Qianyou 8A × You 2894B gave the highest seed yield and also gave significant SCA effect for this character which indicated that it should be the best hybrid for seed yield. Cross lines Qianyou 3A × Qianyou 8B gave the highest SCA effect, the yield was also high, should be the second choice for seed yield. The highest and positive SCA effect for oil content was found in the cross of lines Qianyou 7A × Qianyou 6B, which gave oil content of 38.9%. This indicates that the magnitude of SCA effect may not correspond to the expression of the character as the characters is the sum of mean, GCA and SCA effects and environmental effect.

Most crosses with significant SCA effects for seed yield also showed significant SCA effects for yield related traits. For example, crosses lines Qianyou 3A × Qianyou 8B, You 2894A × Qianyou 6B and Qianyou 8A × QH303-4B also showed significant SCA effects for pods per plant. Crosses lines IIA × QH303-4B, Qianyou 6A × You 2341B and You 2894A × Qianyou 6B also showed significant SCA effects for seeds per pod. Cross lines IIA × Qianyou 5AB also showed significant SCA effects for 1,000-seed weight. These indicate that there are some relationships between the magnitude of SCA effects for seed yield and that of yield related traits.

Parents of some crosses with high SCA effects were both negative, or one negative and one positive, or both positive in GCA effects. These indicate that high SCA effects can be resulted from any parents with high or low GCA effects.

The crosses of lines Qianyou 3A × You 157B and Qianyou 3A × Qianyou 7B showed significantly negative SCA effects, and both crosses gave the shortest days to flowering and days to maturity. This indicates that these two crosses can be used for improving early flowering and early maturity lines.