STUDIES IN POLYPLOID VARIETIES OF SUGAR BEETS

XIV. Use of Cytoplasmic Male Sterility in the Production of Triploid Hybrids, and their Performance in Trials[1)]

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Introduction

Recently the cytoplasmic male sterility is effectively used for the seed production of commercial hybrid varieties and triploid varieties in sugar beets (McFARLANE *et al.* 1961, BANDLOW 1964). Triploid hybrids with a high combining ability indicate a superiority to parental diploid and tetraploid plants in their gross sugar productions (MATSUMURA 1953, KNAPP 1956, HELMERICK *et al.* 1965).

The seeds of pure triploids are efficiently produced by crossing diploid male sterile plants with tetraploid pollinators or by reciprocal crossing, i. e. between tetraploid male sterile plants and diploid pollinators.

The present investigation deals with the application of the cytoplasmic male sterility both at diploid and tetraploid levels, in relation to the production of triploid hybrids.

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Materials and Methods

Diploid male sterile strains were produced from the crossing between an introduced strain with the cytoplasmic male sterility and a Japanese variety, Hon-iku 192. Tetraploid male sterile strains were induced from M-strains by

1) Contribution from the Plant Breeding Institute, Faculty of Agriculture, Hokkaido University, Sapporo, Japan.

colchicine treatment and then this trait was transferred into a tetraploid normal strain, 4192. The male sterile types which were contained in the male sterile strains were classified into complete sterile (C. S.), semi-sterile type-b, (S. S. b), semi-sterile type-a (S. S. a) and normal type (N) based on the classification which was used in previous reports (NAGAO and KINOSHITA 1962).

Trial seeds were produced from crossings between normal diploid and tetraploid plants and from crossings by use of diploid or tetraploid male sterility. Normal tetraploids, H-4002, and normal diploids, H-2002, were mix-planted in a ratio of 3 : 1 and the seeds were harvested separately from diploids and tetraploids (Cross combination A). Tetraploid male sterile strains, 4M-59 and 4M-60 which were produced by the crossing with the plants randomly chosen from the normal strain 4192, and which consisted of male sterile plants in excess of 90%, were mix-planted with normal diploid plants of H-2002 in a ratio of 3 : 1, and the seeds were obtained from tetraploid male sterile plants (Cross combination B, C and D). Diploid male sterile strains, M-14, M-35 and M-41 which were produced by the crossing with the pollinators randomly chosen from Hon-iku-192 and which consisted of lower frequencies of male sterile plants (57-78%), were mix-planted with normal tetraploids, H-4002 or 4192 in a ratio of 3 : 1 and the seeds were obtained from diploid male sterile plants (Cross combination E, F and G).

Frequency of diploid, triploid and tetraploid individuals in the eight populations was examined in the seedling stage by counting the chromosome numbers. Germination tests were carried out in the greenhouse by using the eight triploid hybrids together with the parental strains of diploid and tetraploid.

In 1960 and 1961, yield trials were conducted at the experimental field of the Hokkaido University in Furano, which is located in the central part of Hokkaido. The strains used in the experiments are listed in Table 1. The triploid hybrids which were produced experimentally by using diploid and tetraploid male sterility as described in the above were planted together with their parental diploid and tetraploid strains. The experimental design was a randomized block with four replications. The plot size was 5 m^2 and spacings were 50 cm between rows and 23 cm between plants within rows. The seeds were planted on May 10th and the plants were harvested on October 18th in 1960. Farming and application of fertilizer were done in the usual manner. In the middle of the growth period (July 28th), 20 plants were chosen in each strain for a survey of plant height, length and width of leaf blade and number of leaves. Yield of roots and sugar contents were investigated after harvesting and the data were treated with variance analysis. In 1961, two introduced

TABLE 1. List of strains used in yield trial

No.	Name of strain	Ploidy[1]	Type of cytoplasm	Description
1	Hon-iku-192	2X	N	Japanese standard variety
2	M-16	2X	S	Diploid male sterile strain
3	H-2002	2X	N	A selection from Donyu-2go
4	H-4002	4X	N	Tetraploid strain produced from H-2002
5	4192	4X	N	Tetraploid strain produced from Hon-iku-192
6	4M-50	4X	S	Tetraploid male sterile strain
7	H-4002×H-2002	3XA,[2] (4X)	N	Triploid hybrid produced from 4X by mix-planting of 2X and 4X
8	H-2002×H-4002	3XB,[3] (2X)	N	Triploid hybrid produced from 2X by mix-planting of 2X and 4X
9	4M-50×H-2002	3XA	S	Pure triploid hybrid produced with tetraploid male sterility
10	4M-59×H-2002	3XA	S	ditto
11	4M-60×H-2002	3XA, (4X)	S	Almost pure (97%) triploid hybrid, produced with tetraploid male sterility
12	M-14×H-4002	3XB, (2X)	S	Impure triploid hybrid produced with diploid male sterility
13	M-35×4192	3XB, (2X)	S	ditto
14	M-41×H-4002	3XB, (2X)	S	ditto
15	Polyrave	3X (2X, 4X)	N	Polyploid variety introduced from Netherlands
16	Trirave	3X (2X)	S	Triploid variety introduced from Netherlands

1) Parenthesis means that 4X and/or 2X is involved in 3X population.
2) Triploids from tetraploid mother plants.
3) Triploids from diploid mother plants.

varieties, 'Polyrave' (mixed population with diploids, triploids and tetraploids) and 'Trirave' (triploid hybrid produced with diploid male sterile plants) were joined newly in the trial. The same design and procedure were used for this experiment likewise. However, the seeds were sown on May 5th and the plants were harvested on November 1st. Plant height and number of leaves were first surveyed on June 27th at the middle stage of growth and then measured on July 21st at the maximum growth of the leaves. The susceptibility to the leaf spot disease by *Cercospora* was recorded by the degree of injury with visual observation before the harvest.

Experimental Results

A. Frequency of male sterile types in the male sterile strains

Under the digenic assumption on the pollen restoring genes for the sterility-inducing cytoplasm, a special pollen parent with the genotype, $N\ xx\ zz$ (designated as type-O) is desirable when crossed with the complete sterile plants with the genotype, $S\ xx\ zz$. However the frequency of the type-O plants was estimated to be relatively low percentages in various populations (KNAPP 1955, NAGAO and KINOSHITA 1962, IMANISHI 1968). Therefore, the authors tentatively used the male sterile strains which were crossed with the pollinators randomly chosen from normal varieties.

The frequency of the male sterile types in M- and 4M-strains are shown in Figure 1. Tetraploid male sterile strains, 4M–50, 4M–59 and 4M–60 indicated higher percentages of practical male sterile plants (C.S. and S.S.b) than in diploid male sterile strains, M–14, M–35 and M–41.

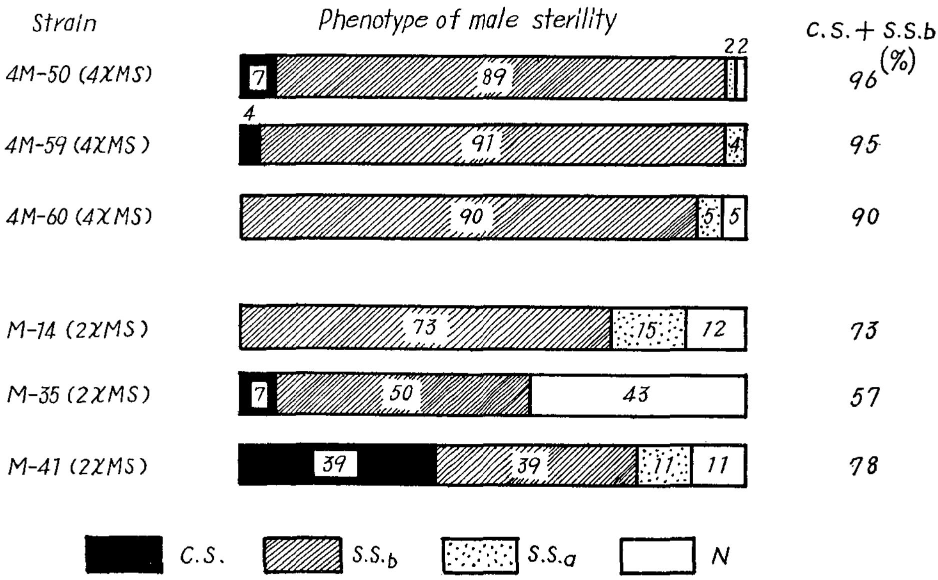

Figure 1. Phenotypic ratios of male sterile type in the male strile strains.

Another series of cross experiments was conducted with the semi-sterile type-b plants, too. Semi-sterile type-b plants in diploids and tetraploids were crossed with normal plants randomly sampled from Hon-iku-192, at diploids, 4192 and 4398, at tetraploids. Though the segregation mode in progenies is different depending upon the genotypes in S.S.b plants which was used as

TABLE 2. Phenotypic ratios of male sterility in the progenies of the crosses between S.S.b type and pollinators (N cytoplasm)

Ploidy	Cross combination	Percentage of male sterile type				Number of plants
		C.S.	S.S.b	S.S.a	N	
2X	K-3-11 (S.S.b)×192	9	68	18	5	22
	〃 -12 (〃)× 〃	2	81	14	4	57
	〃 -16 (〃)× 〃	26	26	40	10	20
	Mean	12.3	58.3	24.3	6.3	—
	M-8 (S.S.b)×192	8	78	11	3	36
	〃-21 (〃)× 〃	0	93	7	0	15
	〃-30 (〃)× 〃	5	70	10	15	20
	〃-32a (〃)× 〃	27	73	0	0	11
	〃-32b (〃)× 〃	15	65	8	12	26
	〃-32c (〃)× 〃	0	35	25	40	20
	Mean	9.2	68.0	10.0	11.7	—
4X	4M-48 (S.S.b)×4398	0	94	0	6	16
	〃 〃 ×4192	3	80	3	13	61
	4M-50 〃 ×4398	7	89	2	2	135
	〃 〃 ×4192	0	89	11	0	37
	〃 〃 ×4192*	5	76	7	13	128
	4M-59 〃 ×4398	4	91	4	0	45
	4M-60 〃 × 〃	0	90	5	5	21
	Mean	2.7	87.0	4.6	5.6	—

* Observation in different populations.

female parents, the total average frequency of C.S. and S.S.b was more than 70% in diploids and about 90% in tetraploids (Table 2). These results show that even if plants of random samples from the above varieties are used as the pollinators, a relatively high percentage of C.S. and S.S.b types which are safe in pollen shedding can transmit to progenies through mother plants of complete sterile and semi-sterile type-b especially at tetraploid level. However in a case where it is desired to obtain 100% incidence of male sterile plants, the type-O plants should be selected and employed as the pollinator.

B. Triploid seed production by the use of male sterility

In the trial seed production by the use of male sterility, seed productivity of parental diploids and tetraploids are as shown in Table 3. There were no

TABLE 3. Seed productivity of diploid and tetraploid parents for the production of the triploid seeds

Cross combination	Ploidy	Plant height cm	Number of primary branches	Seed setting %	Seed weight per plant g	Number of plants
A. H-4002 (4X) ×	4X	124.3	11.0	73.1	59.6	9
H-2002 (2X)	2X	127.8	16.5	89.9	116.0	4
B. 4M-50 (4XMS) ×	4X	130.6	12.0	79.8	49.8	4
H-2002 (2X)	2X	129.5	15.3	95.8	42.0	4
C. 4M-59 (4XMS) ×	4X	126.1	13.8	86.5	36.1	12
H-2002 (2X)	2X	134.8	13.3	86.1	25.8	4
D. 4M-60 (4XMS) ×	4X	108.1	10.0	83.9	67.3	9
H-2002 (2X)	2X	111.7	11.0	74.4	113.0	3
E. M-14 (2XMS) ×	4X	135.5	8.0	82.3	8.5	2
H-4002 (4X)	2X	148.2	18.9	85.7	52.3	10
F. M-35 (2XMS) ×	4X	111.5	11.0	77.0	22.5	2
4192 (4X)	2X	136.2	17.0	92.2	70.2	6
G. M-41 (2XMS) ×	4X	127.5	10.0	68.5	5.0	2
H-4002 (4X)	2X	122.1	13.3	76.7	18.6	8
Mean	4X	123.4	10.8	78.7	35.5	
	2X	130.0	15.0	85.8	62.6	
4X-2X		−7.4	−4.2	−7.1	−27.1*	

* Significant at 5% level.

significant differences between diploids and tetraploids as to plant height, number of primary branch and percentage of seed setting, while seed weight per plant in tetraploid is significantly lower than that in the diploid counterpart, having nothing to do with the male sterility.

In seed fertility and germination rate, triploid hybrids from the various combinations showed a variation over a wide range (Table 4). There is a possibility that the nature of parental tetraploids, the condition of an isolated field and a discrepancy of flowering time between diploids and tetraploids affect the quality of hybrid seeds. A slight delay of germinating date and decrease of seedlings per seed ball were observed in a tetraploid strain and the triploid

TABLE 4. Germination test of various triploid hybrids and their parents

Strain or combination	Ploidy	Number of germs per seed ball	Number of fertile germs per seed ball	Seed fertility %	Germination rate %		Average date of germination		Average number of seedlings per seed ball	
					Sand	Soil	Sand	Soil	Sand	Soil
H-2002	2X	3.1	2.6	83.2	88	86	4.8	4.8	3.1	2.3
H-4002	4X	2.3	0.9	39.7	88	76	6.6	5.2	1.3	1.2
H-4002×H-2002	3XA[1)]	2.4	1.9	79.5	96	98	5.5	5.7	1.8	1.6
H-2002×H-4002	3XB[2)]	2.5	2.1	83.3	100	100	5.1	5.2	2.0	2.1
4M-50×H-2002	3XA	2.5	1.9	76.8	92	82	5.7	5.8	1.6	1.7
4M-59× 〃	3XA	2.2	1.0	47.3	72	30	5.4	5.1	1.4	1.3
4M-60× 〃	3XA	2.0	1.4	71.7	92	82	5.2	5.8	1.4	1.2
M-14×H-4002	3XB	2.5	1.9	73.8	84	88	5.2	5.4	1.7	1.8
M-35×4192	3XB	2.4	2.1	90.7	98	96	5.3	5.6	1.7	1.9
M-41×H-4002	3XB	2.1	1.5	74.0	74	76	4.9	5.3	1.3	1.5

1) Triploids produced from tetraploid mother plants.
2) Triploids produced from diploid mother plants.

hybrids when compared with a diploid strain.

As shown in Figure 2, 100% or 97% triploid seeds were obtained from the crossing between tetraploid male sterile strains and diploid pollinators (B, C and D), whereas a smaller percentage of triploid seeds (34–57%) were produced from diploid male sterile strains (E, F and G). In the progenies of

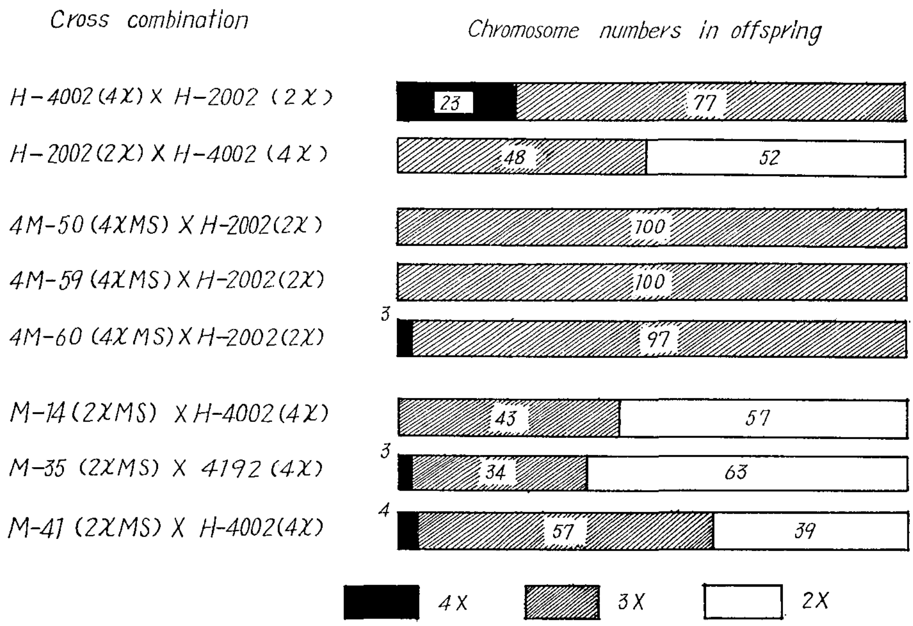

Figure 2. Frequency of diploid, triploid and tetraploid in various populations of triploid hybrids.

the crossing between normal tetraploids and diploids, the frequency of triploids was 77%, while it decreased to 48% in the progeny of the reciprocal direction of the cross, diploids × tetraploids. These results indicate that a higher percentage of triploids produced with tetraploid male sterile plants is due to a higher percentage of male sterile plants in 4M-strains and a faster growth of haploid pollen tubes from diploid pollinators than that of diploid pollens from normal tetraploids which are contained in male sterile tetraploids in 4M-strains. It is also shown that the use of imperfect male sterility in diploids is not effective for the production of a higher proportion of triploids.

C. Agronomic characters of triploid hybrids produced with male sterile parents

The results on the agronomic characters in the trial of 1960 are shown in Table 5 and 6. In these tables, it is noted that the parental tetraploids

TABLE 5. Agronomic characters of various triploid hybrids and their parents in mid growth period in 1960

No.	Strain	Ploidy	Plant height cm	Length of leaf blade cm	Width of leaf blade cm	Number of leaves
1.	Hon-iku-192	2X	51.7 (100)*	25.1 (100)	15.4 (100)	16.7 (100)
2.	M-16	2X	50.1 (97)	25.3 (101)	17.1 (111)	19.3 (116)
3.	H-2002	2X	53.3 (103)	26.9 (107)	17.0 (110)	17.0 (102)
4.	H-4002	4X	44.2 (85)	25.0 (100)	18.3 (119)	18.9 (113)
6.	4M-50	4X	48.1 (93)	24.4 (97)	17.7 (115)	16.5 (99)
7.	H-4002×H-2002	3XA	47.4 (92)	25.0 (100)	16.8 (109)	15.5 (93)
8.	H-2002×H-4002	3XB	52.1 (101)	25.6 (102)	17.0 (111)	16.5 (99)
9.	4M-50×H-2002	3XA	53.0 (102)	25.7 (102)	17.4 (113)	18.2 (109)
10.	4M-59× »	3XA	51.2 (99)	29.7 (107)	17.0 (110)	18.9 (113)
11.	4M-60× »	3XA	53.2 (103)	27.5 (110)	16.5 (107)	17.0 (102)
12.	M-14×H-4002	3XB	51.0 (99)	26.0 (104)	17.2 (112)	17.7 (106)
13.	M-35×4192	3XB	49.2 (95)	26.5 (106)	16.7 (108)	19.4 (116)
14.	M-41×H4002	3XB	57.3 (111)	27.3 (109)	13.6 (89)	16.7 (100)

* Relative number against the standard variety (Hon-iku-192).

decrease in plant height in comparison with the triploid hybrids and diploid strains, while the width of leaf blade increases slightly. The number of plants in each plot was counted before the harvest. The number of plants in a tetraploid strain, H-4002, and a diploid male sterile strain, M-16, decreased significantly from that of the diploid standard variety, Hon-iku-192. Regarding the yield of roots and gross sugar production, the triploid hybrid produced with a tetraploid male sterile parent, 4M-50 × H-4002, showed the best results. The tetraploid, H-4002, and the triploid hybrid, 4M-59 × H-2002, produced a high yield of roots, while H-4002 and the triploids, H-4002 × H-2002 and H-2002 × H-4002, showed a high sugar production. The results of a field test in 1961 are shown in Table 7 anb 8. Tetraploid strains, 4192, H-4002 and 4M-50, showed a decreased plant height from the initiation of the growth. They were also characterized by wide and thick leaves in comparison with diploid strains and triploid hybrids. The number of plants decreased significantly in tetraploid strains, 4192, 4M-50, and triploid hybrids, M-14 × H-4002 and M-41 × H-4002. As to the resistance to the *Cercospora* leaf spot disease, the diploid strain, H-2002, the tetraploid strain, H-4002 and their triploid hybrids, H-4002 × H-2002 and H-2002 × H-4002, showed a relatively high degree of resistance. The yield of roots and sugar content were significantly

TABLE 6. Yield test of various triploid hybrids and their parents in 1960
(Yield per 10 a)

No.	Strain or combination	Ploidy	Number of plants	Total weight kg	Weight of crown and root kg	Root weight I kg	Root weight II[1] kg	Sucrose		Purity (%)	Gross sugar kg
								Percentage	Index[2]		
1.	Hon-iku-192	2X	7650 (100)	3988 (100)	1418 (100)	2571 (100)	2458 (100)	15.68 (100)	23.32	90.98 (100)	348 (100)
2.	M-16	2X	5500 (72)	3568 (89)	1056 (74)	2512 (98)	2444 (99)	15.54 (99)	23.23	91.07 (100)	344 (99)
3.	H-2002	2X	6500 (85)	4659 (117)	1670 (118)	2989 (116)	2861 (116)	16.32 (104)	23.85	91.06 (100)	423 (122)
4.	H-4002	4X	5350 (70)	4498 (113)	1435 (101)	3063 (119)	2992 (122)	16.51 (105)	23.97	91.37 (100)	451 (130)
6.	4M-50	4X	7250 (95)	4008 (100)	1118 (79)	2890 (112)	2846 (116)	15.49 (99)	23.19	90.25 (99)	397 (114)
7.	H-4002×H-2002	3XA	7450 (97)	4300 (108)	1409 (99)	2891 (112)	2824 (115)	17.53 (112)	24.77	92.54 (102)	458 (132)
8.	H-2002×H-4002	3XB	7900 (103)	4608 (116)	1642 (116)	2966 (115)	2871 (117)	16.84 (107)	24.23	92.41 (102)	448 (129)
9.	4M-50×H-2002	3XA	7400 (97)	4888 (123)	1795 (127)	3094 (120)	3018 (123)	16.76 (107)	24.17	91.97 (101)	465 (134)
10.	4M-59× 〃	3XA	7900 (103)	4205 (105)	1146 (81)	3059 (119)	2972 (121)	15.31 (98)	23.04	90.82 (100)	412 (119)
11.	4M-60× 〃	3XA	8500 (111)	4276 (107)	1539 (109)	2737 (106)	2650 (108)	16.00 (102)	23.58	90.64 (100)	385 (111)
12.	M-14×H-4002	3XB	6750 (88)	3472 (87)	1152 (81)	2321 (90)	2270 (92)	14.67 (94)	22.50	89.51 (98)	296 (85)
13.	M-35×4192	3XB	6700 (88)	3685 (92)	1390 (98)	2296 (89)	2210 (90)	15.67 (100)	23.32	91.88 (101)	319 (92)
14.	M-41×H-4002	3XB	7050 (92)	3744 (94)	1379 (97)	2365 (92)	2292 (93)	15.84 (101)	23.46	91.55 (101)	332 (95)
L. S. D. (5%)			974		N. S.		512		0.48		73

1) Root weight excluded small roots having the diameter below 1 cm.
2) Transformed data to angle of arcsin $\sqrt{\%}$

TABLE 7. Agronomic characters of various triploid hybrids and their parents in the mid growth period in 1961

No.	Strain or combination	Ploidy	June 27		July 21			
			Plant height cm	Number of leaves	Plant height cm	Length of leaf blade cm	Length of leaf width cm	Number of leaves
1.	Hon-iku-192	2X	30.0	13.0	53.0	24.2	19.3	19.7
3.	H-2002	2X	29.9	14.0	51.8	23.8	17.6	23.5
4.	H-4002	4X	26.2	11.4	46.3	18.6	16.9	17.7
5.	4192	4X	25.8	10.5	46.3	21.3	21.9	16.0
6.	4M-50	4X	26.0	11.0	48.2	23.1	20.8	16.7
7.	H-4002×H-2002	3XA	30.9	12.6	53.2	27.7	17.7	17.2
8.	H-2002×H-4002	3XB	30.9	13.0	53.8	25.7	19.0	21.0
9.	4M-50×H-2002	3XA	31.0	12.5	54.7	27.0	18.6	19.1
10.	4M-59× 〃	3XA	29.0	12.3	51.0	24.6	18.6	16.9
11.	4M-60× 〃	3XA	29.1	12.8	52.9	24.6	19.0	17.9
12.	M-14×H-4002	3XB	27.4	11.8	47.4	22.2	19.5	17.5
13.	M-35×4192	3XB	28.7	12.9	50.8	24.2	19.2	18.8
14.	M-41×H-4002	3XB	29.3	13.1	51.1	24.8	16.5	18.5
15.	Polyrave	3X	26.9	12.8	47.2	23.8	18.0	19.6
16.	Trirave	3X	27.6	12.5	46.9	22.6	17.5	18.2

TABLE 8. Yield test of various triploid hybrids and their parents in 1961

(Yield yer 10 a)

No.	Strain or combination	Ploidy	Number of plants	Degree of injury by the disease[1)]	Total weight kg	Weight of crown and root kg	Root weight kg	Brix %	Sucrose		Purity %	Gross sugar kg
									Per-centage	Index[2)]		
1.	Hon-iku-192	2X	8100 (100)	3.63 (100)	5765 (100)	1957 (100)	3808 (100)	20.2 (100)	16.52 (100)	23.96	87.3 (100)	550.2 (100)
3.	H-2002	2X	8700 (107)	2.78 (77)	6731 (117)	2253 (115)	4478 (118)	20.0 (99)	16.75 (101)	24.14	89.0 (102)	672.0 (122)
4.	H-4002	4X	6800 (84)	2.35 (65)	4396 (76)	1470 (75)	2926 (77)	19.9 (98)	16.85 (102)	24.23	90.0 (103)	448.2 (81)
5.	4192	4X	6400 (79)	2.95 (81)	5967 (104)	1949 (100)	4018 (106)	18.8 (93)	16.33 (99)	23.83	88.5 (101)	582.9 (106)
6.	4M-50	4X	7260 (90)	3.48 (96)	5342 (93)	1632 (83)	3710 (97)	19.9 (99)	16.51 (100)	23.96	88.7 (102)	543.3 (99)
7.	H-4002×H-2002	3XA	7160 (88)	2.53 (70)	7151 (124)	2321 (119)	4830 (127)	20.4 (101)	17.52 (106)	24.75	92.4 (106)	782.2 (142)
8.	H-2002×H-4002	3XB	8060 (100)	2.65 (73)	6843 (119)	2288 (117)	4556 (120)	20.9 (103)	17.38 (105)	24.64	90.2 (103)	715.2 (130)
9.	4M-50×H-2002	3XA	9460 (117)	3.43 (94)	6769 (117)	2348 (120)	4421 (116)	20.6 (102)	16.57 (100)	24.04	87.6 (100)	641.8 (117)
10.	4M-59× 〃	3XA	8360 (103)	2.98 (82)	5955 (103)	1823 (93)	4132 (109)	19.6 (97)	16.45 (100)	23.92	89.5 (103)	607.7 (110)
11.	4M-60× 〃	3XA	6900 (85)	3.93 (108)	5613 (97)	1609 (82)	4005 (105)	19.8 (98)	16.59 (100)	24.04	88.7 (102)	592.2 (108)
12.	M-14×H-4002	3XB	6800 (84)	3.70 (102)	5431 (94)	1808 (92)	3622 (95)	18.5 (92)	16.08 (97)	23.63	91.2 (105)	529.8 (96)
13.	M-35×4192	3XB	7860 (97)	4.03 (111)	4912 (85)	1743 (89)	3169 (83)	18.3 (91)	15.59 (94)	23.26	89.0 (102)	439.5 (80)
14.	M-41×H-4002	3XB	6500 (80)	3.73 (103)	4871 (85)	1824 (93)	3048 (80)	18.8 (93)	15.94 (96)	23.53	92.2 (106)	446.5 (81)
15.	Polyrave	3X	7900 (98)	5.10 (140)	5485 (95)	1443 (74)	4042 (106)	17.8 (88)	14.90 (90)	22.71	87.4 (100)	525.9 (96)
16.	Trirave	3X	9100 (112)	5.05 (139)	5707 (99)	1508 (77)	4200 (110)	18.5 (92)	15.24 (92)	22.98	88.4 (101)	567.7 (103)
	L. S. D. (5%)		1284	0.63		418	409			0.68		118.3

1) Leaf spot disease by *Cercospora*.

2) Transformed data to angle of arcsin $\sqrt{\%}$.

higher in triploid hybrids, H-4002 × H-2002 and H-2002 × H-4002, than the standard variety, Hon-iku-192. Triploid hybrids, H-4002 × H-2002, H-2002 × H-4002, 4M-50 × H-2002 and 4M-59 × H-2002, and the diploid strain, H-2002 indicated a relatively high gross sugar production.

Throughout the results of two years, the triploid hybrids produced with tetraploid male sterile plants, showed higher yields in both root weight and suger content in comparison with triploid hybrids in similar combinations which were produced with diploid male sterile plants (Figure 3). This seems to

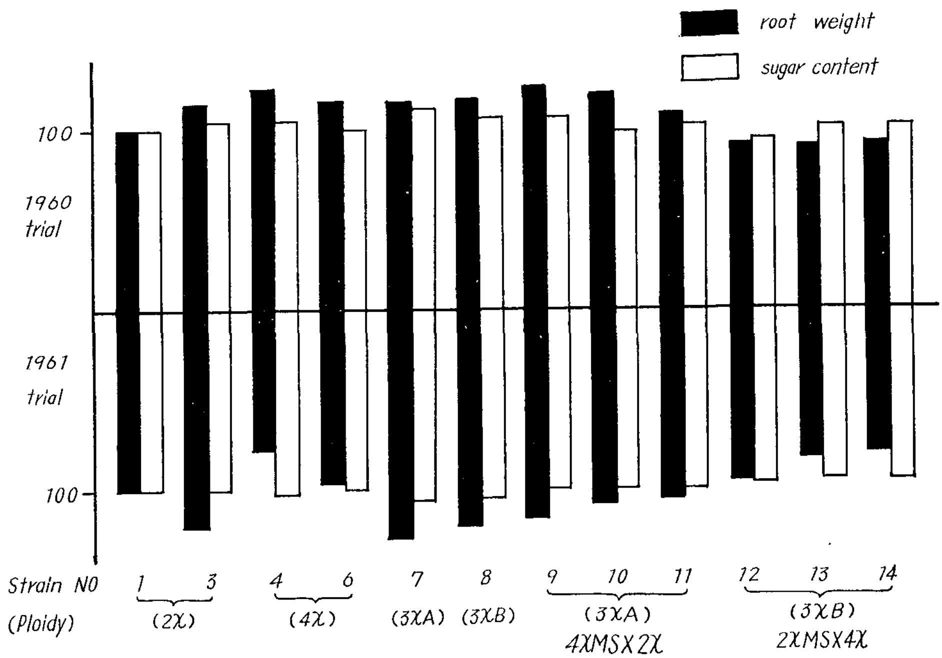

Figure 3. Yield test of various triploid hybrids.

depend on the high proportion of triploids in the triploid hybrids produced with tetraploid male sterile plants. As suggested by BANDLOW (1964), the combining ability of pure triploid hybrids is as important for the yield as the proportion of triploids.

D. Breeding scheme

Triploid hybrids are usually produced by a gingle cross between diploid and tetraploid inbreds or strains. Based on the authors' experimental results, a breeding scheme of triploid hybrids by the use of male sterility both at the diploid and tetraploid levels may be postulated. This is diagramatically shown

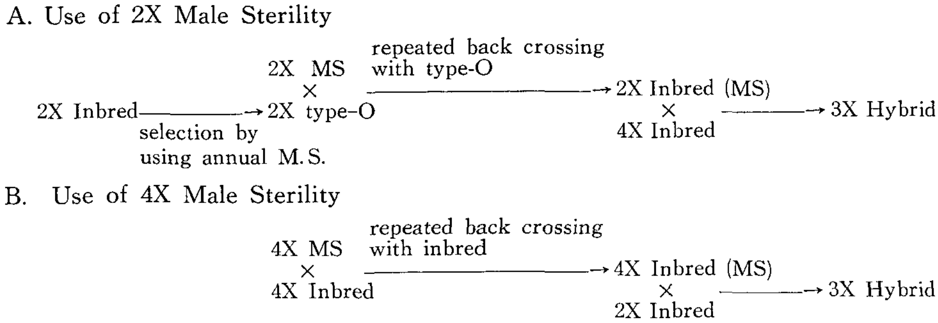

Figure 4. Breeding scheme on triploid hybrids by the use of male sterility.

in Figure 4. In the use of diploid male sterility, type-O plants must be selected from a normal cytoplasm strain by progeny tests of the crosses between an annual complete sterile plant and pollinators from the normal strain. Then diploid male sterile plants are back-crossed with type-O plants as recurrent parents to produce diploid inbreds or strains with male sterility. Pure triploid hybrids are obtained by crossing between a diploid male sterile inbred or strain and a tetraploid inbred or strain. If the seed productivity of a diploid male sterile inbred is insufficient, a single cross hybrid can be replaced for the crossing with a tetraploid inbred or strain.

In the use of trtraploid male sterility, tetraploids of type-O plants are not necessarily provided because of the fact that more than 90% of the plants show practical male sterility in the progeny of the crossing between male sterile plants and pollinators chosen at random from the strain. The male sterile character is introduced to desirable inbreds or strains by successive back crossing. Thus, pure triploid hybrids are produced by crossing between a tetraploid male sterile inbred or strain and a diploid inbred or strain. Finally, combining ability should be testified in triploid hybrids which was produced by the use of diploid or tetraploid male sterility.

Discussion and Conclusion

Owen (1948) proposed a scheme on the utilization of male sterility in breeding of hybrid varieties. He also suggested the use of this character to produce pure triploids. Diploid male sterile strains were used to produce the triploid varieties (Stewart and Gaskill 1952, Ellerton and Hendriksen 1959). In this report, the authors propose the the scheme on the utilization of male sterility both at diploid and tetraploid levels, for triploid seed production. In a trial seed production of triploids, the seed productivity of tetraploid parents was lower than that of diploid parents, while the seed fertility, the germination rate and the average date of germination were not different between the seeds from diploid and tetraploid plants. Almost pure triploids were obtained from the crossing between tetraploid male sterile strains and diploid pollinators, whereas the use of diploid male sterile strains which were produced without the use of type-O plants and contained partially male feritile plants, was not effective for the production of pure triploids. It is considered that the complete male sterile plants which is produced by the use of type-O plants, are indispensable for the triploid seed production between diploid male sterile plants and tetraploid pollinators. The use of tetraploid male sterile plants which was produced without the use of type-O plants and contained the small number of male fertile plants was efficient enough to obtain pure triploids.

Yield tests in gross sugar production covering two years showed that the triploid hybrids from the crosses between tetraploid male sterile strains and diploid pollinators are superior to parental diploid and tetraploid varieties and the triploid hybrids from the reciprocal crosses.

According to the results by BANDLOW (1964), some of the triploid hybrids which were produced with diploid male sterile plants were superior to the diploid control variety. However the combinations with the highest proportion of triploids was not always the best ones but in some cases they were the ones with good combining ability. A preliminary test showed that the yields of almost pure triploid hybrids which were produced with the tetraploid male sterile plants were significantly better than the diploid standard variety. The authors' experimental results indicated that the use of male sterile tetraploids has a potential value for an efficient seed production of triploids.

Summary

1. Tetraploid male sterile strains which were crossed with the pollinators randomly chosen from normal varieties without the use of type-O plants, indicated a large percentage (90–96%) of practical male sterility (C. S. and S. S. b.) and these strains were used for the trial seed production of triploid hybrids. Diploid male sterile strains were also used for the production of triploid hybrids in a similar way.

2. Although the seed productivity of tetraploid parents was lower than that of diploid parents, the seed fertility, the germination rate and the average date of germination were not different between the diploid and tetraploid parents, whether they are male sterile or not.

3. Almost pure triploids were obtained from the crossing between tetraploid

male sterile strains and diploid pollinators whereas the use of diploid male sterile strains which were produced without the use of type-O plants was not effective for the production of pure triploids.

4. Yield tests in gross sugar production covering two years showed that the triploid hybrids with the use of tetraploid male sterile strains are superior to parental diploid and tetraploid varieties, and the triploid hybrids with the use of diploid male sterile strains.

5. The authors proposed the scheme on the utilization of male sterility both at diploid and tetraploid levels. In the use of tetraploid male sterility, the pollinators which were randomly chosen from the normal varieties can be used, instead of the use of type-O plants. Thus, the use of tetraploid male sterile strains has a potential value for an efficient seed production of pure triploids.

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