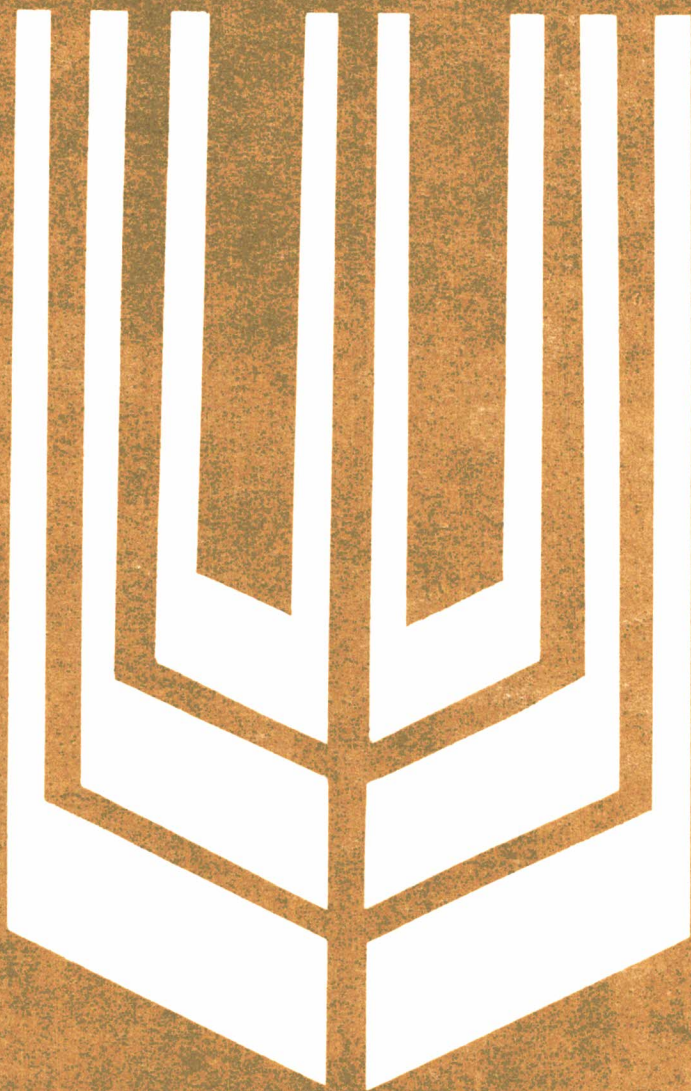


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AMPHIPLOID TRITICUM MONOCOCCUM L. x SECALE CEREALE L. (AARR)  
- A NEW FORM OF TETRAPLOID TRITICALE

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Summary. By colchicine treatment of diploid wheat-rye (T.monococcum x S.cereale) F<sub>1</sub> hybrid the mixoploid plants were obtained. Based on the vegetative propagation of the sectors with doubled chromosomal number and continual selection on the cytological examination basis in their vegetative progeny the stable amphiploid plants with AARR genome formula were produced, as it was impossible to obtain by self-pollination using gametes of plants after colchicine treatment. According to the hitherto existing results there is much higher fertility of these amphiploid plants in cross-pollinations than in self-pollination.

Index words: diploid wheat-rye hybrid, colchicine treatment, amphiploid plants, cross-pollination, self-pollination

#### INTRODUCTION

Many attempts were taken in the past in order to obtain hybrids between diploid wheat and rye. In the last years they were taken again in connection with the aim of analysis of tetraploid level of ploidy of Triticale (Kiss 1960, 1965; Rigin 1965; Chaudry 1968; Krolow 1970, 1974; Moss 1979; CIMMYT Report 1974.). Only Kiss (1960.) succeeded in obtaining two viable seeds from such crosses, but plants have died as a completely sterile.

In similar experiments in Poznań the vigorous hybrid plant has been derived on which functional female gametes were found to occur occasionally (Sodkiewicz 1982.). Attempts have been made to obtain a new tetraploid Triticale form with AARR genome structure by a doubling number of chromosomes of this hybrid.

#### MATERIAL AND METHODS

Clone of plants of F<sub>1</sub>-hybrid T.monococcum var. macedonicum x S.cereale cv. Dankowskie Złote consisted more than 400 individuals have been used. Young plants in tillering stage have been treated with colchicine in two ways:

- by tillers, using cotton wick soaked with 0,4% colchicine solution (by 2-3 days),
- by roots, using the solution of 0,05 or 0,075% colchicine with addition 2 or 3% DMSO and 18 or 5 h of soaking, respectively.

Somatic chromosome number was checked on aceto-orceine squash preparations made from tips of secondary roots formed after colchicine treatment.

The degree of development of the male gametes was estimated on the basis of observations of the anther morphology and using pollen staining with a mixture of acetocarmine and glycerine.

The function of female gametes were revealed as a result of open or hand pollination with pollen varying degree of ploidy. A diploid pollinator in hand made crosses was the rye cv. Pancerne, whereas in free pollination experiment a pollen donor was alloplasmic form of rye cv. Dańskowskie Złote with Triticum timopheevi cytoplasm. As a tetraploid pollinator was used the mixture of Triticale lines with the chromosome number incompletely stabilized and similar to tetraploid. They were obtained at the Institute of Plant Genetics of the Polish Academy of Sciences in Poznań as a result of reduction in the chromosome number of hexaploid Triticale (Żapinski et al. 1980). The third pollinator was the mixture of 6x-Triticale lines from the breeding programme of Plant Breeding Station "Łaski" of the Poznań Plant Breeders kindly supplied by Docent Dr. T. Wolski.

#### RESULTS AND DISCUSSION

223 plants of the clone of F<sub>1</sub>-hybrid were colchicine treated and subjected to pollination in different ways. 109 of them and 147 untreated (control plants) were exposed to free pollination of pollen varying degree of ploidy. Diploid, tetraploid and hexaploid pollen donors (mentioned in detail in material and methods) were used on separate, space-isolated plots. Taken as a whole 75 seeds have been set on colchicine treated plants (tab. 1A). Hybridization of F<sub>1</sub>-hybrid T.monococcum x S.cereale with all pollen donors was successful. Despite of the distinct dependence of the results on the cross combination colchicine treatment have increased the functionality of female gametes as compared to untreated material. Paralelly, on the same plots, some spikes of the colchicine treated plants with well developed anthers were isolated with paper bags to be selfed. Opposite to results with alien pollen no seed have been obtained on selfed spikes.

On the second part of colchicine treated material growed in greenhouse or plastic tunnel and subjected to hand pollination of spikes the very similar results have been obtained (tab. 1B). Generally, results of seed setting were strongly dependent on a pollen donor. Pollinations with Triticale pollen were relatively more effective than with rye. The most suitable pollen parent was 6x-Triticale. In contrast to it all attempts to selfing plants were unsuccessful.

The spikes used for selfing were specially selected and most of them have had spontaneously dehiscent anthers. The pollen staining in such anthers with a mixture of acetocarmine and gly-

Table 1. Results of pollinations of dihaploid and mixoploid plants of  $F_1$ -hybrid T. monococcum x S. cereale in different pollen conditions.

Pollen donor	Dihaploid plants		Mixoploid plants /after colchicine treatment/	
	Number of pollinated spikes flowers	Seeds % poll. flowers	Number of pollinated spikes flowers	Seeds % poll. flowers
A. Free pollination				
$F_1$ T.monoc. x S.cereale <sup>+</sup>	—	—	36	2900
Secale cereale 2x=14	86	0	71	2564
Triticale 4x=28	126	9	445	27982
Triticale 6x=42	107	3	27	1212
				16
				0,00
				0,08
				0,20
				1,32
B. Hand-made pollinations				
$F_1$ T.monoc. x S.cereale <sup>+</sup>	—	—	31	1565
Secale cereale 2x=14	86	0	24	1632
Triticale 4x=28	126	9	42	3110
Triticale 6x=42	107	3	29	1756
				82
				0,00
				0,12
				3,21
				4,67
+ -- mixoploid plants				



cerine has revealed from 48 to 79 % well developed grains. Subsequent additional pollination some of these spikes with alien pollen resulted in hybrid seeds, showing partial female fertility of florets.

In these circumstances new attempts have been made to obtain amphiploid plants with AARR genome pattern in a vegetative way, based on the cytological analysis of mixoploid plants after colchicine treatment and the vegetative propagation of the sectors with doubled chromosomal number.

The results of pollination of the vegetative offspring obtained from mixoploid plants of the F<sub>1</sub>-hybrid have revealed the female fertility the same as in untreated plants, indicating absence of the sectors with doubled chromosome number. This fact indicates the existence of strong competition between cells with different number of chromosomes inside chimeric plants and natural selection processes directed against the sectors with doubled number of chromosomes.

In order to produce stable plants with 28 chromosomes in all cells efforts have been made with the newly prepared 38 mixoploid plants to overcome natural selection processes inside chimeras via artificial selection of stems. For this purpose, plant shoots of the F<sub>1</sub>-hybrid T.monococcum x S.cereale have been colchicine treated using cotton wick soaked with colchicine. Physiological reaction of different tillers on colchicine was observed. On the basis of this observations 11 most convenient plants after regeneration were selected. These plants were partitioned into tillers and those, which showed reaction on colchicine were selected and planted. Somatic chromosomal number have been counted in tips of secondary roots. The tillers with 28 chromosomes were cultivated into plants and vegetatively propagated into clones. The chromosome numbers of plants belonging to one clone were checked again. As a result of such work two hybrid plants with 2n=28 chromosomes, which in all subsequent partitions have given with no exception shoots with the same number of chromosomes, have been obtained.

Preliminary data on the fertility of spikes of the 28-chromosomal plants are given in table 2. Seed setting have increased markedly. However, the results showed generally the same dependence on the pollen parent as the observed before in mixoploid plants. The best results were obtained with 6x-Triticale pollen. The mean percent of seed setting in case of pollination with alien pollen was 11,32 %. In contrast to the results with alien pollen no seed have been obtained from the self-pollinated flowers. In repeated experiments, using different times of pollination and different times of flowering we were able to obtain a few seeds, but nevertheless still the general rule exists that in self-pollination fertility is markedly lower than in cross-pollination with any kind of Triticale pollen. Some examinations have been undertaken to explain the reason of this phenomena.

This 28-chromosomal plants in their qualitative morphological characters, especially the shape of spikelets and florets, strongly remind morphological features of the F<sub>1</sub>-hybrid T.mono-

Table 2. Preliminary results of fertility on amphiploidal plants /AARR/ T. monococcum x S. cereale with different pollen donors.

Clon	Pollen donor	● Number of spikes	Number of pollinated flowers	Number of seeds	Seeds % poll. flowers
A1	2x S. cereale	12	1010	125	12,38
A2	2x S. cereale	10	862	78	9,05
Σ A1+2	2x S. cereale	22	1872	203	10,84
A2	+ 4x Triticale	1	66	6	9,09
A1	6x Triticale	6	511	88	17,22
A2	6x Triticale	7	558	43	7,71
Σ A1+2	6x Triticale	13	1069	131	12,25
A2	8x Triticale	2	111	13	11,71
Σ A1+2	all pollen donors	38	3118	353	11,32
A1	self	2	95	0	0,00
A2	self	3	230	0	0,00
Σ A1+2	self	5	325	0	0,00

coccum x S.cereale (Sodkiewicz 1982.), but their quantitative characters are evidently different. These plants are taller, stems thicker and larger leaves and spikes than in the F<sub>1</sub>-hybrid plants with 2n=14 chromosomes. Consequently they seem to be amphidiploidal tetraploids with the genomes AARR. Giemsa C-banding of chromosomes have confirmed such a chromosomal constitution. According to this genome pattern the above mentioned amphiploidal plants should be considered as the new tetraploid form of Triticale, different in many cytogenetical aspects from obtained earlier 4x-Triticale (Krolow 1974.).

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