STUDIES ON INDUCED MUTATION OF SESAME MALE STERILITY

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Abstract

The dry seeds of the high yielding cultivar, Yuzhi-4, were irradiated with 300, 500 and 700 Gy of ^{60}Co-gamma rays. 3277 M_1 plants were harvested separately as single plants and also in bulk, by doses. In M_2, the single plant seeds were grown in progeny rows and the bulked seeds were grown as bulks. 25 male sterile plants were screened from M_2. 10 of the 25 male sterile plants were from the progenies of the single plant seeds and 15 were from the progenies of the bulked seeds. In further genetic research of the 25 male sterile plants in M_3 and M_4, 6 separate genic male sterile (GMS) lines were identified. Their male sterility was stable and was controlled by a pair of alleles, male fertility being dominant to sterility.

1. INTRODUCTION

Sesame is an oil crop which can show a high degree of heterosis. Its hybrids' yields can increase by 20–30%, and in the best a 60% increase can be obtained. Sesame heterosis breeding research is progressing slowly because male sterile breeding material is scarce and not ideal. Cytoplasmic-genic male sterile (CGMS) material has not been found in the world so far. The existing genic male sterile (GMS) materials are not good. Their male sterility rates are low (less than 50%) and their agronomic characters are not ideal. Induction of mutation is an efficient method for creating new breeding materials, so research was carried out to create new male sterile breeding material that would be ideal and be used for heterosis breeding directly.

2. MATERIALS AND METHODS

In M_1, the dry seeds of the high yielding cultivar,'Yuzhi-4', were irradiated with 300, 500 and 700 Gy of ^{60}Co-gamma rays. All M_1 plants were selfed and harvested individually by treatment. The number of the harvested capsules from the single plant varied: 5–10 when the capsules were large, 10–15 if medium and 15–20 if they were small. In M_2, the seeds from the single plants were grown in progeny rows and the mixed seeds of each dose treatment were grown in a bulk. The anthers of each M_2 plant were observed carefully. The male sterile plants whose anthers did not contain any pollen or contained only a little pollen were labeled at early flowering stage. All male sterile plants were observed every day during the whole flowering period. Their male sterile expression was recorded. All male sterile plants were crossed with their sib-plants at 5 a.m., before the bees started work. Wheat straw pieces were used to cover the stigmas of the male sterile plants after pollination to prevent out-crossing. The capsules of each male sterile plant were harvested separately at maturity. In M_3, the seeds from the single capsules of the male sterile plants were grown separately. The male sterility of M_3 plants was recorded. The segregation ratio of the male sterile and fertile plants in the progenies of the single capsule seeds were counted. The heterozygous fertile (*Ms/ms*) plants (sib-plants of the male sterile plants) which segregated among the progenies of the single capsule seeds were selfed, and the male sterile plants which segregated from them were crossed with the heterozygous male fertile plants (sib-plants) which segregated from the progenies of the same capsules and with the homozygous fertile (*Ms/Ms*) plants (non-irradiated parent plants). In M_4, the seeds from the selfed *Ms/ms* plants and from the male sterile plants which were crossed with the *Ms/ms* plants and with the *Ms/Ms* plants were grown separately. The male sterile expression of all M_4 male sterile plants was checked further. The frequencies of male sterile and fertile plants in the progenies of the three combinations were recorded.

3. RESULTS AND DISCUSSION

3.1. Development of M_1 plants

The single plant seeds of 3277 M_1 plants and the mixed seeds of all M_1 plants of each dose treatment were harvested. 1147 of 3277 M_1 plants were from 300 Gy treatment, 1125 from 500 Gy treatment and 1005 from 700 Gy treatment.

3.2. Screening for male sterile plants in M_2

In M_2, the seeds from each plant of 3277 M_1 plants were grown in progeny rows and the mixed seeds of all M_1 plants of each dose treatment were grown in a bulk; 25 male sterile plants were recovered. They were numbered as 95ms-1, 95ms-2, 95ms-3, 95ms-4, 95ms-5, 95ms-6, 95ms-7, 95ms-8, 95ms-9, 95ms-10, 95ms-11, 95ms-12, 95ms-13, 95ms-14, 95ms-15, 95ms-16, 95ms-17, 95ms-18, 95ms-19, 95ms-20, 95ms-21, 95ms-22, 95ms-23, 95ms-24, 95ms-25. 9 of the 25 male sterile plants came from 500 Gy treatment, 16 from 700 Gy treatment, and none from the 300 Gy treatment. Ten male sterile plants, 95ms-1, 95ms-2, 95ms-3, 95ms-4, 95ms-5, 95ms-6, 95ms-7, 95ms-8, 95ms-9 and 95ms-10 were found among the single plant progenies and the other 15 male sterile plants were screened from the bulk progenies from each dose treatment. During the whole flowering period, the 10 male sterile plants had different male sterile expressions. They are described below.

95ms-2 and 95ms-19 gave completely male sterile plants. During the whole flowering period their anthers had no pollen. They were completely male sterile when they were selfed and could bear seeds when they were crossed with their sib-plants. The anthers of 95ms-2 were brown, while those of 95ms-19 were green. The anthers of both were flat and a little smaller than normal.

In 95ms-1, 95ms-8, 95ms-9 and 95ms-10 the male sterility could be environmentally affected. Their anthers contained a little pollen but showed self-sterility when the temperature was high late in the flowering period. The anthers were normal and showed self-fertility when temperatures were low at the early flowering stage. Their anther color and shape were different, i.e. greenish and flat when they were sterile, and normal when they were fertile.

95ms-3, 95ms-4, 95ms-5, 95ms-6, 95ms-7, 95ms-12, 95ms-13, 95ms-20, 95ms-21 and 95ms-23 were unstable sterile plants. Their anthers sometimes contained a little pollen, sometimes contained no pollen. At times, all anthers in a flower contained a little pollen, sometimes only one or two where the others contained no pollen. Their anther color was special; one half was greenish and the other half was white. Their anther shape varied; some were small and flat, and some were long and flat. They expressed fertility when anthers contained pollen, and sterility when anthers contained no pollen.

95ms-11, 95ms-14, 95ms-15, 95ms-16 and 95ms-17 gave male sterile plants whose anthers were malformed. Their anthers were triangular and very small, being about one fifth or one tenth of the normal anther size. The anther color was yellow. They were male sterile during the whole flowering period.

95ms-18 and 95ms-25 were the male sterile plants whose anther color and shape were unstable; their anther color was sometimes green, and sometimes yellow or white-green. Their anthers were sometimes big, sometimes small and sometimes crescent-shaped.

95ms-22 and 95ms-24 were false male sterile plants. They expressed male sterility during 3–5 days when the male sterile plants were being counted, but they always expressed male fertility later.

3.3. Identification and preliminary genetic research of male sterile mutants in M_3

3.3.1. Identification of 6 male sterile mutants

In M_3, 25 male sterile mutants behaved as follows: i) two mutants (95ms-1 and 95ms-25) did not have any progeny because their seeds were unfilled; ii) 6 male sterile mutants (95ms-2, 95ms-3, 95ms-4, 95ms-5, 95ms-6 and 95ms-7) segregated for male sterile and -fertile plants continued; iii) the other 17 male sterile mutants did not produce any male sterile plants.

The male sterility of the M_3 male sterile plants which segregated among the progenies of the 6 male sterile mutants, was stable. During the whole flowering period their anthers did not contain any pollen; they were completely sterile when they were selfed. The anther color of the male sterile plants from 95ms-2 was brown and all anthers of all the male plants from the other sterile mutants were green. The anther shape of all male sterile plants was flat and a little shorter than the normal anthers.

3.3.2. Genetic research of the 6 male sterile mutants

Segregation was observed for male sterile and fertile plants in the progenies of each single capsule seeds of each male sterile mutant. The progenies of some single capsule seeds produced male sterile and -fertile plants, some only produced male fertile plants, and none produced only male sterile plants. The result showed that the 6 male sterile mutants that gave male sterile and -fertile segregation in M_3 were genic male sterile (GMS) mutants. Progenies of the single capsule seeds segregated male sterile and -fertile when the capsules resulted from crosses with heterozygous fertile (*Ms/ms*) plants in M_2, and produced only male fertile plants when the cross was with homozygous fertile (*Ms/Ms*) plants in M_2.

Segregation ratio was observed in M_3 for male fertile vs. -sterile plants in all the progenies that were raised from each different male sterile mutant and that segregated. The segregation ratio of male fertile and -sterile plants was 38:30 for 95ms-2, 41:38 for 95ms-3, 57:52 for 95ms-4, 88:75 for 95ms-5, 74:71 for 95ms-6 and 37:34 for 95ms-7. The result showed that the observed frequencies did not deviate significantly from the expected monogenic segregation (1:1) ratio (Table I). The findings showed a good fit to monogenic control of male sterility, with the fertility allele being dominant.

TABLE I. SEGREGATION FOR MALE -STERILE AND -FERTILE PLANTS IN ALL THE PROGENIES THAT WERE RAISED FROM THE SINGLE CAPSULES OF THE SIX MALE STERILE MUTANTS

Male sterile mutants	Observed ratio F:S	Expected ratio F:S	X^2 value	P value
95ms-2	38:31	1:1	0.523	0.5-0.25
95ms-3	41:38	1:1	0.051	0.9-0.75
95ms-4	57:52	1:1	0.147	0.75-0,5
95ms-5	88:75	1:1	0.883	0.5-0.25
95ms-6	74:71	1:1	0,028	0,9-0.75
95ms-7	37:34	1:1	0.056	0.9-0.75

3.4. Further genetic research of 6 male sterile mutants in M_4

In M_4, segregation was observed for male -sterile and -fertile plants in the progenies of the selfed heterozygous fertile (*Ms/ms*) plants, the male sterile (*ms/ms*) plants crossed with heterozygous fertile (*Ms/ms*) plants and the male sterile (ms/ms) plants crossed with homozygous fertile (*Ms/Ms*) plants. The segregation ratios of male sterile and fertile plants in the progenies of 3 combinations are listed in Table II. The observed frequencies in the three combinations did not deviate significantly from the expected values. The high probability values showed good fit to the 1:1, 1:0 and 3:1 ratios, further confirming a monogenic control of the male sterility with male fertility completely dominant over male sterility.

TABLE II. SEGREGATION FOR MALE -STERILE VS. -FERTILE PLANTS IN THE PROGENIES OF *ms/ms* x *Ms/ms*, *ms/ms* x *Ms/Ms* AND THE SELFED *Ms/ms*

Cross/self	Observed ratio F:S	Expected value F:S	X^2 value	P value
95ms-2xMs/ms	621 : 603	1:1	0.236	0.750–0.500
95ms-2xMs/Ms	163 : 0	1:0	–	–
Selfed Ms/ms	225 : 69	3:1	0.290	0,750–0.500
95ms-3xMs/ms	716 : 660	1:1	2.198	0.250–0.100
95ms-3xMs/Ms	157 : 0	1:0	–	–
Selfed Ms/ms	219 : 72	3:1	0.0014	>0.995
95ms-4xMs/ms	1167:1086	1:1	2.842	0.100–0.050
95ms-4xMs/Ms	125 : 0	1:0	–	–
Selfed Ms/ms	243 : 75	3:1	0.2683	0.750–0.500
95ms-5xMs/ms	843 : 819	1:1	0.318	0.750–0.500
95ms-5xMs/Ms	184 : 0	1:0	–	–
Selfed Ms/ms	234 : 72	3:1	0.2788	0.750–0.500
95ms-6xMs/ms	1109:1026	1:1	3.149	0.100–0.050
95ms-6xMs/Ms	198 : 0	1:0	–	–
Selfed Ms/ms	254 : 76	3:1	0.5818	0.500–0.250
95ms-7xMs/ms	1126:1053	1:1	2.306	0.250–0.100
95ms-7xMs/Ms	175 : 0	1:0	–	–
Selfed Ms/ms	213 : 65	3:1	0.3069	0.750–0.500

4. CONCLUSION

Induced mutation is an efficient method of creating new breeding material. The 6 GMS lines induced by ^{60}Co gamma rays possessed good economic characters and resistance to disease. They were similar to their parent (a good cultivar, Yuzhi-4) in all ways except their male sterility. They have been used for heterosis breeding.

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