葯培養によるAegilopsの半数性アルビノ植物の育成

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SHORT COMMUNICATION

PRODUCTION OF HAPLOID ALBINO PLANTS OF *AEGILOPS* BY ANTHER CULTURE[1)]

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The genome composition of polyploid species can be reconstructed from chromosome pairing of interspecific or intergeneric hybrids involving the species in question. However, in many cases such estimation is often inconclusive due to the difficulty in distinguishing auto- from allo-syndetic pairing. In doubtful cases chromosome pairing of polyhaploid plants may provide the desirable information (Sakamoto 1964). Recently anther culture as an efficient method for producing haploids in higher plants was practiced first in *Datura* by Guha and Maheshwari (1964), thereafter in *Nicotiana* by Nakata and Tanaka (1968) and in *Brassica* by Kameya and Hinata (1970). However, in monocotyledons induction of haploid plants succeeded so far only with *Oryza* by Niizeki and Oono (1968) and with *Setaria* by Ban *et al.* (1971). The above mentioned authors have devised several ways how to induce polyhaploid plants of complex polyploid species of the tribe Triticeae mainly with the help of anther culture.

The basic medium used in our experiment was Miller's (Miller 1963) which was used for the anther culture of *Oryza* by Niizeki and Oono (*loc. cit.*). It was supplemented with 2.21 mg/*l* 2, 4-D (2, 4-dichlorophenoxyacetic acid). The solution was adjusted to pH 6.0 and medium was solidified with 10 mg/*l* agar.

The following species of *Aegilops* were used by us: *Ae. caudata* L., *Ae. crassa* Boiss. (6x), *Ae. comosa* Sibth. et Sm., *Ae. cylindrica* Host., *Ae. ovata* L., *Ae. speltoides* Tausch, *Ae. squarrosa* L., *Ae. triuncialis* L., *Ae. umbellulata* Zhuk., *Ae. variabilis* Eig and an artificially synthesized amphiploid (designated CCC^uC^u) between *Ae. caudata* and *Ae. umbellulata* (Kondo 1941). The anthers were aseptically planted on the surface of the agar and were incubated in test tubes at 25°C under about 100 lux inflorescent light. Anthers containing pollen grains approximately at uninuclear stage were planted.

About 20 days after planting yellowish calluses started to proliferate from the inside of the anthers. Callus formation occurred only in anthers of CCC^uC^u, an amphiploid between *Ae. caudata* and *Ae. umbellulata*. No callus formation was observed in the anthers of the remaining species employed. From 206 anthers of CCC^uC^u planted in eight test tubes only two calluses were produced, thus the induction rate could be assessed at 1.0 per cent.

80 days after planting the calluses were divided aseptically into two parts. The

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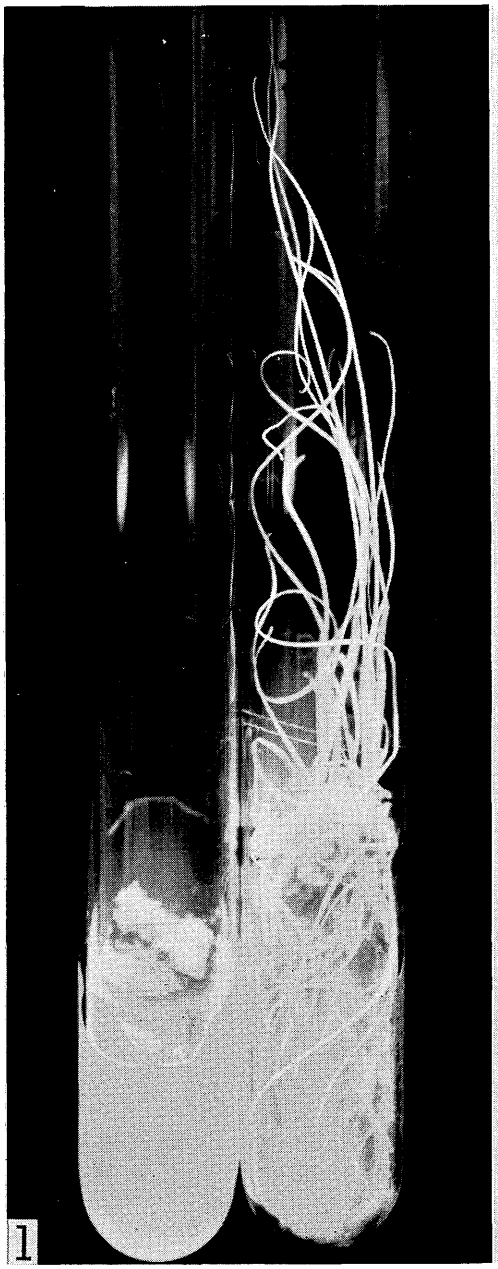

Fig. 1. left: Callus on the basic medium containing 2.21 mg/*l* 2, 4-D.
right: Young haploid plants grown on the basic medium without 2, 4-D.

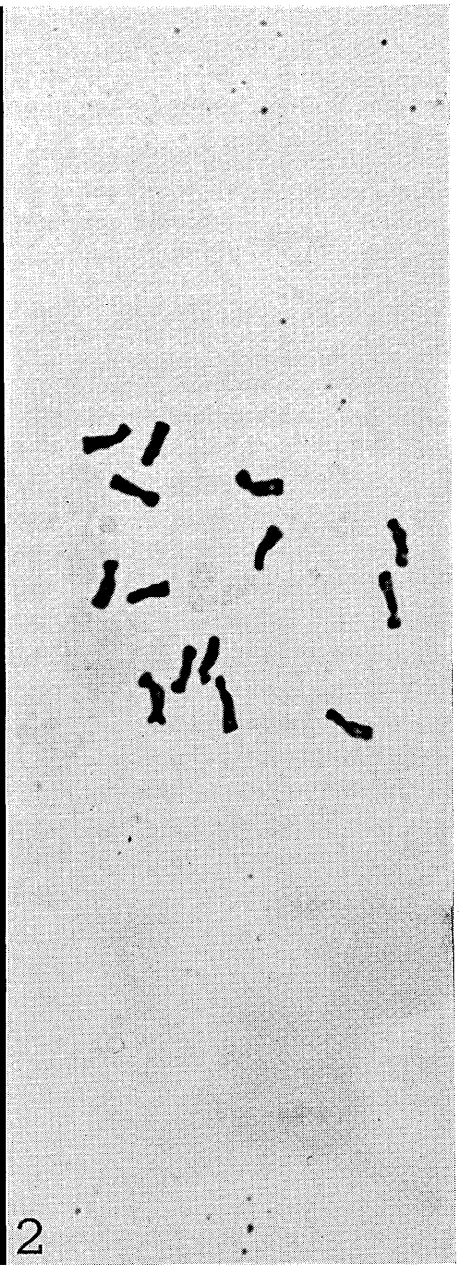

Fig. 2. Haploid chromosome number ($2n=14$) in a root-tip cell of a newly developed plant.

first part was subcultured on the same callus producing medium. They continued to give normal growth (Fig. 1). In order to induce organ formation the second part was transferred to the same medium as mentioned above omitting 2, 4-D. About two weeks after transplanting to this medium sprouting from the callus appeared and many plantlets took shape. All were albino with no trace of chlorophyll formation in coleoptile and leaves (Fig. 1). Since CCC^uC^u is a synthetic tetraploid plant ($2n=28$) derived from two diploid species of *Aegilops*, the root tips were collected from these newly developed plants and their chromosome numbers were examined by the aceto-carmine squash technique. All cells of the albinos were haploid, $2n=14$ (Fig. 2). 117 days after tansplanting an albino plant with seven leaves flowered. However, the spike of this plant included only a single spikelet without pistil and stamens.

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