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Improving outcrossing rate in rice (*Oryza sativa* L.)

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Floral traits that affect the outcrossing rate in rice are discussed; those that facilitate pollen dispersal and those that facilitate stigma reception of pollen are enumerated. Except for the variability of stigma and anther characteristics, variability for nearly all traits that influence outcrossing is large in *Oryza sativa.* Variability for stigma and anther characteristics is found in allogamous species. Two approaches (backcrosses and pedigree selection, and recurrent selection) used to transfer the long stigmas of wild species to *O. sativa* are described, and results are given.

Success in breeding and growing hybrid rice commercially in China has created great interest among rice breeders throughout the world. However, the natural outcrossing rate in rice cultivars is extremely low and hybrid seed production, using male sterility or gametocides, presents some difficulties. Increasing the potential outcrossing rate would facilitate use of hybrids in other areas of the world.

Floral traits and outcrossing aptitudes of rice species

The *Oryza sativa* species

The inflorescence of *O. sativa* is a terminal panicle of perfect flowers. Each spikelet has a branched stigma, six stamens, and two lodicules. At blooming, the flowers open rapidly. Usually the anthers dehisce just before or immediately after the spikelets open. The flowers remain open from 1 to 3 h and close after anthesis; they never reopen.

Some floral traits, such as stigma size, stigma exsertion, and flower opening duration, affect the rate of outcrossing in rice. *O. sativa* stigmas are small and slightly feathery. Usually the stigma remain inside the glumes during and after anthesis. In some cultivars, some stigmas remain outside the glumes after anthesis and fertilization. This characteristic is more frequent in indicas.

The rate of exserted stigma, which ranges from 0 to 90%, is correlated with stigma length. Parmar et al (1980), Virmani et al (1980), Sarkar and Miah (1983a), and Taillebois (1983) reported stigma lengths of 0.4–1.6 mm. The duration of receptivity is variable and can be longer than 5 d (Virmani and Tan 1982, Yoshida 1981).

Anther length ranges from 0.9 mm to 2.8 mm (Parmar et al 1980, Taillebois 1983, Virmani and Athwal 1973) and correlates highly with number of pollen grains per anther. Oka and Morishima (1967) found 700-2,500 pollen grains/anther. After pollen emission, the duration of fertilizing capacity is rather short, generally less than 5 min.

O. sativa, because its floral structure is not well adapted to cross-pollination, is highly autogamous. In general, natural cross-pollination is less than 1%, depending on environmental conditions and cultivars. When male sterile plants are used, the outcrossing rate is much higher; in China, up to 45% seed set was obtained. Sarkar and Miah (1983b) observed natural outcrossing rates of 0.5-2.5%. Silitonga (1985) reported a maximum seed set of 14%, with a mean of 6%. Outcrossing rates up to 92% were reported for individual plants (Virmani and Tan 1982). At the National Research Center for Rice and Beans (CNPAF), in an indica population of fertile and sterile plants, outcrossing averaged 7.8%.

Male sterile spikelets behave differently from fertile spikelets. At CNPAF, male sterile spikelets had better stigma exsertion and stayed open longer. Parmar et al (1980) reported similar observations and also mentioned a relationship between length and exsertion of stigma and size and function of anthers. In general, anthers with sterile pollen are associated with persistent protruding stigmas.

Hoff and De La Torre (1981) reported a high correlation between seed set and stigma exsertion in male sterile plants. Their results suggest that male sterile plants with 100% exserted stigmas should have about 80% seed set in good pollination conditions.

Allogamous species

Some wild *Oryza* species of the sativa group are partially or completely allogamous. Oka and Morishima (1967) gave a natural outcrossing rate of 20-45% for *O. perennis* (asiatic type). Sakai and Narise (1959) observed an outcrossing rate of 7-50% among different strains of *O. sativa* f. *spontanea. O. longistaminata,* a wild species from Africa, is the most allogamous of all *Oryza* species. Some populations of self-incompatible plants depend on outcrossing for seed multiplication.

The main floral traits of these allogamous species are

- large and feathery stigmas (2.5 mm long for *O. longistaminata)* which remain outside the glumes after anthesis;
- very large anthers (5.5 mm) with more than 7,000 pollen grains/anther in some strains of *O. longistaminata* (Oka and Morishima 1967);
- very good panicle exsertion;
- pollen with high fertility duration, up to 9 min for *O. perennis* (Oka and Morishima 1967); and
- pollen emission lasting up to 9 min after flower opening.

Chinese hybrid seed production techniques

China is the only country in the world where farmers use hybrid rice technology on a large scale. To produce hybrid seeds, male and female parents are planted alternately in specific row ratios; the most common ratios are 1:6 and 1:8. Because the natural rate of outcrossing on male sterile plants is insufficient for economic hybrid seed production, the Chinese promote cross-pollination by

- planting across the wind direction to increase pollen dispersal on female plants;

- clipping the flag leaves of male sterile and restorer lines at booting to facilitate pollen circulation;
- applying 20-30 ppm gibberellin to male sterile or restorer lines at initial heading to promote emergence of the basal part of panicle from the leaf sheath;
- supplementing pollination by rope pulling or rod shaking every 30 min during flowering.

Flower synchronization of male and female plants is basic to obtaining a high seed yield. Two planting dates are used for the restorer line to get good synchronization.

With this technique, outcrossing rates range from 15 to 45%, with a maximum 14% reported (Virmani and Tan 1982). Chinese are now getting hybrid seed yields of more than 1 t/ha.

The technique does require a lot of manpower. In China, 1 kg of hybrid seed costs as much as 6-10 kg of conventionally bred varieties. However, only 16-25% of the seeding rate used for conventional varieties is needed for hybrid rice. In countries where manpower is very expensive or in areas where higher seeding rates are necessary, such as in upland rice, this seed production technique is not economically acceptable. Parents perfectly adapted to cross-pollination are needed if hybrid seeds are to be produced for those situations.

Increasing outcrossing rates

Traits that influence outcrossing

Several traits (stigma and anther size, panicle exsertion, aspect of flag leaf) affect outcrossing. All these characters must facilitate pollen dispersal and stigma reception of the pollen grains.

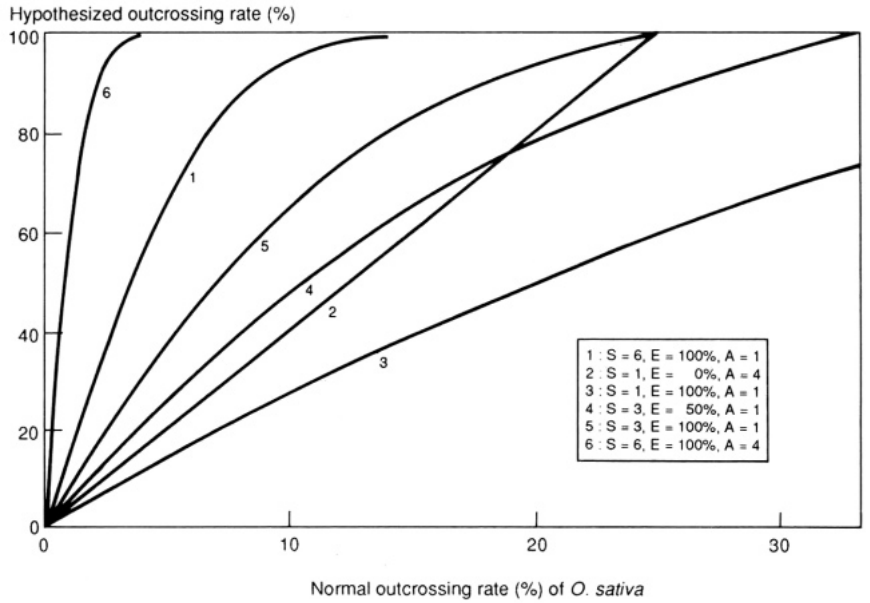
The characteristics that facilitate pollen dispersal are

- many pollen grains per anther;
- long stamen filament and dehiscence of anthers after they are completely outside the glumes;
- pollen with longer capacity of fertilization;
- good male panicle exsertion;
- male parent taller than female parent; and
- small and horizontal flag leaf, which does not interfere with pollen circulation in both parents.

The characteristics that facilitate stigma reception of pollen grains are

- longer and very feathery stigmas, which increase the reception area;
- stigmas that remain outside the glumes after anthesis, increasing the time stigmas are accessible to pollen;
- longer duration of stigma receptivity; and
- good female panicle exsertion and small and horizontal flag leaf, facilitating pollen access to stigma.

Of all the traits influencing outcrossing, stigma characteristics are probably the most efficient in enhancing seed set on male sterile plants. Figure 1 shows hypotheses



1. Theoretical seed set on male sterile plants (based on different hypotheses of stigma area, stigma exsertion, and pollen grains per anther) compared to conventionally bred *O. sativa.* Assumed stigma receptivity period: 3 d. A = pollen grains per anther (1 for *O. sativa,* 4 for *O. longistaminata*); E = stigma exsertion, S = stigma area (1 for *O. sativa,* 6 for *O. longistaminata*).

about the modification of stigma area, stigma exsertion, and number of pollen grains per anther. With a male parent having *O. longistaminata* anthers, a perfect seed set on the male sterile plants is obtainable only when natural outcrossing on normal plants is higher than 25%. With a female parent having *O. longistaminata* stigma, a perfect seed set is obtainable if natural outcrossing on normal plants is higher than 14%.

Selection of plants adapted to outcrossing

The variability in *O. sativa* of nearly all traits that influence outcrossing is very large. There should be no need to resort to wild species to enhance cross-pollination. Using the variability already in *O. sativa* would reduce problems of linkage with undesirable traits and cross-incompatibility and simplify breeding. However, the variability of stigma and anther characteristics in allogamous species is not included in *O. sativa.* As these two traits probably influence outcrossing most, it would be a good alternative to transfer them from wild allogamous species.

The transference of allogamous characteristics from wild species to *O. sativa* was first studied by Virmani and Athwal (l974). The wild species used was *O. sativa* f. *spontanea,* which is cross-compatible with *O. sativa.* They found that shorter anthers and stigmas showed partial dominance. But these wild rice characteristics were linked with genes controlling undesirable wild rice traits. To break the undesirable linkage, a biparental mating approach was proposed.

In Brazil, CNPAF and IRAT are transferring the long stigmas of the wild species *O. longistaminata* to *O. sativa* (indica and japonica types). Two approaches have been used. One is based on successive backcrosses and pedigree selection, the other on recurrent selection. Results so far indicate that

- the *O. longistaminata* stigma is a dominant character;
- the *O. longistaminata* anther is a partially dominant character;
- stigma and large anther are linked characters. No selection pressure was applied for large anthers, although nearly all lines with large stigmas had large anthers. Even so, anthers in the backcross are always smaller than those of *O. longistaminata;*
- perfect *O. longistaminata* stigmas occur in less than 10% of the plants of a backcross. In F_2 generation of a backcross plant with large stigmas, the rate is 5-10%.
- all plants in BC_1 and BC_2 with *O. longistaminata* stigmas are partially or completely male sterile and seed shedding is extremely high, although from BC_3 on, there are no sterility problems and seed shedding decreases.

The use of backcrosses and pedigree selection is expected to result in A and B lines (indica and japonica) with *O. longistaminata* stigmas, by 1987.

Transforming maintainer varieties in A and B lines with *O. longistaminata* stigmas is time-consuming and of low utility, because only a few of the transformed varieties would be used as parents of commercial hybrids. For now, the female parent will be extracted directly from a segregating population where all plants are maintainers with good stigmas. A male sterile gene will be introduced in the population. Such a population will be bred through a process of recurrent selection. To assure a high level of recombination and to maintain selection pressure for outcrossing aptitude, only open-pollinated seeds on male sterile plants will be harvested. After several cycles of selection, the plants should have the *O. longistaminata* stigma character with no undesirable linkages.

The other important favorable traits for outcrossing, such as flag leaf aspect and panicle exsertion, are easily found in *O. sativa.*

Special attention must be given to the “eui” gene described by Rutger and Carnahan (1981). This recessive gene produces nearly twice-as-long uppermost internodes, with no other effects. This character should facilitate windblown dispersal of pollen onto female plants and would make it possible to use another system of hybrid seed production, in which male and female plants could be mixed in transplanting. Because male parents are taller, it is easy to eliminate them after blooming by pulling a herbicide wick across the field at the level of the male panicles.

References cited

Hoff B J, De La Torre M (1981) Stigma exsertion in rice and its effect on the seed set of male-sterile plants. Pages 240-243 *in* 73rd annual progress report of the Rice Experiment Station, Crowley, Louisiana.

Oka H I, Morishima H (1967) Variations in the breeding system of a wild rice *Oryza perennis.* Evolution 21:249-258.

Parmar K S, Swaminathan M S, Siddiq E A (1980) Variation in reproductive organs of rice with reference to male incompatibility index. Indian J. Genet. Plant Breed. 40(1):262-271.

Rutger J N, Carnahan H L (1981) A fourth genetic element to facilitate hybrid cereal production - a recessive tall in rice. Crop Sci. 21:373-376.

Sakai K I, Narise T (1959) Studies on the breeding behavior of a wild rice (*Oryza rufipogon* and *Oryza perennis*). Inst. Genet. Jpn. 9:64-65.

Sarkar H C, Miah N M (l983a) Extent of natural outcrossing of V20a, a cytoplasmic male-sterile (cms) line in Bangladesh. Int. Rice Res. Newsl. 8(6):7-8.

Sarkar H C, Miah N M (1983b) Local indica rice varieties with desirable floral traits influencing outcrossing. Int. Rice Res. Newsl. 8(5):3.

Silitonga T S (1985) Natural outcrossing of cytoplasmic male-sterile V20a. Int. Rice Res. Newsl. 10(6):8-9.

Taillebois J (1983) New prospects for the production of F_1 hybrid seed: transfer of allogamous characters of *O. longistaminata* A. Chev. to *O. sativa* L. [in French]. Agron. Trop. 38(4):303-307.

Virmani S S, Athwal D S (1973) Genetic variability in floral characteristics influencing outcrossing in *Oryza sativa* L. Crop Sci. 13:66-67.

Virmani S S, Athwal D S (1974) Inheritance of floral characteristics influencing outcrossing in rice. Crop Sci. 14:350-353.

Virmani S S, Khush G S, Yang Rancui (1980) Rice cultivars possessing some desirable floral traits influencing outcrossing. Int. Rice Res. Newsl. 5(4):4-5.

Virmani S S, Tan Zhonghe (1982) Stigma receptivity of some cytoplasmic male-sterile lines of rice. Int. Rice Res. Newsl. 7(6):3.

Yoshida S (1981) Fundamentals of rice crop science. International Rice Research Institute, P.O. Box 933, Manila, Philippines. 269 p.

Notes

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