

Contributions of the Haploid Method to Plant Breeding

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Introductory remarks

Development in methods of inducing haploids with a considerable frequency will help us to use haploids in breeding, as homozygotes can be produced immediately after doubling the the chromosomes of haploids. The senior author has been advocating adoption of what is called "the haploid method" in plant breeding since the idea occurred to him in his studies regarding the induction of haploid plants. Encouragingly enough, a considerable amount of results concerning the haploid method have been achieved by many investigators in the practical field of plant breeding. Here the progress of the haploid method will be outlined.

Proposal of the haploid method

Katayama (1934) obtained many haploids in *Triticum monococcum* with a frequency of as high as 17.6% by pollinating normal stigmas wity x-rayed pollen. This frequency was the highest among those which had so far been attained in experimental induction of haploids. After that, he advanced his haploid method in plant breeding (Katayama, 1950), and then advocated the significance of pollen culture as a means of producing haploids in a review about haploidy in plants (Katayama and Nei, 1964). In the same year, Guha and Maheshwari (1964) preliminarily reported success in anther culture in India, implying successful pollen culture; their conclusive results were reported two years later (Guha and Maheshwari, 1966). Encouraged by these works, Tanaka and Nakata (1967, 1968, 1969) achieved production of haploids of tobacco by anther culture and moreover succeeded in doubling the chromosomes of the haploids by means of the colchicine method (Blakeslee and Avery, 1937). Reviewing these works, Katayama and Tanaka (1969) confidently proposed again tne haploid method in plant breeding.

Methods of haploid production

Ever since the haploid plant was first found in *Datura stramonium* (Blakeslee

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et al., 1922), haploids have been recognized in many different plant species, though their occurrences are rare in nature. Katayama and Nei (1964) enumerated the following methods for increasing the frequency of occurrence of haploids: (1) intergeneric or interspecific hybridization; (2) pollination with X-rayed pollen; (3) delayed pollination; (4) screening of haploids among twin seedlings; (5) selection for appropriate genotypes; and (6) utilization of alien cytoplasms. Of these methods of artificial production of haploids, pollination with X-rayed pollen (Katayama, 1934), delayed pollination (Kihara, 1940), and cytoplasm substitution (Kihara, and Tsunewaki, 1962) were especially effective in haploid production. In addition, Kasha et al. (1970) and others succeeded in raising haploids in barley using a unique phenomenon of chromosome elimination that occurs in culturing embryos of certain interspecific hybrids. Nevertheless, it might be anther culture, as described in the next section, that best assures to produce a large number of haploids constantly.

Success in anther culture

At the Hatano Tobacco Experiment Station, Japan, Tanaka and Nakata (1967, 1968, 1969) succeeded in the anther culture of tobacco to produce a large number of haploids and in reconstitution of homozygous diploids from the anther-derived haploids through chromosome doubling using the colchicine method. These works marked an epoch in showing the possibility of the practical application of the haploid method. Applying this method to the practical tobacco breeding, they actually produced an excellent variety at the Iwata Tobacco Experiment Station, Japan. Successes in haploid production by anther culture in tobacco were also reported in France (Bourgin and Nitsch, 1967; Nitsch, 1969; and others). In this respect, Niizeki (1970) reviewed anther cultures from a standpoint of plant breeding, and the Agriculture, Forestry and Fishery Council, Japan (1974) compiled the materials concerning anther culture techniques by the help of various research institutes and experimental stations concerned.

Achievement of the haploid method in tobacco breeding

Based on the haploid method which was first realized in tobacco by Drs. Tanaka and Nakata, a plan of tobacco breeding was undertaken at the Iwata Tobacco Experiment Station and produced excellent results, leading to an outstanding commercial variety. The results were reported in a series of papers from the first (Kadotani, 1969) to the fifth (Nakamura et al., 1975) which reported the success of the haploid method. They employed the haploid method in improving the resistance to bacterial wilt of leading tobacco varieties in Japan with mild smoking quality, and finally, produced an excellent improved variety, F211 which showed a satisfactory adaptability in the locality test and has come to be widely cultivated (Nakamura et al., 1974). It can

well be said that this successful application of the haploid method in tobacco breeding is really a frontier achievement of modern breeding and is credited to Japan.

Application of the haploid method to rice and other crops

Following the success of Drs. Tanaka and Nakata in the anther culture of tobacco, Niizeki and Oono (1968, 1971) obtained comparable results in the anther culture of rice. Thereafter, anther culture was attempted in various crops such as *Brassica* (Kameya and Hinata, 1970), *Solanum* (Irikura and Sakaguchi, 1972), *Hordeum* (Clapham, 1973), and others. As to the utilization of haploids in plant breeding, Melchers (1972) also referred to the issue, and particularly, in China (Ouyang et al., 1973; Institute of Genetics (Academia Sinica), 1974; and others), the haploid method seems to have been generalized in plant breeding by the name of pollen plant breeding (cf. Niizeki, 1976).

Closing remarks

It is our great pleasure to see that the haploid method, which was originally proposed by the senior author (Katayama) has come into practical use. Although the haploid method does not seem to be applicable in breeding all crops, even limited application would be a reward to the study of the haploid method. Further development of the haploid is desired.

Literature Cited

- Agriculture, Forestry and Fishery Council (Japan) 1974. Studies on the performance in anther culture technique. (Japanese). Results No. 76: 1-127.
- Blakeslee, A. F. and Avery, A. G. 1937. Methods of inducing chromosome doubling in plants. *Journ. Hered.* **28**: 393-411.
- Blakeslee, A. F. Belling, J., Farnham, M. E. and Bergner, A. D. 1922. A haploid mutant in the Jimson Weed, *Datura stramonium*. *Science* **55**: 646-647.
- Bourgin, J. P. and Nitsch, J. P. 1967. Obtention de *Nicotiana* haploides à partir d'étamines cultivées in vitro. *Ann. Physiol. vég.* **9**: 377-382.
- Clapham, D. 1973. Haploid *Hordeum* plants from anthers in vitro. *Zeit. Pflanzenzüchtg.* **89**: 142-155.
- Guha, S. and Maheshwari, S. C. 1964. In vitro production of embryos from anthers of *Datura*. *Nature* **204**: 497.
- and — 1966. Cell division and differentiation of embryos in the pollen grains of *Datura* in vitro. *Nature* **212**: 97-98.
- Institute of Genetics (Academia Sinica) 1974. Investigation on the induction and genetic expression of rice pollen plants. *Scientia Sinica* **17**: 209-222.
- Irikura, Y. and Sakaguchi, S. 1972. Induction of 12-chromosome plant from anther culture in tuberous *Solanum*. *Potato Research* **15**: 170-173.
- Kadotani, N. 1969. Studies on the haploid method of breeding by anther culture in tobacco I. Production of diploid plants from haploid by pith and root tissue culture. *Bull.*

- Iwata Tobacco Exp. Sta. No. 2: 73-77.
- Kasha, K. J. and Kao, K. N. 1970. High frequency haploid production in barley (*Hordeum vulgare* L.). *Nature* **225**: 874-875.
- Kameya, T. and Hinata, K. 1970. Induction of haploid plants from pollen grains of *Brassica*. *Japan. Journ. Breed.* **20**: 82-87.
- Katayama, Y. 1934. Haploid formation by X-rays in *Triticum monococcum*. *Cytologia* **5**: 235-237.
- 1950. Studies on the haploidy in relation to plant breeding I. Introductory contribution. *Bull. Miyazaki Univ. (Nat. Sci.)* No. 1: 28-30.
- Katayama, Y. and Nei, M. 1964. Studies on the haploidy in higher plants. *Rep. Lab. Pl. Breed., Fac. Agr., Miyazaki Univ.* No. 2: 1-78.
- Katayama, Y. and Tanaka, M. 1969. Studies on the haploidy in relation to plant breeding V. Further proposal of haploid method in plant breeding. *Seiken Zihō* **21**: 37-44.
- Kihara, H. 1940. Formation of haploids by means of delayed pollination in *Triticum monococcum*. *Bot. Mag. Tokyo* **54**: 178-185.
- Kihara, H. and Tsunewaki, K. 1962. Use of an alien cytoplasm as a new method of producing haploids. *Japan. Journ. Genet.* **37**: 310-313.
- Melchers, G. 1972. Haploid higher plants for plant breeding. *Zeit. Pflanzenzüchtg.* **67**: 19-32.
- Nakamura, A., Yamada, T., Kadotani, N., Itagaki, R. and Oka, M. 1974. Studies on the haploid method of breeding in tobacco. *Sabrao Journ.* **6**: 107-131.
- Nakamura, A., Yamada, T., Oka, M., Tatemichi, Y., Eguchi, K., Ayabe, T., and Kobayashi, K. 1975. Studies on the haploid method of breeding by anther culture in tobacco V. Breeding of mild flue-cured variety F211 by haploid method. *Bull. Iwata Tobacco Exp. Sta.* No. 7: 29-39.
- Nitsch, J. P. 1969. Experimental androgenesis in *Nicotiana*. *Phytomorphology* **19**: 389-404.
- Niizeki, H. 1970. Anther culture and plant breeding. (Japanese). *Recent Advances in Plant Breeding* No. 11: 45-51.
- 1976. Pollen breeding. (Japanese). *Shizen* No. 368: 38-47.
- Niizeki, H. and Oono, K. 1968. Induction of haploid rice plant from anther culture. *Proc. Japan Acad.* **44**: 554-557.
- and ——— 1971. Rice plants obtained by anther culture. *Les Cultures de Tissus Plantes. Colloques Internationaux du C. N. R. S.* No. 193: 251-257.
- Ouyang, T. W., Hu, H., Chuang, C. C. and Teng, C. C. 1973. Induction of pollen plants from anthers of *Triticum aestivum* L. cultured in vitro. *Scientia Sinica* **16**: 79-95.
- Tanaka, M. and Nakata, K. 1967. On the formation of haploid plants. (Speech, Symp. October Meeting Japan, Soc. Breed.).
- and ——— 1968. On the formation of haploid plants. (Japanese). *Recent Advances in Plant Breeding* No. 9: 23-32.
- and ——— 1969. Tobacco plants obtained by anther culture and the experiment to get diploid seeds from haploid. *Japan. Journ. Genet.* **44**: 47-54.