Mutagenic Improvement of Shattering Characteristic of the Restorer Line of a Hybrid Rice “Xieyou 9308”

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Abstract: The newly developed hybrid rice combination “Xieyou 9308” (Xieqingzao A/T9308) has been regarded as a super-high-yielding rice variety, of which the yield potential reached as high as 12 t/ha. However, its high shattering characteristic (as high as 60%) has limited its wider application. In the current experiment, a non-shattering mutant line, M9308, was developed from T9308 by gamma irradiation. Comparisons were made but no marked differences were found between T9308 and M9308 as well as between their F1 hybrids crossed to Xieqingzao A for major agronomic and grain quality characters as well as resistance to diseases. Genetic analysis indicated that the non-shattering character of M9308 was governed by a single recessive gene.

Key words: hybrid rice; non-shattering; mutation; high shattering character

Xieyou 9308, a cross of Xieqingzao A/T9308, bred by China National Rice Research Institute, is a new hybrid rice registered in Zhejiang Province in 1999. It has super-high-yielding potential and superior grain quality as well as multiple disease resistance. In a demonstration experimental field in Xinchang City, Zhejiang Province in 1999, its yield reached 12.2 t/ha in a 6.6 ha scale paddy field, which surpassed the first stage breeding goal of 12 t/ha set for super hybrid project in China. Therefore, Xieyou 9308 was regarded as one of the excellent super hybrid rice varieties in China.

During the commercialization of this hybrid rice, it has been found that Xieyou 9308 has a high shattering rate than conventional rice varieties, and this character was inherited from its restorer line T9308. It has become a problematic issue for this otherwise promising new variety; large amount of seeds dropped to the field before or during harvest, which sometimes caused serious yield losses. It was particularly significant when it encountered strong wind and severe rain before harvest. The high shattering character has also made it not suitable for mechanized harvest. To overcome this problem, dried seeds of T9308 were irradiated with gamma rays, and non-shattering mutants were selected for developing middle shattering hybrid rice, which is otherwise the same as or similar to Xieyou 9308. Such a mutant restorer line, M9308, was developed and its performance was studied in this experiment.

MATERIALS AND METHODS

Development of non-shattering mutant

The experimental seeds of T9308, the restorer line of hybrid combination “Xieyou 9308”, were provided by China National Rice Research Institute. Dried seeds with the moisture content of about 13.5% were treated with gamma rays at dosages of 200 Gy, 300 Gy and 400 Gy in a cobalt-60 source in Zhejiang University. M1 seeds were sown and transplanted in hill to produce M2 seeds, which were harvested in bulk. The M2 seeds were sown and transplanted in a single seedling. At maturity, low-shattering mutants of M2 were screened individually by hand pinching the panicles one by one. Plants with the least shattering were selected, and further tested in comparison with T9308. The method is to let panicles freely drop onto the cement ground from two meters above, and the shattering rate (shattered grains/total grains) was calculated for each selected plant. Plants with a shattering rate less than 20% were selected as non-shattering mutants.

Comparative analysis

The most promising non-shattering mutant line, M9308, was selected and further analyzed. The shattering rate of M9308, T9308, and their F1 hybrid with Xieqingzao A, as well as the agronomic traits, grain quality and disease resistance were compared pair-wise.

Genetic analysis of non-shattering mutation

The mutant line, M9308, was crossed with T9308, and the segregation of shattering character was investigated by measuring the shattering rate of each of the resulting F2 plants.

RESULTS

Selection of non-shattering mutant M9308

Three M2 populations were generated by using the irradiation of different dosages. No non-shattering or low shattering mutant was selected from the 200 Gy gamma rays derived M2 population (15 000 plants). Two such mutants were
selected from the 300 Gy gamma rays derived M₁ population (35,500 plants), and one from the 400 Gy M₂ population (22,500 plants). Therefore, the mutation rate was quite low, ranging from 0 to $5.63 \times 10^{-6}$.

Among these three mutants, one mutant line, M9308, was selected for further analysis because it was almost non-shattering and most similar to its parental variety T9308 in plant shape and other characters.

**Shattering and agronomic performance of M9308**

The mutant line, M9308, in M₁ generation, was already homozygous, and grew uniformly in the field. A comparison was conducted for the shattering rate between M9308 and T9308 at different degree of maturity (Fig. 1). From 10 to 35 days after heading, the shattering rate of T9308 increased dramatically from 0 to about 67%, while that of M9308 only slightly increased to the maximum of 2.5% at full maturity.

M9308 had similar phenotype to T9308 in the field, both had fewer tillers, straight, slightly curl and dark green leaves, and slender grains. No significant difference was found between M9308 and T9308 in major agronomic traits including days from sowing to heading, plant height, flag leaf length, numbers of tillers per plant, total seeds number per panicle, seed setting rate and 1000-grain weight.

**Performance of F₁ hybrids**

The shattering character of F₁ hybrid between Xieqingzao A and M9308 was significantly improved in comparison to Xieqingzao A/T9308. It had a shattering rate of 29.8% in sharp contrast to 65.0% of Xieqingzao A/T9308. This rate was comparable with that of Xieqingzao B (33.5%).

The F₁ hybrids of Xieqingzao A/M9308 and Xieqingzao A/T9308 had almost same agronomic traits, both displayed compact plant type, tall and straight flag leaf, large panicle, and strong hybrid vigor (Table 1), and so did the grain quality and disease resistance parameters (Table 2).

For further evaluating its combining ability, which is the key character for a restorer line, crosses of M9308 were made, together with T9308, with three CMS lines Zhenshan 97A, II 32A and Longtefu A. The shattering character and major agronomic traits of the hybrids were observed and compared in pairs between those of M9308 and those of T9308. The only significant difference observed between two sets of hybrid was the shattering rate. The average rate of shattering was 28.0% (26.7% to 28.8%) for the hybrids of M9308, and 60.6% (58.0% to 62.3%) for that of T9308. There was no significant difference between the hybrids of M9308 and T9308 crossed to the same CMS line of other traits.

**Inheritance of non-shattering mutation**

The F₁ hybrid plants between M9308 and T9308 had an average shattering rate of 54.2%, which was far higher than their mathematic mean (34.7%) and that of M9308 (2.5%), it was slightly lower than that of T9308 (67.2%). Therefore,

**Table 1. Comparison on major agronomic characters between the mutant M9308, the parent T9308, and hybrid rice combinations originated from them.**

<table>
<thead>
<tr>
<th>Material</th>
<th>Shattering rate (%)</th>
<th>Days from sowing to heading</th>
<th>Plant height (cm)</th>
<th>Flag leaf length (cm)</th>
<th>No. of tillers per plant</th>
<th>Panicle length (cm)</th>
<th>Seed setting rate (%)</th>
<th>1000-grain weight (g)</th>
<th>Yield per plant (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>M9308</td>
<td>2.5±0.3</td>
<td>94.0</td>
<td>90.2±1.5</td>
<td>37.6±1.3</td>
<td>9.6±1.1</td>
<td>25.2±0.8</td>
<td>194.6±15.4</td>
<td>90.0±2.0</td>
<td>22.8±0.3</td>
</tr>
<tr>
<td>T9308</td>
<td>67.2±2.4</td>
<td>94.0</td>
<td>89.6±1.5</td>
<td>37.4±1.5</td>
<td>8.8±1.3</td>
<td>25.5±0.7</td>
<td>187.4±15.9</td>
<td>89.7±2.1</td>
<td>22.5±0.3</td>
</tr>
<tr>
<td>XQZA/M9308</td>
<td>29.8±1.7</td>
<td>102.0</td>
<td>98.4±2.3</td>
<td>36.2±1.6</td>
<td>14.0±1.4</td>
<td>26.5±1.1</td>
<td>225.0±13.2</td>
<td>92.0±0.7</td>
<td>26.2±0.3</td>
</tr>
<tr>
<td>XQZA/T9308</td>
<td>65.0±1.6</td>
<td>103.0</td>
<td>100.4±2.5</td>
<td>37.6±0.8</td>
<td>14.2±1.3</td>
<td>26.3±1.3</td>
<td>219.0±11.6</td>
<td>92.4±0.5</td>
<td>26.6±0.2</td>
</tr>
</tbody>
</table>

**Table 2. Comparison on grain quality and disease resistance between Xieyou M9308 and Xieyou 9308, originated from the mutant M9308, and the original parent T9308, respectively.**

<table>
<thead>
<tr>
<th>Combination</th>
<th>Grain length (mm)</th>
<th>Length to width ratio</th>
<th>Chalkiness (%)</th>
<th>Translucency (%)</th>
<th>Alkali spreading value</th>
<th>Gel consistency (mm)</th>
<th>Amylose content (%)</th>
<th>Blast resistance (score)</th>
<th>Blight resistance (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>XQZA/M9308</td>
<td>7.2</td>
<td>3.1</td>
<td>26.6</td>
<td>4.0</td>
<td>4.4</td>
<td>56.0</td>
<td>21.5</td>
<td>4.0</td>
<td>1.9</td>
</tr>
<tr>
<td>XQZA/T9308</td>
<td>7.2</td>
<td>3.2</td>
<td>28.2</td>
<td>4.2</td>
<td>4.2</td>
<td>58.3</td>
<td>20.9</td>
<td>4.3</td>
<td>1.8</td>
</tr>
</tbody>
</table>
non-shattering seemed to be almost recessive to high shattering.

The distribution of F2 plants with various shattering rate had been illustrated in Fig. 2. It is a bimodal curve, with a break around 20%. The F2 plants could thus be divided into two types, low shattering and high shattering, fitting into a typical Mendelian ratio of 3:1 (36:15) for single gene inheritance mode.

**DISCUSSION**

It has been well documented that high shattering rate is a wild character. In wide cross between wild rice and cultivated rice, the hybrids tend to have high shattering rate, indicating that high shattering is a dominant character relative to low or non-shattering. However, the inheritance of shattering in crosses within cultivated rice is more complicated. In some crosses, both of high and low shattering rate had been reported as dominance to the opposition [3, 4]. Huang et al [5] recently reported that the shattering character of 14 F1 hybrids between high shattering sterile lines and low shattering restorer lines exhibited a high shattering character, which implied that high shattering character was dominant. From the data obtained in this experiment, we can speculate that the shattering rate in rice might be controlled by a set of allelic genes, of which the higher shattering is partially dominant. For example, the shattering allele in T9308 (high shattering, ca. 67%) is partially dominant to that in Zhenshan 97, Xieqingzao and II 32B (middle shattering, ca. 30%), while the latter is partially dominant to M9308 (non-shattering, ca. 2%).

Shattering is an important agronomic trait, neither too high nor too low shattering rate is preferred. In areas where rice is harvested and threshed by manual equipment, the shattering rate should be a little higher than that in areas using electronic threshers. For most rice varieties commercially used currently, the shattering rate (determined by dropping the panicle from 2.0 m above onto ground as described above) is around 25% (most japonica rice) to 30% (most indica rice). Although the mutant restorer line M9308 developed in this experiment had very low shattering rate (some 2%), but the hybrid F1 plants using it as the restorer line had the shattering rate similar to its CMS line. Hybrid rice with a shattering rate of 25% to 30% was preferable for the sake of easy threshing and minimum yield losses. Therefore, the non-shattering mutant M9308 will not incur a new problem of difficulty to shatter in hybrid rice production.

Based on the above findings, the hybrid rice, Xieqingzao A×M9308, could be a new super hybrid rice combination with appropriate shattering character while keeping all other merits of Xieyou 9308.

**REFERENCES**