



Figure 4. Total percentages of iron extracted by HEEDTA and EDDHA from chlorotic and nonchlorotic tomato leaf tissues harvested at three successive times after planting

not be ignored, since much of the iron is relatively resistant to removal and may reside in the wall of the cell.

Similar fractionations of iron in stem and root tissues provided little clue as to the forms of iron in chlorotic as compared with nonchlorotic tissue because of inconsistent data.

#### Literature Cited

- (1) Bennett, J. P., *Soil Sci.* **60**, 91 (1945).
- (2) Bowen, H. J. M., Cawse, P. A., Thick, J., *J. Exptl. Botany* **13**, 257 (1962).
- (3) Greene, R. E., Bullock, J. S., Maier, R. H., *Agron. J.* **54**, 363 (1962).
- (4) Hoagland, D. R., Arnon, D. I.,

- "Water-Culture Method for Growing Plants without Soil," Calif. Agr. Expt. Sta. Circ. 347 (revised) (1951).
- (5) Jackson, M. L., "Soil Chemical Analysis," Prentice-Hall, Englewood Cliffs, N. J., 1960.
  - (6) Jacobson, L., Oertli, J. J., *Plant Physiol.* **31**, 199 (1956).
  - (7) Maier, R. H., Cattani, R. A., *Life Sciences* **4**, 391 (1965).
  - (8) Mills, C. F., *Biochem. J.* **57**, 603 (1954).
  - (9) Oserkowsky, J., *Plant Physiol.* **8**, 449 (1933).
  - (10) Schmid, W. E., Gerloff, G. C., *Ibid.*, **36**, 226 (1961).
  - (11) Tiffin, L. O., Brown, J. C., *Science* **135**, 311 (1962).
  - (12) Wallihan, E. F., *Am. J. Botany* **42**, 101 (1955).

Received for review January 29, 1965. Accepted February 9, 1966. Approved for publication as Technical Paper No. 930 by the Director of the Arizona Agricultural Experiment Station, Tucson, Ariz. Investigation partly financed by grants from the National Science Foundation (Grant GB-96) and Geigy Chemical Corp., Ardsley, N. Y.

## MINERAL NUTRITION IN PLANTS

### Chemical Fractions of Plant Potassium, Calcium, and Magnesium as Influenced by Soil Treatment

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Total, water- and acid-soluble potassium, calcium, and magnesium have been studied in tomato stem and leaf tissue as a function of soil applications of potassium and magnesium. Total leaf potassium was highest as a result of potassium and magnesium applications. Highest amounts of water-soluble plant magnesium resulted from the high rates of potassium and magnesium applications. Potassium applications did not inhibit plant uptake of magnesium. Approximately 95% of both stem and leaf potassium was water and acid soluble. More stem and leaf calcium was acid soluble than was water soluble. However, more than half of the stem and leaf calcium was not soluble in water or acid. Proportionately more plant magnesium was extracted with acid than with water.

IN AN EFFORT to understand better mineral nutrition of plants with regard to potassium, calcium, and magnesium, it would be helpful to obtain further information on the forms of these elements in certain plant parts. This approach is not only essential for the plant scientist, but information is now available indicating that plant forms of elements strongly influence animal utilization of the element ingested (5, 17).

The total amount of an element in a tissue is not a reliable criterion to evaluate its relation to growth and function. Elucidation of the active form (4) of the nutrient should be a continual goal. A prerequisite to research on the available or active forms of potassium, calcium, and

magnesium in plants is predicated on a knowledge of the forms of occurrence of the element within selected tissues, and how that form changes as a function of environmental components.

The purpose of this investigation was to contribute information on the total, water- and acid-soluble amounts of potassium, calcium, and magnesium in tomato stem and leaf tissue as a function of soil applications of potassium and magnesium. Previous data on total amounts of potassium, calcium, and magnesium in tomato plants have been compiled (7, 9).

#### Materials and Methods

**Materials.** Laveen loam, 0 to 12

inches, was obtained from the Mesa Experimental Farm, Mesa, Ariz. After collection, the soil was air-dried, crushed with a wooden rolling pin, and sieved through a plastic screen with 10-mm. openings.

The inside surfaces of No. 10 tin cans were coated with asphalt emulsion, and each treatment described below was replicated three times. The necessary amount of soil for each can was placed in a twin-shell plastic blender, and the appropriate nutrients were added. The mixture was blended for 20 minutes and then was transferred to the respective container.

**Base Nutrient Applications.** The base nutrient application for all treat-

**Table I. Soil Treatments**

Treatment Designation	Description
A	24 lbs. K/A, <sup>a</sup> no magnesium added
B	24 lbs. K/A and 15 lbs. Mg/A from Sul-Po-Mag <sup>b</sup>
C	80 lbs. K/A and 50 lbs. Mg/A from Sul-Po-Mag <sup>b</sup>
D	24 lbs. K/A, <sup>a</sup> 15 lbs. Mg/A from Granules <sup>c</sup>
E	80 lbs. K/A, <sup>a</sup> 50 lbs. Mg/A from Granules
F	80 lbs. K/A, <sup>a</sup> no magnesium added

<sup>a</sup> From K<sub>2</sub>SO<sub>4</sub>.

<sup>b</sup> A trade name for the double sulfate of potash-magnesia; contains about 40% K<sub>2</sub>SO<sub>4</sub> and 55% MgSO<sub>4</sub>. Furnished by the International Minerals and Chemical Corp., Skokie, Ill.

<sup>c</sup> An inert carrier sprayed with magnesium sulfate and binding agent. Furnished by Minnesota Mining and Manufacturing Co., St. Paul, Minn.

**Table II. Dry Weights of Tomato Plant Tissue as Influenced by Treatments**

Treatment	Means-Dry Wt., Grams <sup>a</sup>		
	Stem	Leaf	Total
A	5.37a	7.41a	12.78a
B	5.73a	7.90ab	13.63ab
C	5.94a	8.86bc	14.80bc
D	5.58a	7.48a	13.06a
E	5.79a	7.80a	13.59ab
F	6.40a	9.16c	15.56c

<sup>a</sup> In comparison between weight of plant parts and total weight *vs.* treatments, means with like symbols do not differ significantly at the 5% level of probability.

ments on a per acre basis consisted of: 350 pounds of nitrogen as ammonium nitrate; 75 pounds of phosphorus as monocalcium phosphate; 20 pounds of iron as the ferric chelate of ethylenediamine di(*o*-hydroxyphenylacetic acid) (FeEDDHA); 12 pounds of manganese as the manganese chelate of ethylenediaminetetraacetic acid (MnEDTA); 9 pounds of zinc as ZnEDTA; and 6 pounds of copper as CuEDTA. The magnesium and potassium treatments are shown in Table I. All soils were held at approximately 90% of the moisture equivalent (10).

Eight seeds of tomato, *Lycopersicon esculentum*, variety Early Pak were planted in each can in a glass greenhouse and, after emergence, were thinned to three plants per can. The plants were harvested after 8 weeks of growth. At harvest, the plants were cut at the soil surface, and the aerial portion was divided into leaflets (referred to as leaves), and stems and petioles (referred to as stems). The harvested plant material was frozen immediately with dry ice, lyophilized, and ground to pass a 60-mesh screen, and stored at -18° C., for subsequent chemical analysis.

Tissue was analyzed for total, water-soluble and acid-soluble potassium, cal-

**Table III. Concentration and Total Amount of Element Fractions in the Stems and Leaves of Tomato Plants**

Treatment	Total		H <sub>2</sub> O Soluble		Acid Soluble		% Total Extracted with	
	Mg./gram	Total mg.	Mg./gram	Total mg.	Mg./gram	Total mg.	H <sub>2</sub> O	Acid
POTASSIUM Stems								
A	10.0	53.2 <sup>a</sup>	9.2	48.7 <sup>a</sup>	9.3	49.6 <sup>a</sup>	91.7	93.4
B	9.3	53.2 <sup>a</sup>	8.7	49.4 <sup>a</sup>	9.3	53.3 <sup>a</sup>	92.9	100.0
C	8.7	51.5 <sup>a</sup>	7.8	46.5 <sup>a</sup>	8.2	48.6 <sup>a</sup>	90.3	94.4
D	10.0	55.6 <sup>a</sup>	9.2	51.1 <sup>a</sup>	9.5	52.9 <sup>a</sup>	91.9	95.1
E	8.8	51.0 <sup>a</sup>	8.0	46.2 <sup>a</sup>	8.8	51.0 <sup>a</sup>	90.6	100.0
F	7.8	49.6 <sup>a</sup>	7.5	48.1 <sup>a</sup>	8.0	51.2 <sup>a</sup>	97.0	100.0
Leaves								
A	38.0	281.8a	36.0	267.6a	37.3	276.8a	95.0	98.2
B	39.0	307.6ab	36.5	287.9a	36.8	290.6ab	93.6	94.5
C	36.8	324.5bc	35.3	312.8ab	36.3	321.5bc	96.4	99.1
D	39.8	297.9ab	37.3	279.1a	39.0	291.8ab	93.7	98.0
E	38.7	300.9ab	36.3	283.0a	38.0	296.2ab	94.1	98.4
F	37.5	343.5c	36.5	334.4b	36.3	332.1c	97.4	96.7
CALCIUM Stems								
A	12.3	65.5ab	2.2	11.4c	4.5	23.9a	17.4	36.5
B	14.0	80.1c	1.2	6.7ab	4.7	26.7a	8.4	33.3
C	9.7	57.2a	1.7	9.8bc	3.5	20.8a	17.1	36.4
D	13.7	76.1bc	1.3	7.4ab	3.7	20.4a	9.7	28.8
E	11.2	64.1ab	2.0	5.7a	4.0	23.1a	8.9	36.0
F	10.8	68.8abc	1.0	5.4a	4.3	27.2a	9.3	39.5
Leaves								
A	24.7	181.4a	6.0	43.5	12.7	93.9	24.0	51.8
B	23.0	181.2a	5.8	46.4	12.7	100.6	25.6	55.5
C	21.7	191.8a	5.0	44.1	10.7	94.2	23.0	49.1
D	24.5	182.9a	5.5	41.1	12.5	93.5	22.5	51.1
E	23.0	178.8a	4.2	32.8	11.7	91.5	18.3	51.2
F	22.3	203.9a	5.8	52.7	12.5	114.5	25.8	56.2
MAGNESIUM Stems								
A	6.2	33.1a	2.3	12.5	5.0	26.7	37.7	80.7
B	5.7	32.4a	3.0	17.3	5.0	28.4	53.4	87.7
C	6.2	36.6a	3.3	19.8	5.7	33.6	54.1	91.8
D	5.2	28.7a	3.5	19.7	4.7	26.1	68.6	91.0
E	5.5	31.7a	2.5	14.3	4.7	26.9	45.1	84.9
F	5.0	32.0a	3.5	22.4	4.3	27.2	70.0	85.0
Leaves								
A	7.2	52.7a	5.5	40.7a	6.2	45.2	77.2	85.8
B	6.8	54.1a	5.2	40.8a	6.7	52.3	75.4	96.7
C	6.5	57.6a	5.7	50.1b	6.0	53.1	87.0	92.2
D	6.8	51.4a	5.3	40.0a	6.5	48.8	77.8	94.9
E	7.2	55.8a	5.8	45.3ab	7.0	54.6	81.2	97.8
F	7.3	66.9a	5.0	45.8ab	6.3	57.2	69.1	86.3

<sup>a</sup> In comparison between treatments and a particular form of element, means with like symbols do not differ significantly at the 5% level of probability.

cium, and magnesium. Potassium was determined by a flame photometer method (4) and calcium and magnesium by a chelometric titration (2, 3).

**Acid-Soluble Extraction.** A 200-mg. sample of tissue was allowed to soak for one hour at 6° C., in 50 ml. of a 3% acetic acid solution. The mixture was transferred to a homogenizing flask and homogenized for 2 minutes in a high speed Vir-Tis homogenizer. The resultant slurry was filtered through Whatman No. 42 paper, and the residue was washed with 3% acetic acid. To the filtrate, 15 ml. of concentrated perchloric acid (60 to 62%) and 15 ml. of concentrated nitric acid were added, and the entire mixture was evaporated to dryness. To the oxidized white residue, 10 ml. of 0.01N nitric acid was added to solubilize the residue.

The solution was filtered through Whatman No. 42 paper into a 100-ml. volumetric flask and aliquots of 10 ml. each were treated to remove interfering phosphate (2). The water extraction was conducted in the same manner except that deionized water was used in place of acetic acid. Since these were separate extractions, the data presented herein reflect some overlapping, —e.g. some water-soluble forms are extracted with the acid-soluble extraction.

**Results and Discussion**

The leaf tissue of tomato comprised the largest proportion of the total dry vegetative weight produced above ground (Table II). Soil treatment had no significant effect on the amount of stem tissue produced, but did influence the amount of leaf tissue produced. The

**Table IV. Total Uptake of Potassium, Calcium, and Magnesium by Tomato Plants<sup>a</sup>**

Treatment	Plant Uptake, <sup>b</sup> Mg./Pot		
	K	Ca	Mg
A	334.9a	246.9a	85.9a
B	360.8ab	261.3a	86.5a
C	376.0bc	249.0a	94.2a
D	353.5ab	259.0a	80.1a
E	351.9ab	242.9a	87.5a
F	393.1c	272.7a	98.4a

<sup>a</sup> Total nutrient in stem and leaf portions.

<sup>b</sup> In comparison of an individual nutrient with treatments, means with like symbols do not differ significantly at the 5% level of probability.

largest amount of leaf tissue was associated with the highest application of potassium. There was no reduction in plant growth as a function of the high potassium application. Reports of potassium inhibiting magnesium uptake and, hence, restricting growth are numerous (7, 8, 14).

The concentration and amounts of potassium fractions in tomato stems and leaves are shown in Table III. The values for total potassium agreed with those previously reported (7, 9). Stem potassium was not affected significantly by soil treatment. Approximately 95% of the total stem potassium was extracted with water and 3% acetic acid.

The largest total amount of leaf potassium resulted from the high rate of treatments of Sul-Po-Mag and potassium alone. A high rate of potassium with magnesium in the granule form resulted in a depression of leaf potassium when compared with potassium applied alone. Approximately the same proportion of leaf potassium was extracted with water and acid as was found for stem potassium.

Leaf tissue contained a higher concentration of potassium than did the stem tissue. This is in agreement with what has been reported for the potassium distribution in timothy and bromegrass (6).

The largest amount of total stem calcium resulted from the low rate of magnesium application (Table III). The high rate of magnesium and/or potassium applied reduced the amount of stem calcium. There was little effect of soil treatment on water- and acid-soluble stem calcium. Approximately 23% of the total stem calcium was extracted with water and acid as compared with four times more potassium extracted with the

same solvents. Acid extracted about three times more stem calcium than did the water extraction. Approximately 60% of the calcium in the stems was in a chemical form(s) not soluble in acid. There were no significant differences in total, water- and acid-soluble leaf calcium as a consequence of treatment. More leaf calcium was extracted with water and acid than was stem calcium; however, about 50% of the leaf calcium was not soluble in either water or acid. More of the leaf calcium was acid soluble than was stem calcium. The higher concentration of calcium in tomato leaves is in agreement with the accumulation of calcium in the leaves of certain grasses (6).

The total amounts of calcium in stems and leaves compared favorably with that reported earlier for tomato (7, 9).

No significant differences in plant magnesium fractions in tomato stems resulted from the soil treatments (Table III). Approximately 70% of the stem magnesium was extracted with water and acid. This was about twice the amount of stem calcium extracted with water and acid. No significant differences were observed for total and acid-soluble leaf magnesium as a result of the treatments. The largest amounts of water-soluble leaf magnesium occurred with the high rates of potassium and magnesium applications. Application of potassium to the soil did not reduce significantly the content of magnesium in the stems and leaves of tomato plants. In general, more leaf magnesium was soluble in acid and water than was stem magnesium. A greater proportion of the leaf magnesium was extracted with acid as compared with water which was similar to the extraction pattern for calcium. Over half of the stem and leaf magnesium was water soluble which agrees with previous work (12, 13) for grasses and clovers.

The total content of magnesium in tomato stem and leaf tissue was within the range previously reported (7, 9).

These results show that, particularly for calcium and magnesium, these elements existed in different chemical forms as a function of anatomical tissue. A better understanding of specific chemical forms of nutrients within certain plant parts would aid in interpreting soil-plant-animal relationships. Todd (11) has focused attention on the importance of chemically characterizing plant magne-

sium with respect to animal ingestion and utilization of the plant magnesium. The same can be expressed for potassium and calcium.

Slight differences in total potassium uptake were observed as a result of treatment (Table IV), however, there were no significant differences in the total plant uptake of calcium and magnesium. Application of potassium did not reduce magnesium uptake.

#### Acknowledgment

The authors express their appreciation to N. S. McNutt for his aid in the chemical analyses. This research was supported in part by a grant-in-aid from the International Mineral and Chemical Corp., Skokie, Ill., and by Western Regional Research Funds, W-67.

#### Literature Cited

- (1) Altman, P. L., Dittmer, D. S., "Biology Data Book," Federation Amer. Societies Exptl. Biology, Washington, D. C., 1964.
- (2) Bullock, J. S., Johnson, G. V., Maier, R. H., Fuller, W. H., *Soil Sci. Soc. Am. Proc.* **25**, 515 (1961).
- (3) Cheng, K. L., Bray, R. H., *Soil Sci.* **72**, 449 (1951).
- (4) Jackson, M. L., "Soil Chemical Analysis," Prentice-Hall, Englewood Cliffs, N. J., 1958.
- (5) Mills, C. F., *Soil Sci.* **85**, 100 (1958).
- (6) Pritchard, G. I., Pigden, W. J., Folkins, L. P., *Can. J. Plant Sci.* **44**, 318 (1964).
- (7) Reuther, W., ed., "Plant Analysis and Fertilizer Problems," American Institute Biological Sciences, Washington, D. C., 1961.
- (8) Russell, E. J., "Soil Conditions and Plant Growth," 9th ed., Longmans, Green & Co., London, 1961.
- (9) Spector, W. S., ed., "Handbook of Biological Data," W. B. Saunders Co., Philadelphia, Pa., 1956.
- (10) Stanberry, C. O., Cooper, L. R., Maier, R. H., Wilson, J. P., *Proc. Am. Soc. Hort. Sci.* **85**, 672 (1964).
- (11) Todd, J. R., *J. Agr. Sci.* **56**, 411 (1961).
- (12) *Ibid.*, **57**, 35 (1961).
- (13) *Ibid.*, **58**, 277 (1962).
- (14) Wallace, A., "Solute Uptake by Intact Plants," A. Wallace, Los Angeles, Calif., 1963.

Received for review June 24, 1965. Accepted January 26, 1966. Published with the approval of the Director of the Agricultural Experiment Station as Technical Paper No. 1015.