old budline counterparts did their yield surpass that of old budline trees. Although the oldest nucellar line originated in 1945; juvenility still limited production in early years of the trees (3).

Because fruit quality is not affected and early yields are greater, it seems advantageous to the grower interested in early returns to plant old budline red grapefruit on sour orange rootstock in areas where tristeza is not a problem. The saving in labor and materials resulting from use of smaller trees is also worthy of consideration.

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## Effects of Induced Polyploidy in Cucumbers<sup>1</sup>

O. S. Smith<sup>2</sup> and R. L. Lower<sup>3</sup> North Carolina State University, Raleigh

Abstract. Autotetraploid cucumbers were induced by soaking diploid seed in solutions of colchicine. Triploids were made by reciprocal crosses between diploids and tetraploids. The fruit yield of the polyploids was approx <sup>1</sup>/<sub>2</sub> that of diploids. The fertility of the tetraploid plants as measured by viable seed was approx 1/5 that of diploids. The reduced seed yields of tetraploids appeared to be due to reduced no. of functional male gametes and genic factors. Differences in seed yields were measured among tetraploid lines. Tetraploid by diploid crosses were extremely infertile in either direction.

With the advent of once-over mechanical harvesting of pickling cucumbers, the no. of fruit a plant will set simultaneously has become of primary importance. Previous work with cucurbits has shown that polyploids of this family have reduced seed yields, with triploids having very few if any seeds (1, 3, 4, 5, 6). This study was made to determine if reduced seed yields affected the no. of fruit a plant would set simultaneously and if the polyploids have enough potential fertility for commercial production.

## Materials and Methods

Tetraploid cucumber plants of several cultivars were produced by soaking diploid cucumber seed in 0.5 to 1.0% aqueous colchicine solutions for 6 to 24 hr at 21°C. Plants of the  $CO^5$  generation displaying morphological characteristics similar to a naturally occurring tetraploid, RT<sup>4</sup>, were grown in the greenhouse. Pollen diam was used as the primary screening aid in the selection of tetraploids. Chromosome counts were made on some plants in order to authenticate ploidy level based on pollen sizes. The percentage of pollen stainable with 2% acetocarmine was determined for all lines used in the study. Tetraploids were selfed and crossed, and also crossed reciprocally with diploids. Resulting progenies were grown in the greenhouse and in the field. Completely random designs with 7 plants of each line in the greenhouse and 10 plants of each line in the field were used. The no. of pollinations and the no. of fruit set per plant was recorded for those plants grown in the greenhouse, and the no. of fruit set per plant for plants grown in the field. Data were collected on fruit length to diam

ratio (L/D), skin toughness, and flesh firmness (measured with a USDA fruit Pressure Tester, Magness, .79 cm tip). The skin toughness was computed as the difference between 2 pressure test readings taken on each fruit - 1 on the upper 3rd of the fruit with the skin intact and the other on the lower 3rd of the fruit with the skin removed. The 2nd measurement was used as a measure of flesh firmness. Pressure test and L/D measurements were taken on immature fruit that were approx 4.5 cm in diam. The no. of full-size seed coats per fruit, the no. of full seed per fruit, and the seed size for diploid and polyploid lines of the  $C_{O}$ and  $C_1$  generations were recorded. In this study the no. of full seed per fruit was used as an estimate of female fertility and the total no. of full-size seed coats was used to estimate the no. of functional female gametes produced or potential fertility.

## **Results and Discussion**

Pollen diam was used as the major criterion in preliminary determinations of ploidy level. The pollen from tetraploid plants was similar in size to RT, the naturally occurring polyploid, and significantly larger than pollen from diploid plants (Table 1). Distributions of diploid, triploid, and tetraploid pollen diam were plotted and there was little overlap between pollen size of diploid and tetraploid plants. The triploid pollen diam were quite irregular with the mean square

Table 1. The effect of ploidy level on cucumber pollen size and stainability of 5 cucumber cultivars.

Cultivar	Pollen diam <sup>z,y</sup>		Percent stainabley	
	2 N	4N	2 N	4N
Poinsett	47.1	57.6	99.0	81.0
Tablegreen 65	48.6	65.0	97.0	47.5
N. H. Tiny Dill	45.1	57.4	98.0	85.0
S. C. Gynoecious 3	50.3	63.3	96.0	81.0
MSU 713-5	49.3	60.9	96.0	80.0
RT		59.8		88.0
LSD .01	8.1	73	14	.96

 $^{z}1$  unit = 1.2 $\mu$ .

yValues are means of 10 measurements.

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<sup>&</sup>lt;sup>2</sup>Graduate Assistant, Department of Horticultural Science.

<sup>&</sup>lt;sup>3</sup>Associate Professor, Department of Horticultural Science.

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 $<sup>{}^{5}</sup>C_{O}$  = treated generation;  $C_{1}$  = progeny of treated generation.

being significantly larger than that of the diploids or tetraploids.

In 4 out of 5 cases the percent stainable tetraploid pollen was significantly lower than that of diploid pollen (Table 1). However, stainable pollen ranged from 47.5 to 88% in the tetraploid lines. The percent stainable pollen in triploid plants ranged from 20-45% with an average of 30.1%. The triploid flowers had much less pollen than the tetraploid or diploid flowers. No differences were observed in the amount of pollen produced by diploid and tetraploid flowers.

The effects of increased chromosome sets on morphological characteristics were similar to those discussed by other authors working with polyploid cucumbers (2, 5, 7). These include: increases in the size and amount of serration of the leaves, increases in the size and degree of ruffling of the blossoms, and increases in the size and wt of the seeds. Fruit of the tetraploid plants had significantly lower L/D ratios than diploid fruit of the same cultivar (Table 2). The differences between ploidy levels for skin toughness and flesh firmness were not significant. Fruit of the triploid plants were highly variable, and not enough were available to accurately compare with diploid or tetraploid fruit.

The seed of the tetraploids and triploids were larger than those of diploids (Table 3). Both the length and width of tetraploid seeds were significantly greater than those of diploid seeds. The length and width of triploid seed did not differ significantly from that of the female parent. The tetraploid seed was heaviest, diploid seed next, and triploid seed was the lightest. The embryo and cotyledon of triploid seed was visibly smaller than those found at the other ploidy levels and occupied approx 1/3 of the total vol within the seed coat.

The germination of triploid seed was very low. Only 6.5% of the seed containing embryos from 18  $4N^{\circ}$  x 2nd crosses germinated as compared to 60% germination of the full tetraploid seed and 90% germination of the full diploid seed. However, if fruit of  $4N^{\circ}$  x 2Nd crosses were kept uncut for 2-3 weeks after harvest, a much higher percentage of seeds were found to have sprouted within the fruit. Therefore it appears that many of these triploid seed that would not germinate when planted in the soil did contain viable embryos.

Tetraploid x tetraploid and 4N x  $2N\sigma$  crosses were as successful as  $2N \ge 2N$  crosses on greenhouse grown plants. Diploid plants set an average of 2 fruit per plant when pollinated, whereas tetraploid plants rarely set more than 1 fruit per plant. Triploids used as pollinators would not induce fruit set at any of the 3 ploidy levels. Triploid plants did set fruit when either tetraploids or diploids were used as a pollen source. Several female flowers were pollinated on all of the triploid plants, and rarely did these plants set more than 1 fruit per plant.

The fertility of the  $C_1$  generation as measured by full seed per fruit was compared among ploidy levels grown under comparable conditions (Table 4). In the field the tetraploids produced fewer full seed per fruit than when grown in the greenhouse. No difference was detected for diploid lines.

Table 2. The effect of ploidy level on 3 fruit characters of cucumber.

	Variable <sup>z</sup>			
Line	Ploidy level	Fruit L/D	Skin toughness	Flesh firmness
MSU 713-5 x Poinsett	2 N	3.48	5.6	11.0
	4N	2.52	6.6	11.4
MSU 713-5 x Tablegreen 65	2 N	3.11	6.4	11.0
	4N	2.38	6.6	10.4
S. C. Gynoecious 3	2N	2.50	10.6	11.6
	4N	1.73	7.8	9.0
LSD .01		.70	4.45	3.83

<sup>z</sup>Values are means of 10 measurements.

Table 3. The effects of ploidy level on seed size and wt of cucumber.

		Seed size <sup>z</sup>	ize <sup>z</sup>	
Ploidy level <sup>y</sup>	Length (mm)	Width (mm)	Wt <sup>x</sup>	
2N 🛞	13.8	8.8	2.5332	
2N x 4N	13.1	8.8		
4N 🛞	15.3	9.6	3.7812	
4N x 2N	16.2	9.8	-	
3N x 2N	15.1	9.4	2.0846	
LSD .01	1.0	0.8	0.5148	

<sup>z</sup>Values are means of 10 measurements.

<sup>y</sup>Female parent always designated 1st.

 $x_{Expressed}$  as g/100 seed.

Triploid lines rarely set seed.

The fertility of the tetraploid lines ranged from almost complete sterility to about 1/5 that of diploid lines. Significant differences were found between the viable seed yields of several of the tetraploid lines in the C<sub>O</sub> generation. Significant differences between crosses of the low yielding tetraploid lines

Table 4. The no. of full seeds per cucumber fruit produced under field and greenhouse conditions.

	No.	Full seeds/fruit		
Type of pollination	of fruit	Range	Mean <sup>z</sup>	
4N (O.P. <sup>y</sup> field)	47		7.1 <sup>b</sup>	
4N x 4N (field)	30	2-75	16.1 <sup>at</sup>	
4N x 4N (greenhouse)	93	7-134	48.3 <sup>a</sup>	
3N x 2N (greenhouse)	14	0-9	3.3b	
3N x 3N (greenhouse)	0	-	-	
2N x 2N (O.P. field)	36	_	274.6 <sup>c</sup>	
2N x 2N (greenhouse)	10	92-340	206.1 <sup>c</sup>	

<sup>z</sup>Mean separation by Duncan's multiple range test, 5% level.

YO.P. = open-pollinated in field of diploid and tetraploid plants.

were also observed in the  $C_1$  generation. These differences indicate that genetic factors probably account for part of the variation in the fertility levels observed. Additional evidence that these differences were partly due to genetic causes is indicated by the following observations. Some tetraploid lines

Table 5. Total no. of full-size seed coats per cucumber fruit produced in diploid-tetraploid crosses.

	Cross <sup>z</sup>			
Generation	2N x 2N control	2N x 4N	4N x 2N	4N x 4N
$\overline{C_1}$ (field)	217.6 <sup>a</sup>	53.5 <sup>b</sup>	165.4 <sup>a</sup>	59.3 <sup>b</sup>

<sup>Z</sup>Mean separation by Duncan's multiple range test for comparisons made within generations, 1% level.

that were nearly self-sterile would produce viable seed when outcrossed to other tetraploid lines. When the average no. of full seed produced by the  $C_1$  generation was regressed on the average no. of full seed produced by corresponding lines in the

Table 6. No. of full seed produced in diploid-tetraploid cucumber crosses.

Cross	No. of pollinations	No. of fruit set	Avg no. of full seed/fruit
4N x 2N <sup>Z</sup>	65	44	0.98
2N x 4N <sup>z</sup>	46	25	0.08
4N x 2N <sup>y</sup>	55	41	0.21

<sup>z</sup>In these crosses RT was used as the 4N parent.

<sup>y</sup>Crosses used the induced tetraploids as either the female or male parent.

 $C_O$  generation, the correlation coefficient was 0.65; significant at the 1% level.

The total no. of full-size seed coats was determined at diploid and tetraploid levels and used as an estimate of functional female gametes (Table 5). The seed coats produced in  $4N \ge 2N$ crosses indicated that the tetraploid lines produced female gametes in no. (165.4) comparable to those of diploids (217.6). When the tetraploid was used as the female parent the no. of seed coats was not significantly different from that of the diploid, but the difference between the diploid and tetraploid seed yields was significant when the tetraploid was used as the male parent. Evidence from pollen stainability counts and the reduction in full-size seed coats when tetraploids were used as pollen parents indicated that tetraploids produced significantly fewer functional male gametes than diploids, however, this should not be a limiting factor.

The extremely low no. of full triploid seed produced in diploid-tetraploid crosses may be attributable to physiological imbalances caused by the triploid chromosome no. (Table 6). Fruit set was induced when the tetraploid was either the male or female parent, but the seed yield was extremely low when the tetraploid was the male parent. This may in part be due to the reduced pollen counts of the tetraploid blossoms. However, the highest average seed yield was only about 1 seed per fruit.

Selection would probably be effective in increasing the seed

yields of the tetraploids. The low seed yields of the tetraploid by diploid crosses suggest that it would not be commercially feasible to produce large quantities of usable triploid seed. This and the lack of evidence that the polyploids are capable of increased fruit set indicates that polyploids will not be used to increase the yields of cucumber crops.

Although fruit yield of polyploids was less than that of diploids, the increase in chromosome no. did not influence the taste or quality of the fruit to an observable extent.

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# Manganese Enrichment of Tomato and Onion Seed<sup>1</sup>

## Ross J. Traverse<sup>2</sup> and Jerald W. Riekels<sup>3</sup> University of Guelph, Guelph, Ontario, Canada

Abstract. Soaking tomato seeds in MnSO4 solutions of concentrations greater than 0.5 and 1 M MnSO4 inhibited germination during treatment without affecting the viability of the seeds. The emergence and early growth of tomato seedlings and the emergence of onion seedlings in soil was greater using seeds previously treated with 1 M MnSO4 than with untreated seeds or with seeds treated with 2 and 2.5 M MnSO4. These treatments had no effect on onion seedling growth. Soaking seeds in 1 M MnSO4 was effective in supplying the Mn requirements of tomato plants grown in Mn deficient solutions for Approx 40 days. Shorter periods of normal growth were obtained by treating the seeds with less than 1 M concn of MnSO4.

The amount of Mn retained after desorption and washing was greater with each increase in the soaking temp (0, 10, 20, and  $30^{\circ}$ C). A substantial amount of the Mn retained by the tomato and onion seeds after soaking appeared to be located on the seed coat or in the "outer space" of the tissue. With onion seeds, an additional portion of the Mn retained after soaking was located on the exchange sites of the seeds.

The aims of research on seed soaking either with water or salt solutions, have been to stimulate growth and occasionally to supply a nutrient to the plant. Increases in yield have been reported for various crops using seeds treated with salts of P (15), Mn (5, 15), and Mo (4, 18), but no research has been conducted to determine the precise nutritional benefit to be derived from soaking seeds in various salt solutions. Since smaller quantities of the nutrient-element salts could be applied to the seeds in comparison to soil applications, toxic nutrient imbalances and fertilizer pollution could be minimized. Also in soils with a low amount of a particular nutrient, the developing

seeds and seedlings would receive a supply of the deficient element.

The emergence of tomato and pepper seedlings was stimulated by treating the seeds with 2% solutions of K<sub>3</sub>PO<sub>4</sub> and KNO<sub>3</sub>, and to a lesser extent, with distilled water and 2% NaCl (6). Delays in germination have occurred, however, with high concn of salt in the treatment solutions (15, 17). A stimulation of emergence and early seedling growth is desirable on organic soils in Canada and northern United States where emergence is normally slow because of cold soil temp early in the spring. Since crops grown on organic soils frequently develop Mn deficiency, Mn was selected for study. We consider nutritional enrichment and growth stimulation simultaneously by determining the nutritional value of soaking seeds in Mn solutions that enhance seedling emergence without initiating germination during treatment or affecting seed viability.

The salts in treated seeds have been reported to be held superficially since heavy losses occurred when treated seeds were washed (11) and cotton seed analysis indicated that N, P, and K did not penetrate the embryo (3). However, Mn, Cu (3),

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<sup>&</sup>lt;sup>2</sup>Present address: Department of Mines, Agriculture and Resources, Government of Newfoundland and Labrador, St. John's, Newfoundland. <sup>3</sup>Department of Horticultural Science.