Manifestation of Male Sterility in Autotetraploids Derived from Diploid Male-Sterile Radishes (*Raphanus sativus L.*)

Hiroshi OGURA and Takeomi ETOH
(*Laboratory of Vegetable Crops*)
(Received for publication August 30, 1974)

Introduction

In vegetable-crop plants, various types of male sterility have been observed by many workers, most part of which being elucidated to be the male sterility controlled either by the simple recessive gene or by the interaction between the gene and the cytoplasm concerning male sterility (Jones *et al.*[10], Katsumata *et al.*[12], Ogura[18], Owen[20]). A new type of cytoplasmic male sterility induced by interspecies crossing was recently added (Pearson[21]), too. On the other hand, the developments of normal pollen grains were occasionally observed in some male-sterile lines when they were grown under a given environmental condition. Those facts seem to suggest that in the manifestation of male sterility, the male sterilities were both unstable and susceptible to some environmental conditions, especially to temperature and nutritional conditions (Barham *et al.*[1], Borchers[2], Dickson[3], Edward[4], Johnson[9], Nieuwhof[16], Pearson[21], Rundefeldt[22]).

It is well known that the tetraploids derived from diploids by chromosome doubling express various morphological or physiological characteristics; such as the delay in growth (Fukushima *et al.*[7], Kihara *et al.*[13], Nagamatsu *et al.*[15], Nishiyama[17], Shinohara[23]), the enlargement in organs or tissues (Francis *et al.*[5], Kadota *et al.*[11], Oka[19], Tamai *et al.*[25]), the increase or the decrease in fertility, the alteration in their components or the tolerance to radiation and so on (Fujii[6], Murakami *et al.*[14], Nagamatsu *et al.*[15], Takasugi *et al.*[24], Tamai *et al.*[25], Tanaka[26]).

In the present studies the authors attempted to make clear the relationship between the manifestation of male sterility and polyploidy, by means of the morphological and cytological observations of the normal and male-sterile plants in diploid and tetraploid conditions.

Materials and Methods

In this experiment were used normal diploid cultivar, Kuroba-riso, and a new male-sterile diploid Japanese radish, *Raphanus sativus* L., (Ogura[18]), which was elucidated to be a type controlled by the interaction between the simple recessive gene and the cytoplasm concering male sterility. The tetraploid plants derived from the diploid lines mentioned above were added, too.

The method used to induce the tetraploid plants was as follows.

The seeds of normal and male-sterile diploid lines were sown in early autumn in 1973 in 9 *cm* clay pots filled with the sterilized soil, and grown under the glasshouse condition. A part of those was treated with 0.2 % aqueous colchicine solution by means of dropping on the growing points of young seedlings. At the stage of about five leaves they were set in the field. In the following spring, the pollen grains and pollen mother cells were observed microscopically and then diploid and tetraploid were determined.

Results and Discussion

About one third of the diploid seedlings treated with colchicine solution turned into tetraploid (Table 1). The tetraploid plants were relatively easy to be distinguished from the original diploid lines by their external characteristics ; such as the enlargement of flower size, the thickening and the stiffening in the leaves. However, in this

Table 1. Pollen fertility and numbers of tetraploid plants derived from diploid lines by treatment with colchicine solution.

Lines treated	No. of seedlings treated	No. of diploids	No. of tetraploids	pollen fertility	
				fertile	sterile
ms No. 15	20	13	7	0	20
ms No. 21	19	15	4	0	19
Normal 7	20	12	8	20	0

* ms male-sterile

experiment, tetraploid was determined finally by the cytological observation of pollen mother cells (Fig. 1). The size of flower in the tetraploid radishes reported here was larger than that of the original diploid lines, without reference to fertility or sterility in their pollen grains (Fig. 1). In addition to common morphological characteristics the autotetraploid plants show various specific characters, especially decrease in pollen fertility (Fukushima *et al.*[7], Iizuka [8], Nagamatsu *et al.*[15], Shinohara [23]).

The tetraploid plants derived from a new diploid male-sterile line by the colchicine treatment constantly manifested male sterility as if they were diploid plants. On the other hand, all of the tetraploid plants derived from the normal dipolid plants produced usually fertile pollen grains, though their size and the stainability by iron aceto-carmine lacked uniformity a little. Such situation was maintained through the flowering season, but there was no change in the manifestation of pollen fertility in each plant. Those results seem to suggest that the male sterility reported here was very stable and was not to be affected by polyploidy or environmental conditions for the manifestation of male sterility.

The visual states of pollen grains in the tetraploid plants were similar to the cases reported by many workers in various plants. Namely, the size of pollen grains in the normal tetraploid radishes was slightly large, showing in diameter about 1.25 times that of the original diploid radishes.

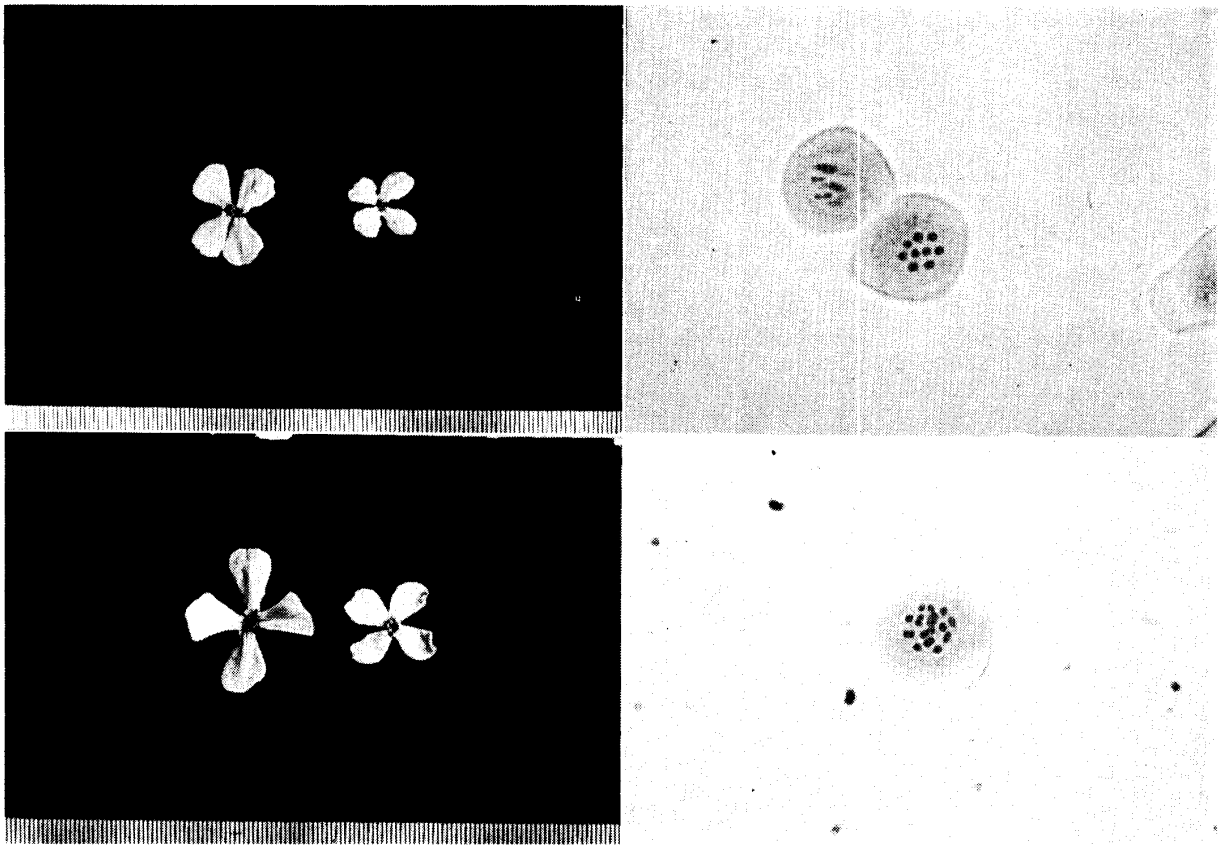

Fig. 1. Comparison of tetraploid flowers and PMCs at first metaphase with those of diploids.

Upper parts are diploid flowers (left is normal, right is male-sterile) and PMCs at first metaphase.
Lower parts are tetraploid flowers (left is normal, right is male-sterile) and PMC at first metaphase.

Summary

Tetraploids of male-sterile and normal plants were derived from each diploid line by the treatment with 0.2 % aqueous colchicine solution, and the relationship between the manifestation of male sterility and polyploidy was observed.

The sizes of flowers and pollen grains in tetraploid plants were a little larger than those of diploid plants. The tetraploid male-sterile plants derived from diploid male-sterile lines manifested constantly male sterility as if they were diploid plants, showing no change at all in the manifestation of male sterility throughout the whole flowering season.

References

1) Barham, W. S. and Munger, H. M. : *Proc. Amer. Soc. Hort. Sci.*, **56**, 401-409 (1950).
2) Borchers, E. A. : *J. Amer. Soc. Hort. Sci.*, **96**, 542-543 (1971).
3) Dickson, M. H. : *J. Amer. Soc. Hort. Sci.*, **95**, 13-14 (1970).
4) Edward, J. R. : *Proc. Amer. Soc. Hort. Sci.*, **83**, 585-589 (1963).

5) Francis, R. R. and Bemis, W. P. : *HortScience,* **9**, 138 (1974).
6) Fujii, T. : *Jap. J. Breed.,* **9**, 253 (1959).
7) Fukushima, E., Tokumasu, S. and Oguro, E. : *J. Hort. Assoc. Japan.,* **18**, 121–128 (1949).
8) Iizuka, M. : *Bull. Res. Inst. Food Sci. Kyoto Univ.,* **7**, 19–25 (1951).
9) Johnson, A. G. : *Nature,* **182**, 1523 (1958).
10) Jones, H. A. and Emsweller, S. L. : *Proc. Amer. Soc. Hort. Sci.,* **34**, 582–585 (1935).
11) Kadota, T. and Ito, K. : *J. Hort. Assoc. Japan.,* **18**, 57–61 (1949).
12) Katsumata, K. and Yasui, H. : *Bull. Hort. Res. Station Minist. Agri. and Forest*, **D-3**, 79–112 (1965).
13) Kihara, H. and Nishiyama, I. : *Rep. Kihara Inst. Biol. Res.,* **3**, 93–104 (1947).
14) Murakami, K. and Mizushima, U. : *Jap. J. Breed.,* **1**, 76–80 (1951).
15) Nagamatsu, T., Anezaki, Y., Kizakihara, C. and Nakamura, M. : *Jap. J. Breed.,* **1**, 172–178 (1952).
16) Nieuwhof, M. : *Euphytica,* **17**, 265–273 (1968).
17) Nishiyama, I. : *Rep. Kihara Inst. Biol. Res.,* **3**, 104–118 (1947).
18) Ogura, H. : *Mem. Fac. Agr. Kagoshima Univ.,* **6**, 39–78 (1968).
19) Oka, H. T. : *J. Hort. Assoc. Japan.,* **11**, 186–202 (1940).
20) Owen, F. V. : *J. Agr. Res.,* **71**, 423–440 (1948).
21) Pearson, O. H. : *J. Amer. Soc. Hort. Sci.,* **97**, 397–402 (1972).
22) Rundefeldt, H. : *Zeits. Pflanzenzuchtg.,* **44**, 30–62 (1961).
23) Shinohara, S. : *J. Hort. Assoc. Japan.,* **17**, 73–76 (1948).
24) Takasugi, K. and Futatsudera, T. : *Jap. J. Breed.,* **9**, 145–153 (1959).
25) Tamai, T., Tokumasu, S. and Yamada, K. : *Jap. J. Breed.* **13**, 143–148 (1963).
26) Tanaka, M. : *Rep. Kihara Inst. Biol. Res.,* **4**, 59–65 (1950).