C_0 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 x 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[1]

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Abstract. Soaking tomato seeds in $MnSO_4$ solutions of concentrations greater than 0.5 and 1 M $MnSO_4$ 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 $MnSO_4$ than with untreated seeds or with seeds treated with 2 and 2.5 M $MnSO_4$. These treatments had no effect on onion seedling growth. Soaking seeds in 1 M $MnSO_4$ 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 $MnSO_4$.

The amount of Mn retained after desorption and washing was greater with each increase in the soaking temp (0, 10, 20, and 30°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_3PO_4 and KNO_3, 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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and Zn (14) have been found in the embryo of seeds after seed treatment. For max nutritional benefit, the treated seeds should retain all of the nutrient supplied during the soaking period. Washing and drying the seeds after treatment was studied to measure losses in seed viability and Mn losses from the seeds. This information would indicate whether hydro seeding or dry seeding should be practiced to obtain max stimulation of growth and nutritional benefit.

Materials and Methods

We used tomato, *Lycopersicon esculentum* Mill. cv. Rutgers, and onion, *Allium cepa* L. cv. Autumn Spice, to study the effect of $MnSO_4$ concn on germination and emergence, on plant growth in Mn deficient solutions, and on Mn retention by seeds. The design for each experiment was a randomized complete block with 3 replications. The results were analyzed by standard analysis of variance with differences between means tested by Duncan's multiple range test.

Germination during seed treatment. To determine a concn of $MnSO_4$ which would inhibit germination during the treatment period without affecting seed viability, seeds were soaked in solutions of $MnSO_4$ at concn of 10^{-6}, 10^{-5}, 10^{-4}, 10^{-3}, 10^{-2}, 10^{-1}, and 1 M (Experiment 1) and 10^{-1}, 2.5 x 10^{-1}, and 5 x 10^{-1} M (Experiment 2). Three ml of each solution and a control using distilled water were added to 50 tomato seeds on 2 circles of filter paper in a 9 cm Petri dish (6). Germination was evaluated at 25°C daily for 14 days. Following this period, the seeds treated with concn of $MnSO_4$ that completely inhibited germination, were seeded in flats of sterilized soil (1/3 sand, 1/3 loam, and 1/3 vermiculite). Seedling emergence from treated seeds was compared with emergence from untreated seeds to indicate the effect of $MnSO_4$ solutions on seed viability.

Emergence from treated seeds. The influence of $MnSO_4$ seed treatments on seedling emergence in flats of sterilized soil was determined using tomato and onion seeds soaked in 1, 2, or 2.5 M $MnSO_4$ in Petri dishes as previously described. The dishes were covered with a polyethylene bag, and stored for 7 days at 25°C. After the soaking period, lots of 25 seeds were treated 4 ways: washed with 50 ml of distilled water and seeded wet, washed then dried on filter paper at 25°C for 18 hr and seeded dry, not washed and seeded wet, and not washed and seeded dry. Washed and unwashed untreated seeds were used as controls. After seeding, the flats were placed in a ventilated greenhouse with 25°C day and 20°C night temp. Emergence was recorded each day for 13 days after seeding. Fresh wt of the seedlings was determined 21 days after planting.

Growth in Mn deficient solutions. Tomato plant growth using treated and untreated seeds was evaluated in complete and Mn-deficient nutrient solutions to determine the length of time a plant could be grown from the Mn added to the seed. The plants were grown in a ventilated greenhouse in 1 liter flasks in continuously aerated nutrient solutions. In each flask, a small rectangle of stainless steel mesh was placed in a bored rubber stopper. A single seed was placed on the mesh and covered with a strip of filter paper, so that the ends of the filter paper were in the nutrient solution to act as a wick and moisten the seed. When the seedlings reached the first true leaf stage, they were removed from the mesh and supported with glass wool.

The composition of the complete nutrient solution was: 6.35 mM $Ca(NO)_3 \cdot 4H_2O$; 1.35 mM $Mg(NO_3)_2 \cdot 6H_2O$; 1.35 mM NH_4NO_3; 2.4 mM K_2HPO_4; 0.65 mM$(NH_4)_2HPO_4$; 0.05 mM KCl; 0.25 mM H_3BO_3; 0.005 mM $MnSO_4 \cdot H_2O$; 0.002 mM $ZnSO_4 \cdot 7H_2O$; 0.0005 mM $CuSO_4 \cdot 5H_2O$; 0.0001 mM H_2MoO_4 (85% MoO_3); and 0.005 mM FeEDDHA. Macronutrients were prepared and purified as described by Steinberg (16); micronutrients were prepared according to Johnson et al. (10). To prevent the addition of Mn to the solutions from sources other than the seed, the solutions were not changed during the experimental period. Demineralized water was added as required.

Plants were grown from treated seeds in Mn-deficient solutions and from untreated seeds (control) in complete and Mn-deficient solutions. Increasing amounts of Mn in the seeds were obtained by soaking the seeds in solutions with various combinations of $MnSO_4$ and $MgSO_4$ to provide a total 1 M salt concn as follows: 0.25 M $MnSO_4$ + 0.75 M $MgSO_4$, 0.50 M $MnSO_4$ + 0.50 M $MgSO_4$, 0.75 M $MnSO_4$ + 0.25 M $MgSO_4$, and 1 M $MnSO_4$ (Table 3, Experiment 1). Due to an error in experiment 1 with the 1 M $MnSO_4$, this treatment along with the 0.75 M $MnSO_4$ treatment was repeated (Table 3, Experiment 2).

Plants were observed daily to determine the first occurrence of Mn deficiency symptoms. The date of any change of color (chlorosis) compared to the control plants was recorded. If chlorosis progressed to typical Mn deficiency symptoms, then the date used to calculate the days of normal growth was that of the first change of color. The experiments were terminated when plants in the complete nutrient solutions began showing macronutrient deficiency symptoms. The fresh wt of each plant was determined at the end of the experiment.

Mn retention by treated seed. A solution of 1 M $MnSO_4$ labelled with ^{54}Mn (initial activity approx 4000 counts/μmole), was used to determine the effect of soaking temp on amount of Mn retained by tomato and onion seeds. Test tubes (50 x 300 mm), each containing 100 ml of the labelled solution, were placed in water baths at 0, 10, 20, and 30°C. Samples of approx 1800 seeds of tomato (4.5 g) and 1800 seeds of onion (7.7 g) were soaked in the aerated solution for 4 days. There were 3 replications at each temp with each crop. After soaking, the seeds were removed from the labelled solution by filtering through cheesecloth and were allowed to dry at room temp. Lots of 400 seeds, taken from each sample after soaking and drying, were weighed and assayed for ^{54}Mn in a NaI crystal scintillation counter.

Desorption was conducted by washing 400 seeds of the samples, previously treated with ^{54}Mn, in 100 ml of non-radioactive 1 M $MnSO_4$. Another 400 seeds from each sample were washed in 100 ml of distilled water. Desorption and washing by the "tea bag" technique (12) were carried out in 50 x 300 mm test tubes in a 5°C water bath for ½ hr. The seeds were dried at 25°C for 2 days in the "tea bag" and then removed and assayed for ^{54}Mn. Results are expressed as total amount (μmoles) of Mn retained by the seeds.

Results and Discussion

Germination during seed treatment. The patterns of tomato seed germination (Table 1, Experiment 1) for concn of $MnSO_4$

Table 1. Effect of concn of $MnSO_4$ on rate of tomato seed germination.

Concn of $MnSO_4$	Days from beginning of germination					
	1	2	3	4	5	14
	No. germinated[z] Experiment 1					
H_2O	12 a[y]	26 b	32 a	36 a	37 a	39 a
10^{-2} M	15 a	32 a	36 a	38 a	38 a	39 a
10^{-1} M	0 b	7 c	23 b	30 b	35 a	40 a
1 M	0 b	0 d	0 c	0 c	0 b	0 b
	Experiment 2					
H_2O	10 a	30 a	36 a	37 a	39 a	41 a
10^{-1} M	0 b	12 b	25 b	32 b	36 a	39 a
2.5 x 10^{-1} M	0 b	0 c	0 c	0 c	3 b	7 b
5 x 10^{-1} M	0 b	0 c	0 c	0 c	0 c	0 c

[z]Cumulative germination, mean of 3 replications of 50 seeds each.
[y]Mean separation by Duncan's multiple range test, within columns in each experiment, 5% level.

up to and including 10^{-2} M and for the water control were similar, hence data for concn of $MnSO_4$ less than 10^{-2} M are not presented. Increasing the concn of $MnSO_4$ to 10^{-1} and 2.5 x 10^{-1} M progressively delayed germination with no germination occurring during the 14 day experimental period for 5 x 10^{-1} M and 1 M $MnSO_4$ (Table 1, Experiment 2). Following the 14 day soaking period, the ungerminated seeds were seeded in flats of sterilized soil. Seedling emergence was 84% from 1 M $MnSO_4$-treated seeds and 80% from untreated seeds which indicated that viability of tomato seeds was not affected by treatment with $MnSO_4$ solutions.

Similar results were reported by Ells (6) using 2% solutions of K_3PO_4, KNO_3, or NaCl and by Hood (9) using 2 M $MgCl_2$. Since different salts produce the same effect, germination apparently is inhibited by the high osmotic pressure of the solution. Uhvits (17) has shown that hydration of seeds decreases as the osmotic pressure of the soaking solution increases. This indicates that tomato seed germination was inhibited with 1 M $MnSO_4$ because the seeds could not take up sufficient water. The delay in germination with the 10^{-1} M $MnSO_4$ solutions may have been due to water uptake slower than that at lower concn.

Loo and Tang (13) reported an acceleration in rate of germination of cabbage, mung beans, and maize with 10^{-3} M $MnSO_4$. We observed no enhancement of germination of tomato seeds at this concn.

Emergence from treated seeds. Tomato seedling emergence was enhanced and growth was greater with the 1 M $MnSO_4$ seed treatment than with the control and the 2 and 2.5 M $MnSO_4$ treatments (Table 2). Total emergence was not reduced by the 2 and 2.5 M $MnSO_4$ except when the seeds were treated with 2.5 M $MnSO_4$ and seeded wet. A few differences were observed between washed and unwashed seeds, but in general the data indicated the seeds need not be washed to remove excess salt before seeding. The data for seeding with wet and dry seeds were not compared statistically, but the fresh wt figures indicate that growth was enhanced by drying the tomato seeds after treatment. However, with 1 M $MnSO_4$, washing or drying the seeds had no effect on emergence 7 days after seeding or on seedling growth (Table 2). These results indicate that many different seeding methods (wet or dry) could be used with Mn enriched seeds to obtain early seedling emergence and greater seedling growth.

Increases in the fresh wt of tomato seedlings with the 1 M $MnSO_4$ seed treatment are attributed to earlier emergence. The increase was not due to the nutritional value of $MnSO_4$ since plant growth with untreated tomato seeds was equivalent to that obtained using seeds treated with 2 and 2.5 M $MnSO_4$.

The pattern of onion seedling emergence was essentially the same as that obtained for tomatoes, and the treatments had no effect on onion seedling growth; consequently the data are not presented.

Growth in deficient solutions. Plants grown from seeds treated with $MnSO_4$ had a longer period of normal growth and greater fresh wt than the plants grown from untreated seeds in Mn deficient solutions (Table 3). Plants grown from seeds treated with 1 M or 0.75 M $MnSO_4$ developed Mn deficiency symptoms sooner and weighed less than the plants grown with 5 μM Mn in solution. Soaking seeds in 0.75 M or 1 M $MnSO_4$ was effective in supplying the Mn requirements of tomato plants for

Table 2. Tomato seedling emergence and growth as influenced by washing and drying the seed after treatment with various concn of $MnSO_4$.

Concn of $MnSO_4$	Washing	Seeded wet: Days after seeding 7	Seeded wet: Days after seeding 13	Seeded wet: Fresh wt mg/plant	Seeded dry: Days after seeding 7	Seeded dry: Days after seeding 13	Seeded dry: Fresh wt mg/plant
		Numbers emerged			Numbers emerged		
1M	washed	14[z]a[y]	18 a	169 a	14 a	19 ab	222 a
1M	none	10 a	17 a	127 b	13 a	21 a	199 ab
2M	washed	1 b	14 bc	116 b	1 b	16 b	161 bc
2M	none	1 b	13 cd	119 b	0 b	20 ab	131 c
2.5 M	washed	2 b	12 cde	113 b	0 b	18 ab	142 c
2.5 M	none	1 b	8 e	73 c	0 b	19 ab	132 c
Untreated	washed	1 b	14 bc	79 c	0 b	17 ab	144 c
Untreated	none[x]	1 b	8 e	97 bc	0 b	19 ab	136 c

[z]Cumulative emergence, mean of 3 replications of 25 seeds each.

[y]Mean separation by Duncan's multiple range test, within columns, 5% level.

[x]Seeds of this treatment were not exposed to water before seeding.

Table 3. Influence of seed treatment with $MnSO_4$ on growth of tomato plants in Mn deficient solutions.

Seed soaking	Mn in solution	G/plant fresh wt	Days to Mn deficiency
		Experiment 1[z]	
none	5 μM	148.7 A[y]	69 A
none	none	0.2 D	25 D
0.75 M $MnSO_4$	none	60.0 B	48 B
0.5 M $MnSO_4$	none	10.5 CD	34 C
0.25 M $MnSO_4$	none	24.0 C	31 CD
		Experiment 2[z]	
none	5 μM	163.7 A	68 A
none	none	0.6 C	18 C
1 M $MnSO_4$	none	84.7 B	41 B
0.75 M $MnSO_4$	none	93.6 B	38 B

[z]Age of plants at harvest: Experiment 1, 69 days; Experiment 2, 69 days.

[y]Mean separation by Duncan's multiple range test, within columns, for each experiment, 1% level.

Table 4. Effect of desorption and washing on removal of Mn from tomato and onion seeds soaked in 1 M $MnSO_4$.

Solution temp °C	Mn in seed after: Initial treatment μmoles/g	Desorption μmoles/g	Desorption %	Washing μmoles/g	Washing %
		Tomato			
0	248 b[z]	30 c	12 b	27 b	12 b
10	238 b	39 bc	16 ab	38 b	16 ab
20	249 b	45 b	20 a	49 a	24 a
30	348 a	60 a	19 ab	58 a	19 ab
		Onion			
0	160 b	57 c	35 a	109 b	68 a
10	213 b	75 bc	35 a	117 b	55 a
20	222 b	82 b	37 a	132 b	62 a
30	304 a	116 a	38 a	172 a	68 a

[z]Mean separation by Duncan's multiple range test, within columns for each species, 5% level.

approx 40 days during the early stages of growth in Mn deficient media. Less growth was obtained by soaking the seeds in concn less than 0.75 M $MnSO_4$, but the plants grown with 0.25 M and 0.50 M $MnSO_4$ did have sufficient size to make foliar sprays of Mn feasible. However, for growth in Mn deficient soil, soaking tomato seeds with 1 M $MnSO_4$ would supply Mn for a longer period of time and fewer foliar applications of Mn would be required for subsequent growth.

Plant growth with treatments of 0.75 M $MnSO_4$, no seed soaking, and no Mn was greater in experiment 2 than in experiment 1 (Table 3). Experiment 1 was conducted during winter months when light intensity and duration were less favorable for rapid growth than for experiment 2 which was conducted in early spring. With faster growth, the Mn supplied by the seed would be depleted sooner in experiment 2 than in experiment 1. This would account for the shorter period of time with no Mn deficiency symptoms for the 0.75 M $MnSO_4$ treatment in experiment 2.

Mn retention by treated seeds. With a soaking temp of 30°C, more Mn was retained by both tomato and onion seeds following the initial treatment and after desorption or washing than was retained by seeds soaked at the lower temp (Table 4). Thus seeds soaked in $MnSO_4$ at high temp would have more Mn available for plant growth than seeds soaked at lower temp.

Washing, to remove diffusable Mn (7), and desorption, to remove exchangeable Mn (8), removed similar amounts of Mn from tomato seeds. This indicates that most of the Mn in tomato seeds is readily diffusable and easily leached from seeds. With onion seeds, Mn was present in both exchangeable and diffusable forms. This indicates that Mn accumulation by seeds is predominantly passive (7), a finding similar to that of other workers (2, 4, 17) in which salt accumulation increased as the concn of the salt in the treating solution increased.

However, since a portion of the Mn was not removed by desorption and the amount of Mn remaining in the tomato and onion seeds after desorption increased with increasing temp, some active absorption (8) of Mn by the seeds may have occurred. Seeds have a semipermeable membrane around the embryo (1, 2) and ions will enter the embryo (3, 14). This indicates that some ions could enter the seeds by active processes.

Many different seed handling methods could be used with Mn enriched seeds which are treated primarily to obtain earlier seedling emergence. Seeds treated with Mn for nutritional purposes, however, should not be washed or seeded by hydroseeding methods, since our results indicated that substantial amounts of Mn can be leached from tomato and onion seeds.

Our study indicated that soaking tomato seeds in 1 M $MnSO_4$ will enrich the seeds with sufficient Mn to sustain normal growth for approx 40 days before additional Mn is required from foliar or soil applications. Thus, adding Mn through combinations of seed treatments and foliar applications would maintain normal growth without excessive accumulation of this element in the soil which might occur with repeated soil applications over a long period of time. Enriching seeds with Mn also would insure that some Mn is available for seedling development in Mn deficient soils.

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