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Chloroplast pigments in relation to magnesium deficiency.

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CHLOROPLAST PIGMENTS IN RELATION TO MAGNESIUM DEFICIENCY

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**CHLOROPLAST PIGMENTS IN RELATION
TO MAGNESIUM DEFICIENCY**

By

Dale Albert Hinkle

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**Presented in Partial Fulfillment of the Requirements
for the Degree of Doctor of Philosophy at the
University of Massachusetts, Amherst, Massachusetts.**

May 1949

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INTRODUCTION

Although magnesium has long been recognized as a necessary element for plant growth, it has been only within the last twenty or thirty years that any attention has been given to a deficiency of this element. Previously magnesium deficiency either did not exist or its symptoms were not recognized in the field. In all probability a deficiency of the element did not exist previous to this time for two reasons. First, the amount of magnesium used by plants is small, and, therefore, the drain upon the soil is not excessive. Secondly, the amount of this element formerly supplied as an incidental constituent was generally sufficient to take care of most crop withdrawals. Organic ammoniates which formerly supplied a tenth of all the magnesium in commercial fertilizers are now largely used in feeding stuffs where they command a higher price. The greatest loss of magnesium as an incidental constituent in fertilizers has been the use of refined potassium salts as a source of potassium in mixed fertilizers. Unrefined potassium salts such as kainite and manure salts, which were once widely used, contained relatively large amounts of magnesium.

Few studies have been made of the effects of magnesium deficiency upon the chloroplast pigments. Some of the recent studies have been concerned with the influence of the potassium ion upon magnesium deficiency and the amount

of magnesium in the leaves of certain plants, but no one has studied the influence of other cations in the same periodic group with potassium. Only one study has been made of the effects of order of plant development in relation to magnesium deficiency.

The purpose of the present investigation was to study the effects of magnesium deficiency on the chloroplast pigments, to determine the influence of the alkali cations on magnesium absorption, and to study the relationship between the order of plant development of certain plants and the degree to which they are affected by magnesium deficiency.

REVIEW OF LITERATURE

No complete review was made of the literature dealing with all phases of magnesium deficiency. Papers were reviewed on the three phases of the problem which were selected for study and a few which were of general interest.

Magnesium Deficiency Symptoms of Some Common Crops

Although magnesium deficiency has been reported to occur on soils both high and low in exchangeable calcium, Boynton (4) reports that for apples it generally is found on soils low in exchangeable calcium and also low in total exchange capacity. Apparently the symptoms of deficiency in apple leaves are somewhat different from those in other plants. Boynton describes them as follows:

"The first visible evidence of magnesium deficiency in apple trees is a fading between the large veins of the older leaves on some shoots or spurs. The faded areas may turn very pale yellow and remain so for some time, or they may scorch rapidly; in either case, necrotic brown 'blotches' develop from them. Some of the dead areas between veins may remain as separate islands surrounded by green tissue, but many of them tend to coalesce as they grow in size, and where they grow together at the leaf margin may produce a marginal leaf scorch indistinguishable from that resulting from potassium deficiency in its later stages."

These symptoms usually developed in late summer after all vegetative growth has stopped and flower initiation has gone on for some time.

Jones (20) has reported magnesium deficiency in corn grown on a sandy loam soil underlaid by a gravelly fine sand at the Massachusetts Agricultural Experiment Station. He

described the symptoms as a striping of the leaf due to loss of green color between parallel veins. These symptoms appeared early in the season, usually about two weeks after emergence. As the season progressed the light green color of the inter-vascular tissue continued to fade until all green color was lost from the leaf. At about this time the leaf ceased to function as a part of the living plant.

In discussing magnesium deficiency in tobacco, McMurtry (26,27) states that symptoms rarely appear in the field until the plants have obtained considerable size. This delay mitigates against affecting leaf size to any considerable extent. The lower leaves first lose their normal green color, starting at the tip and side and working back toward the center and basal parts. Leaves higher up on the plant become affected only after those lower down have become severely affected. The parts of the leaf where the chlorophyll has been destroyed become pale green at first and later may fade to almost white. Apparently not only is the chlorophyll affected, but the yellow pigments as well.

Garner and associates (15) were the first to report magnesium deficiency in tobacco. Since the disease occurred frequently in sandy areas in fields it was given the name "sand drown". Magnesium deficiency is now so common in tobacco grown in the Connecticut Valley and the Atlantic Coastal Plains soils that fertilizer intended for use on this crop in these areas normally carries two per cent of soluble magnesium.

The potato is another crop in which magnesium deficiency is widespread. According to Jones and Brown (19) symptoms of magnesium deficiency in this plant have been observed in all the potato districts of the Atlantic Coastal Plain and as far north as Maine. They described the symptoms as follows:

"The foliage of affected plants is lighter in color than normal. The lower leaves are the first to become affected, since some of their magnesium is withdrawn to be used over again by the new growth. In mild cases only the lower leaves of the plants show symptoms, while the new growth appears healthy. Loss of green color begins at the tips and margins of the lowermost leaves and progresses between the veins toward the center of the leaflets. In advanced states of deficiency the central portion of the individual leaflets becomes chlorotic between the veins and eventually is filled with small brown dead areas. Break-down of the tissue, like loss of color, usually starts at the tip and the terminal leaflet is generally the one most severely affected. The lower leaves of magnesium-deficient plants are brittle, which serves to distinguish them from leaves yellowing naturally through age."

The onset of symptoms can be correlated with rainfall. They usually appear soon after periods of rainy weather. The leaves of deficient plants were found to be very low in magnesium and calcium, but very high in nitrogen. When leaves age normally they have a high calcium and magnesium content and low nitrogen content.

Cooper (9) describes magnesium deficiency in cotton as follows:

"The characteristic magnesium-deficiency symptom in cotton is a purplish-red leaf with green veins. Late in the season it is sometimes difficult to distinguish between the color due to magnesium deficiency and that due to age or maturity, though the latter is apt to be orange rather than purplish red."

Apparently cotton differs from most other plants in that anthocyanin is produced in the leaf following breakdown of the chloroplast pigments. Anthocyanin is universally correlated with an excess of simple sugars. It would seem possible that a lack of magnesium might prevent translocation of carbohydrates out of the leaf. Since magnesium is a carrier of phosphorus, a deficiency of the former might depress the intake of the latter. Phosphorus in turn is known to be necessary for translocation of sugars. In fact the purplish color due to anthocyanin is normally a symptom of phosphorus deficiency in many plants.

Magnesium deficiency is a major problem in all soils in Florida used for citrus (7). Symptoms consist of irregular yellow blotches which start along the midrib of leaves adjacent to the fruit. These eventually coalesce to form an irregular band along both sides of the midrib which gradually enlarges to cover the entire leaf. At this stage the leaf is dropped from the tree. Symptoms may occur at any time during the season, but usually they occur in late summer or early fall.

According to Skinner (30) magnesium deficiency in truck-crops is extensive and widespread in the Atlantic and Gulf Coastal soils. These soils are naturally low in fertility due to their sandy nature. Excessive leaching under cropping conditions have tended to remove what little magnesium these soils may have had. Furthermore, the extensive use of high nitrogen containing fertilizers has

greatly mobilized the soil magnesium. Symptoms in vegetable crops are similar to those of other crops. In tomatoes the lower leaflets are brittle and there is a loss of chlorophyll between the veins. Deficiency in cabbage is manifested by chlorosis of the lower leaves plus a sort of mottled, puckered appearance. Turnips develop a brown area around the leaf rim which dries up and falls out, while the interior part of the leaf is chlorotic and mottled.

From the foregoing review it is evident that magnesium deficiency symptoms follow a general pattern in most plants. The lower leaves are first affected. This is because magnesium is being withdrawn from them to satisfy nutritional requirements at points of growth. In more advanced stages chlorosis moves up the plant toward the meristem which is rarely affected.

Magnesium deficiency may be expected to occur on light, sandy soils that are very acidic in nature. The use of acid forming fertilizers on any soil greatly increases magnesium losses by leaching.

Effect of Magnesium on Chloroplast Pigments

Since the classical researches of Willstätter and Stoll (37) have shown that magnesium is the only metallic constituent in the chlorophyll molecule, it is reasonable to expect that a lack of it would lead to less chlorophyll production. That the amount of chlorophyll is proportional to the amount of magnesium in the nutrient medium has been shown to be true by Mameli (23). In studying the effect of

magnesium on the development of chlorophyll in algae, corn, sunflower and buckwheat, she found the amount of coloring matter extracted was in direct proportion to the magnesium content of the culture solution.

Willstätter and Stoll extracted chlorophyll from a wide variety of plants and always found its magnesium content to be 2.7 per cent. Javillier and Goudchaux (18) extracted chlorophyll from twenty-two widely varying species of plant leaves and determined the amount of magnesium contained in the extract. They found the amount of magnesium contained by the chlorophyll varied from 0.01 to 0.03 per cent of the dry leaf weight. In nine of these species they determined total leaf magnesium and calculated the chlorophyll-contained magnesium as a percentage of the total magnesium. Although their figures varied from 0.8 to 26 per cent, a majority of them fell within the range of 3 to 12 per cent. Most of the variation was due to wide differences in the total leaf magnesium rather than to variations in the chlorophyll-contained magnesium.

Michael (24) studied the effects of magnesium deficiency in sand and water cultures on chlorophyll synthesis in oats, beans, and corn. He found that chlorophyll formation was greatly reduced in cultures in which magnesium was left out. The amount of chlorophyll-bound magnesium in proportion to the total magnesium was small, rising in only a few cases to as much as 20 to 30 per cent. In the case of visible magnesium deficiency the chlorophyll-bound magnesium did not

increase, but in many cases actually decreased. He suggests the possibility of characterizing the degree of deficiency by the ratio of chlorophyll-bound to total magnesium.

Magnesium deficiency not only caused a decrease in chlorophyll, but also lowered the carotene and xanthophyll content of bean leaves. The protein content of the magnesium deficient leaves was also reduced. It was assumed that magnesium deficiency caused a derangement of the carbohydrate and protein economy of the plant and that injury to the chloroplasts was a result of this condition.

It is evident that magnesium must play some role in plant economy other than chlorophyll synthesis since, as we have seen from the results given above, the chlorophyll-bound magnesium always constitutes less than one-quarter, and in most cases, less than one-seventh of the total leaf magnesium. It is believed that it acts as a carrier of phosphorus. Truog and associates (32) have recently thrown additional light on this idea. In peas which were fertilized either with dolomitic limestone or magnesium sulfate the seeds showed an increase not only in magnesium but in phosphorus as well. In fact, increasing the available magnesium in the soil increased the phosphorus content in the pea seeds more than the addition of phosphate fertilizers.

Work by Emerson (13) and Fleischer (14) indicates that magnesium plays a specific role in photosynthesis. Emerson was able to control the chlorophyll content of the

unicellular alga Chlorella by varying the iron content of the culture medium. He was able to show that there is a positive straight line relation between the rate of photosynthesis and chlorophyll concentration. Fleischer repeated Emerson's experiment and fully confirmed his results. In addition, he extended the methods of controlling chlorophyll concentration to varying nitrogen and magnesium. The results for nitrogen experiments were very much like those in which iron was varied, but when magnesium was varied, the rate of photosynthesis was checked long before chlorophyll formation was affected. He concluded from these results that magnesium plays a role in photosynthesis in addition to the formation of chlorophyll.

Kennedy (21) also studied the effect of magnesium deficiency on photosynthesis in Chlorella cells and confirmed Fleischer's results; namely, that a rise in chlorophyll concentration due to more magnesium in the nutrient medium does not cause a corresponding rise in the rate of photosynthesis such as would occur if chlorophyll concentrations were varied by other means. However, when flashing light was used to illuminate the cells and the length of the dark period was varied, photosynthesis per flash per gram of chlorophyll was immensely augmented by increasing the length of the dark period in case of cells rendered chlorotic by growth in magnesium deficient media. No corresponding increase was noted in the case of chlorotic cells due to iron deficiency or in cells grown in full nutrient solution.

Therefore, it was concluded that magnesium is involved in the so-called Blackman reaction.

A few studies have been made of the ratio of the chloroplast pigments in chlorotic and non-chlorotic leaves. Willstätter and Stoll observed that the ratio of the green to yellow pigments in green leaves was remarkably constant. Oserkowsky (28) studied the ratio of the pigments in chlorotic and non-chlorotic pear leaves. Chlorosis in this case was due to growth of the pear trees in highly calcareous soil which induced iron deficiency. Normal leaves were taken from trees which had been sprayed with soluble iron salts. He did not separate carotene from xanthophyll but determined them together as a unit. He states that the coefficient of correlation between chlorophyll and yellow pigments for thirty-one samples of chlorotic and normal leaves was 0.934. The ratio of chlorophyll to yellow pigments was higher in the non-chlorotic leaves than in the chlorotic ones. For example, in eight samples which he described as "severely chlorotic" it averaged 1.8; in nine "moderately chlorotic" samples it was 2.7; and in five samples of "green leaves from treated trees" it was 5.1. The smaller ratio for the chlorotic leaves indicates a much greater deterioration of chlorophyll than of yellow pigments.

Mackinney (25) grew barley seedlings in nutrient solutions deficient in iron. His results indicated that there is a close relation between chlorophyll and the carotinoids, but that the carotene to xanthophyll ratio was markedly

lower in chlorotic leaves. No one seems to have made a study of the ratio of the three pigments when affected by magnesium deficiency.

Holmes and Crowley (17) and Holmes, Crowley and Kuzmeski (16) determined carotene in Boston head lettuce and kale which were grown in field plots treated with lime and magnesium and magnesium alone. No consistent differences in the carotene content of either crop was found that could be attributed to the magnesium treatment. In the case of lettuce, this may have been due to the failure to include the outside leaves.

Brown (6) and others grew Swiss chard in the greenhouse with nutrient solutions in which various elements were left out. Significantly lower carotene content of the chard was obtained from plants receiving insufficient nitrogen, magnesium and iron. On the other hand, calcium, manganese, potassium and phosphorus had no significant effect on the carotene content.

Wynd (38) studied the relation of the chemical properties of the upper eight inches of soil in an area near Midland, Kansas to the carotene content of immature oat leaves. He found the percentage of nitrogen, the base exchange capacity, and the amount of replaceable calcium and magnesium affected the amount of carotene in the leaves. Replaceable potassium and weak acid-soluble phosphorus had no effect.

A summary of these papers reveals the following points:

(a) the chlorophyll-bound magnesium represents a small part of the total leaf magnesium, being within the range of 3 to 15 per cent, (b) magnesium plays a specific role in photosynthesis and is probably involved in the Blackman reaction, (c) nitrogen, magnesium, iron and possibly calcium may influence the carotene content of leaves; but potassium, phosphorus and manganese do not, (d) nothing is known concerning the effects of magnesium deficiency on the ratio of the chloroplast pigments, and (e) the number of studies which have been made concerning the effects of magnesium on the yellow pigments are so few and with so few different plants that additional studies are needed.

Effect of Alkali Cations on Intake of Magnesium

A review of the literature on the effect of alkali cations on the intake of magnesium by plants indicates few studies have been made, except on the influence of the potassium ion.

There are a considerable number of reports on the influence of potassium because fertilization of acid, sandy soils with this element has led in many cases to magnesium deficiency.

Wallace (36) made a study of magnesium deficiency in apples, currants, gooseberries and plums in England. Potassium deficiency was never accompanied by magnesium deficiency, but the latter frequently appeared upon the correction of the former. He states that liberal use of potassium fertilizer may be followed by magnesium deficiency

and that there is little doubt that magnesium requirements are increased by use of potassium. It seems that the latter part of this statement may not necessarily be true.

Potassium probably does not increase the need of the plant for more magnesium, but rather prevents its intake in sufficient amounts.

Kidson, Askew and Chittenden (22) studied magnesium deficiency in apples in New Zealand. They found premature defoliation on trees grown on soil which had received liberal amounts of potassium. Apple leaves from these trees had an abnormally high potassium content and a very low magnesium content. They concluded that the use of high applications of potassium on acid, sandy soils may lead to reduced intake of magnesium due to an unfavorable potassium-magnesium ratio in the soil.

Southwick (31) reports analysis of apple leaves showing different degrees of leaf scorch and found a correlation in the degree of scorch and the magnesium and potassium contents of the leaf. The data suggests that use of potassium fertilizers increased the probability of magnesium deficiency. The critical magnesium level in the leaves for the four varieties tested was 0.25 per cent. However, when the potassium level was high, even this amount did not prevent scorch. He suggests that in commercial orchards potassium should be left out of the fertilizer program until the magnesium content of the soil can be built up.

Tung trees grown in Florida often developed marginal

leaf scorch (11). Analysis of the leaves showed that the affected leaves were much lower in magnesium than normal leaves, and that applications of potassium chloride greatly lowered the magnesium content of the leaves and increased the amount of scorch.

Boynton and Burrell (5) made a study of the effects of potassium on the intake of magnesium in apple leaves in New York state. One of two contiguous rows of McIntosh apples was given five annual applications of potassium sulfate and the other none. Nine of the trees in the potassium-fertilized row developed leaf scorch while none in the row receiving no potassium developed scorch. These nine trees were paired with the nine trees in the adjacent row and samples consisting of fifty leaves from each tree were taken. Composite soil samples consisting of eight borings per tree also were taken. Results indicated that potassium fertilization caused an increase in leaf potassium and exchangeable potassium of the soil, and reduced to a remarkable degree the leaf magnesium and the exchangeable magnesium of the soil. There was a slight reduction in leaf calcium and exchangeable calcium of the soil.

Wallace and associates (35) grew potatoes on plots which during the previous fifteen years had been used for a fertilizer test on black currants. No fertilizer was used in the year in which the potatoes were grown, as it was desired to study the residual effects of previous applications. Leaf symptoms of potassium deficiency were noted

where potassium had not been applied and magnesium deficiency where it had.

Walsh and O'Donohoe (33) found that in potatoes a high level of potassium induced severe magnesium deficiency and also reduced the calcium content of the foliage. On the other hand, the severity of potassium deficiency was positively correlated with the exchangeable magnesium in the soil. Similar results were found to hold true with tobacco, but with this crop high potassium level at low sulfur level did not induce magnesium deficiency. With sugar beets the depressing effect on magnesium intake by potassium was not as pronounced as with potatoes and tobacco. With sugar beets it was also noted that a high level of potassium reduced phosphorus absorption. This effect, however, was probably an indirect effect of lowered magnesium absorption, since the latter has been shown to influence the absorption of phosphorus.

Walsh and Clarke (34) made a thorough study of magnesium deficiency in the tomato. The most important single factor affecting the absorption of magnesium was found to be potassium. The ratio of potassium to magnesium in the foliage was found to be more important than the actual amount of these elements in determining the severity of chlorosis. The extent of control of chlorosis by applications of magnesium sulfate was determined by the potassium level, since potassium was found to depress the intake of magnesium. Chlorosis was observed at high magnesium levels when potassium was also very high.

From the foregoing review it seems evident that the three cations, calcium, magnesium and potassium, mutually influence one another in the processes of absorption by plants. High potassium levels in the nutrient medium reduce the intake of magnesium and calcium, but the intake of magnesium to a much greater extent than that of calcium. High magnesium levels reduce calcium intake to some extent and potassium intake to a much greater extent.

Effect of Magnesium Deficiency on

Different Botanical Species

Eisenmenger and Kucinski (12) have made the interesting observation that among seed plants those of low order of evolutionary development suffer a great deal more from magnesium deficiency than those of high order of development. They observed the growth of a large number of plants belonging to different families on a magnesium deficient plot at the Massachusetts Agricultural Experiment Station. This plot of ground was known as the North Corn Acre and will be referred to again in this report. Nearly all plants belonging to low stages of development became chlorotic. Many in an intermediate stage of development also showed symptoms of magnesium deficiency, but only a few plants of high order of development ever became chlorotic. Among those that did become chlorotic were a few isolated members that have been greatly changed by man through domestication. Corn is a good example of such a plant. Belonging to the Gramineae family it is considered to be very highly

developed, yet it developed chlorosis.

The most bothersome weeds in the eastern United States belong to the highly developed Compositae, Graminaceae and Convolvulaceae families. It was noted that such weeds were as persistent in the magnesium deficient plots as in the magnesium-treated plots, while certain other weeds, such as purslane, wood sorrel and chickweed, all of which are of lower plant orders, did not grow where magnesium was not added.

In tests with seventeen crops on these same plots Beaumont and Snell (2) observed that buckwheat, spinach, turnips, mangels, corn and tobacco were the most severely affected by magnesium deficiency. Small grains, grasses, clover and potatoes were only slightly affected. Additions of magnesium sulfate increased the magnesium content of the crops greatest in those in which the yield and appearance were most affected.

Collander (8) found that certain families of plants have the power to selectively absorb more of some of the cations than others. He found that members of Chenopodiaceae family and buckwheat were rich in magnesium while peas, vetch and oats were relatively poor in it.

Collander's results with the two legumes, peas and vetch, seem to be at variance with results of other workers. Daniel (10) found that the average magnesium content of nineteen species of mature pasture and range grasses grown in Oklahoma was 0.156 per cent while eight species of

mature legumes contained on an average 0.379 per cent magnesium.

Beeson (3) has summarized a large number of analyses of common farm crops. Inspection of his data shows that the average magnesium content of fifteen legumes was 0.36 per cent while eight grasses averaged 0.17 per cent. These analyses show beyond any doubt that legumes have a greater need for magnesium than grasses. Beeson gives the average content of buckwheat and spinach as 0.60 and 0.53 per cent respectively, which is high compared to other plants. This again indicates a considerable need for magnesium by these plants.

EXPERIMENTAL PROCEDURE

History of Field Plots

Samples of plant leaves which were analysed for pigments were obtained from a plot of land known as the North Corn Acre at the Massachusetts Agricultural Experiment Station. This field was first laid out in 1891 by dividing an acre of land into four equal plots, running in a north-south direction. These plots were used in a corn fertilizer test until 1920 when the original purpose was abandoned and an experiment was started to test the residual effects of the previously long continued applications of potassium and phosphorus. Corn was again the test plant in this second experiment which was continued until 1928.

In 1920 chlorosis of corn leaves was noted for the first time. It is probable that it had occurred before this time in mild form and had escaped detection. The plots had not received any lime in the preceding thirty years, so aluminium toxicity was suspected. Accordingly, the north one-half of all plots was given a cross treatment of lime in 1921. Liming did not eliminate the chlorotic condition in the leaves of succeeding crops. Since Garner and his associates (15) had just reported a chlorosis on tobacco, called "sand drown," as being due to magnesium deficiency, it was decided to apply a cross treatment of magnesium sulfate to a strip across the central part of all plots in 1924. This and later magnesium treatments

greatly reduced the amount of chlorosis and indicated it had been due to magnesium deficiency. The results of these experiments were reported by Jones (20).

The cross treatments of lime and magnesium provided four new plots running perpendicular to the original four. In 1929 a third experiment was started on these same plots to determine the relative value of magnesium for the more common crops and to obtain descriptions of deficiency symptoms for these crops. The lime and magnesium cross treatments were continued, but the amounts varied from year to year and were omitted in some years. In 1934, when this experiment was discontinued, seventeen varieties of crops had been grown and observed. These results were reported by Beaumont and Snell (2).

From 1935 to 1947 all four plots have received annual applications of 1000 pounds per acre of a fertilizer made from Ammo-Phos, sodium nitrate and potassium sulfate in proportion to give a 5-8-7 analysis. The limed plots have received during this period annual applications of 1000 pounds per acre of magnesium-free lime. The magnesium treated plots have received 150 pounds of magnesium sulfate per acre each year. Figure 1 shows the experimental plan and treatments during this period.

A large variety of plants have been grown on this area during this period of thirteen years. These have included trees such as peach, apple, elm, oak, willow, black locust, maple and many others. The list also includes flowering

shrubs, ornamental annuals and perennials. Vegetable crops which have been grown include tomato, potato, cabbage, carrot, lettuce, bean, pepper, and egg plant. The purpose behind such a wide variety of plants was to observe the differential effects of magnesium deficiency on plants of different order of evolutionary development.

North Side

<p>Plot D</p> <p>1000 pounds of 5-8-7 fertilizer per acre plus 1000 pounds of limestone per acre</p>
<p>Plot C</p> <p>1000 pounds of 5-8-7 fertilizer per acre plus 1000 pounds of limestone per acre plus 150 pounds of magnesium sulfate per acre</p>
<p>Plot B</p> <p>1000 pounds of 5-8-7 fertilizer per acre plus 150 pounds of magnesium sulfate per acre</p>
<p>Plot A</p> <p>1000 pounds of 5-8-7 fertilizer per acre</p>

South Side

Figure 1. Experimental Plan and Placement of Treatments in the North Corn Acre for the period 1935 to 1947, inclusive.

The study of Eisenmenger and Kucinski (12) concerning the relation of the order of plant development to the need of magnesium was based on observations made of plants grown on these plots. These plots were also used by Holmes and Crowley (17) and Holmes, Crowley and Kuzmeski (16) in their study on the carotene, calcium, magnesium and phosphorus content of Boston head lettuce and kale.

It should be noted that in the earlier studies of Jones (20) and Beaumont and Snell (2) dealing with these plots the same letters A, B, C, and D were used to designate them by starting at the north side with Plot A. In the later reports (16, 17) they were designated by starting with Plot A at the south side. Thus the placements of lime and magnesium have not changed in the twenty-seven years during which they have been used, but the letters used to designate the plots were changed. Plot A was originally called Plot D, Plot B was Plot C, and so on.

Collection and Preparation of Leaf Samples

It was frequently impossible to collect samples for the same species from all four of the field plots since some plants did not grow on the unlimed plots because of too high acidity.

An attempt was made to take samples of leaves of approximately the same age on the plant. This was frequently difficult due to variation in growth and in the effect of disease which affected some plants on the different plots more than others. Magnesium deficient plots often produced

diseased plants whereas the same plant would be practically free from disease on the magnesium treated plots.

In all cases the samples consisted of lower leaves, since these are affected first by magnesium deficiency. Although an attempt was made to take the samples at random from several plants, it is entirely possible that representative samples were not always obtained and this may account for some of the inconsistencies in the data obtained.

The samples were taken from the field plots at various times from June 5 to August 29. As little time as possible was allowed to elapse between the time the samples were picked from the plant and the time the pigments were extracted. This generally was not more than an hour. Usually a set of four samples was taken in the morning of one day and all analyses finished by noon of the next. Precaution was taken to prevent as much loss of pigments by thermal and light decomposition as possible.

On August 21 and again on August 29 sets of samples were taken and placed in a deep freezer at -20°C . The longest time that any one sample remained frozen was approximately six months. In order to determine if any decomposition of pigments occurred during the time the samples remained frozen, a quantity of sugar maple leaves and tobacco leaves were picked. These were cut up into small pieces and thoroughly mixed. One set of samples was withdrawn at once and the pigments determined. The rest were placed in Mason fruit jars and put in the deep freezer.

Sets of samples were withdrawn at various times and analyzed. Table 1 gives the results of these analyses.

Table 1. Effect of Deep Freeze Temperatures on Chloroplast Pigments

Days Stored	Chlorophyll mg./gm.	Xanthophyll mg./gm.	Carotene mg./gm.
Sugar Maple Leaves			
0	6.60	0.85	0.38
30	6.38	0.82	0.36
60	6.45	0.81	0.36
102	6.05	0.77	0.34
180	6.05	0.83	0.38
Tobacco Leaves			
0	11.35	1.47	0.68
30	11.08	1.40	0.60
60	11.60	1.38	0.58
102	11.40	1.34	0.58
180	11.30	1.42	0.66

Tobacco and sugar maple leaves were chosen because these two varieties of leaves represent about the extremes which one might expect in moisture content. Tobacco leaves are "wet" leaves, containing upwards of 90 per cent moisture when picked in the field. On the other hand, sugar maple leaves represent a "dry" type, usually having only about 45 to 50 per cent moisture. It was thought that the amount of moisture might have an effect on the keeping quality of the leaves under deep freeze conditions.

The results showed that moisture was not a factor in

keeping quality of the leaves and that very little decomposition of the pigments occurred for the period during which they were stored.

Methods Used for Pigment Analysis

Chlorophyll was determined by the Photoelectric Colorimetric Method for Total Chlorophyll (1, pp. 140-141), using a Klett-Summerson Glass Cell Industrial model. In this method the pigments are extracted from fresh plant material by grinding with pure quartz sand and 85 per cent acetone. A Waring Blendor was used for samples high in moisture. Chlorophyll in the extract, after having been filtered and made to volume, was determined by reading the amount of light absorbed by a one centimeter column of the extract in a photoelectric colorimeter, using light filter No. 64. A calibration curve for the instrument was prepared by diluting a fresh extract of Norway maple leaves with 85 per cent acetone, and determining the absorption of the various dilutions. A portion of the original acetone extract was transferred to diethyl ether and its chlorophyll content determined by use of a Beckman Spectrophotometer. By calculating the concentrations of the various dilutions it was possible to construct a calibration curve for the instrument.

Carotene was determined by the Barium Hydroxide Method (1, pp. 144-145) on an aliquot of the original acetone extract used to evaluate chlorophyll. In this method chlorophyll is separated from the yellow pigments by absorption on finely divided barium hydroxide which is

removed from the acetone extract by filtration. The yellow pigments remaining are transferred to petroleum ether, and xanthophylls are removed by extraction with 90 per cent methyl alcohol saturated with petroleum ether. Carotene which remains in the petroleum ether phase is dried with anhydrous sodium sulfate and made to volume. Light absorption of a one centimeter column was determined with the colorimeter, using a light filter No. 44. A calibration curve was prepared by making various dilutions of a pure beta carotene solution of known concentration and determining their absorptions values. Carotene used for preparing the original solution was purchased as chemically pure carotene and was further purified by recrystallization.

Xanthophyll was determined by a method devised by the author after corresponding with two authorities in the field of pigment research.¹ The methyl alcohol extracts of xanthophyll were saved and made to volume and light absorption determined as in the case with carotene. The calibration curve for carotene was used to convert these readings into milligrams of xanthophyll per liter. This is possible since the absorption coefficients for xanthophylls are nearly the same as those for carotene.

All pigment results must be expressed on the basis of dried plant tissue, since water content of such tissue varies a great deal. Therefore, simultaneous with weighing

1. Private correspondence with Drs. C. L. Comar and L. A. Moore.

of duplicate samples for pigment analysis, duplicate samples were weighed for moisture determination. These were dried in a steam oven at 60 to 70°C. over night and then placed in an electric oven at 100°C. for another overnight period.

All results of pigment analyses are expressed in terms of milligrams of pigment per gram of oven dried tissue.

EXPERIMENTAL RESULTS

Results of Chlorophyll Analysis on Sample Taken

Early in the Season from North Corn Acre

Magnesium deficiency is not at once evident early in the growing season. New leaves on perennials in the spring are not often chlorotic, but become so after a time. Annuals do not chlorose until they have made some growth. The reason for this delay is that magnesium is mobile in the plant and is withdrawn from the older leaves to new regions of growth. Apparently this process can continue until a certain magnesium content of the leaf is reached at which time chlorophyll synthesis is interfered with. This critical magnesium content is more or less characteristic for different plants.

Practically no chlorosis was noted on the plants growing on the magnesium deficient plots until the last of May. Beginning with June samples were taken of various plants and at intervals during the month. Only the chlorophyll content was determined on these samples. The results of these analyses are reported on Table 2. In this and all tables to follow only the common name of the plant is used. Both the common and scientific names of all plants used in this investigation will be found in the Appendix.

All but one of the plants in Table 2 are perennial. These plants might be expected to suffer less from magnesium

deficiency as compared to annuals since they should have a more widely distributed and deeper root system and thus be able to reach more soil, particularly the subsoil, which is richer in magnesium than surface soil. Nevertheless, the group as a whole showed magnesium deficiency signs even early in the season.

Table 2. Effect of Magnesium Deficiency on Chlorophyll of Plant Leaves, taken in Early Summer, 1947¹

Plant	Sample Date	Plot A, (No Mg)	Plot B, (Mg)	Plot C, (Mg+Ca)	Plot D, (Ca)
		mg./gm.	mg./gm.	mg./gm.	mg./gm.
Rhubarb	June 5	11.09	12.36	13.03	10.37
Mock Orange	" 6	7.08	8.44	7.66	-----
Maltese Cross	" 9	-----	-----	8.32	5.52
Iris	" 9	5.43	5.20	5.82	5.50
Red Maple	" 12	6.55	8.32	7.54	6.31
Pinks	" 13	3.86	8.37	9.91	4.76
Sedum	" 16	7.03	7.72	7.36	8.36
Veronica	" 16	6.59	8.07	7.78	8.32
Coral Bell	" 18	6.22	4.98	5.20	6.19
Bush Honeysuckle	" 27	6.42	-----	8.94	6.29
Beans	" 30	2.98	12.05	9.33	6.84

1. For the period 1935-1947 all plots received annual applications of 1000 pounds of a 5-8-7 fertilizer. In addition plots B and C received annual applications of 150 pounds of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ per acre and C and D 1000 pounds of magnesium-free limestone per acre.

Among the group only iris, sedum, veronica and coral bell failed to increase the chlorophyll content of their leaves as a result of the annual additions of magnesium sulfate. Iris and veronica are highly developed plants and, as will be explained later, such plants do not suffer from magnesium deficiency as much as those lower in the scale of plant evolution. String beans and pinks responded to a remarkable extent from addition of magnesium sulfate. These plants are fairly primitive in the scale of evolution of seed plants.

It will be noted that where direct comparisons can be made seven out of nine of the plants had leaves lower in chlorophyll when grown on the unlimed magnesium deficient Plot A as compared to the limed magnesium deficient Plot D. These results are in agreement with those of other research workers in that magnesium deficiency is more severe on very acid soils.

The reason for a more severe deficiency of magnesium in acid soils as compared to the more basic ones is probably due to the fact that hydrogen ions have a very high replacing power for other ions in the clay complex. The replaced magnesium ions are readily leached from the soil. At more basic soil reactions hydrogen ions are far less numerous and therefore much less magnesium is replaced from the absorbed state.

No consistent differences can be observed between the leaf chlorophyll content of plants grown on the two magnesium

treated plots. Here apparently soil reaction is of no consequence. The amount of magnesium added each year is evidently sufficient to take care of any additional leaching losses that might occur on the unlimed plot.

Plants belonging to the same family may or may not behave in a similar manner to magnesium deficiency. For example, mock orange and coral bell belong to the Saxifragaceae family, and the former was chlorotic early in the season while no chlorosis was observed on coral bell. On the other hand, pinks and maltese cross belong to the Caryophyllaceae family and were about equally affected.

Results of Analysis for Total Leaf Pigments

on Samples from North Corn Acre

Taken During July and August

Samples of leaves were taken during the months of July and August and these were analysed for total chlorophyll, carotene and xanthophyll. Since samples were not always available for the same plant on all four plots the results are given in separate tables. Table 3 gives them for the unlimed plots and Table 4 for the limed plots.

Table 3. Effect of Magnesium Deficiency on the Chloroplast Pigments of Plants Grown in the Unlimed Plots¹

Plant	Chlorophyll		Xanthophyll		Carotene	
	Plot A	Plot B	Plot A	Plot B	Plot A	Plot B
	(No Mg)	(Mg)	(No Mg)	(Mg)	(No Mg)	(Mg)
	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.
Meadow Rue	1.84	6.70	0.31	1.01	0.18	0.34
Apple	5.49	4.54	0.57	0.55	0.29	0.28
Raspberry	5.48	8.10	0.43	0.56	0.17	0.22
Hibiscus	4.19	9.50	0.24	1.20	0.28	0.50
Mock Orange	6.26	8.54	0.89	1.10	0.27	0.42
Petunia	7.43	9.83	0.96	1.27	0.31	0.42
Tobacco	3.40	8.43	0.61	1.04	0.28	0.57
Tomato	2.04	12.20	0.39	1.33	0.30	0.73
Egg Plant	2.47	8.24	0.55	0.94	0.30	0.44
Thunbergia	5.92	6.64	0.81	0.90	0.33	0.37
Oak	8.05	5.46	1.03	0.75	0.38	0.29
Snapdragon	6.81	9.27	0.82	1.14	0.28	0.43
Marigold	11.93	10.64	1.54	1.44	0.28	0.22
Sweet Corn	4.68	10.45	0.59	1.23	0.32	0.71

1. See footnote of Table 2 for plot treatments.

Table 4. Effect of Magnesium Deficiency on the Chloroplast Pigments of Plants Grown in the Limed Plots¹

Plant	Chlorophyll		Xanthophyll		Carotene	
	Plot D	Plot C	Plot D	Plot C	Plot D	Plot C
	(Ca)	(Mg+Ca)	(Ca)	(Mg+Ca)	(Ca)	(Mg+Ca)
	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.
Raspberry	3.84	7.64	0.23	0.54	0.08	0.24
Apple	4.40	4.55	0.54	0.51	0.27	0.21
Hibiscus	4.04	7.10	0.31	1.02	0.26	0.46
Lavatera	6.04	16.42	0.89	2.06	0.34	0.83
Bush Honey-suckle	2.77	8.20	0.44	0.96	0.18	0.54
Carrot	3.16	8.45	0.53	1.05	0.23	0.53
Pepper	11.53	12.23	1.30	1.18	0.49	0.44
Petunia	4.28	9.95	0.66	1.16	0.20	0.45
Tobacco	3.30	8.50	0.50	0.98	0.23	0.50
Tomato	4.42	9.86	0.58	1.02	0.28	0.51
Egg Plant	3.65	7.86	0.60	0.80	0.24	0.41
Oak	5.33	4.57	0.62	0.63	0.28	0.24
Thunbergia	5.95	8.05	0.73	1.03	0.31	0.39
Snapdragon	8.80	9.43	1.20	1.28	0.43	0.45
Marigold	6.77	8.60	0.92	1.24	0.15	0.20
Zinnia	5.36	7.56	0.65	0.87	0.31	0.43
Bunflower	10.51	10.62	1.28	1.12	0.55	0.52
Sweet Corn	7.51	10.76	0.86	1.25	0.50	0.74

1. See footnote of Table 2 for plot treatments.

From an examination of the data in the two tables it is at once evident that magnesium deficiency markedly reduces not only the chlorophyll content of those plants which it affects, but also the yellow pigments.

The literature survey indicated that there are few data with which to compare the results presented here. No one, so far as the author is aware, has studied the effects of magnesium deficiency upon the pigments with plants grown in the field. Michael (26), working in Germany on a much broader subject, determined all three pigments of bean leaves from plants grown in sand supplied with nutrient solutions containing none, a trace, and much magnesium. His results agree with those in the present study.

Brown (5) grew Swiss chard in the green house with nutrient solution and reported that those solutions lacking or low in magnesium produced plants low in carotene. On the other hand, Holmes and associates (16, 17) were unable to find any very consistent differences in the carotene content of Boston head lettuce and kale when grown on the same plots used in the present study.

If magnesium hunger lowers the carotene content of most plant leaves, and from the results reported in the preceding tables there can be little doubt but that it does, then magnesium becomes a very important element in vegetable fertilizers because carotene is known to be the precursor to the synthesis of vitamin A in the animal body.

Table 5 was derived from Tables 3 and 4 by subtracting the various pigment values for the different plants growing on the magnesium deficient plots from the corresponding values for the same plants growing on the magnesium treated plots. An example should make it clear how these values were derived. The last line of Table 3 gives the values of 4.68 and 10.45 which are the milligrams of chlorophyll per gram of oven-dry leaf tissue for sweet corn leaves when the plants were grown on the magnesium deficient Plot A and the magnesium treated Plot B. By subtracting the former from the latter the value of 6.67 is obtained which appears in Table 4 for sweet corn under the column headed "unlimed plots." Exactly the same procedure was followed for sweet corn grown on the magnesium deficient Plot D and the magnesium treated Plot C.

Table 5 records the increases in chlorophyll, xanthophyll and carotene contents in the leaves of the various plants which resulted from the magnesium sulfate treatment when applied with and without lime. The limed plots had a pH of nearly neutral, being about 6.5 to 6.8 while the unlimed plots were exceedingly acid, being about 4.2 to 4.4.

In some cases increases in the pigments were greater when the comparisons were made on the limed plots and in others just the reverse was true. There seemed to be little if any trend in this matter. The reasons are probably due to very unfavorable growing conditions in the 1947 season

Table 5. Increase of Leaf Pigments due to Magnesium Sulfate Treatment on Limed and Unlimed Merrimac Sandy Loam

Plant	Chlorophyll		Xanthophyll		Carotene	
	Unlimed Plots	Limed Plots	Unlimed Plots	Limed Plots	Unlimed Plots	Limed Plots
	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.	mg./gm.
Meadow Rue	4.86	----	0.70	----	0.16	----
Raspberry	2.62	3.80	0.23	0.31	0.05	0.20
Lavatera	----	10.38	----	1.17	----	0.49
Bush Honey-suckle	----	5.43	----	0.52	----	0.36
Hibiscus	5.31	3.06	0.96	0.71	0.22	0.20
Mock Orange	2.28	----	0.21	----	0.15	----
Carrot	----	5.29	----	0.52	----	0.30
Petunia	2.40	5.67	0.31	0.50	0.11	0.25
Tobacco	5.03	5.20	0.43	0.48	0.29	0.27
Tomato	10.16	5.42	0.94	0.44	0.43	0.23
Egg Plant	5.77	4.21	0.39	0.20	0.14	0.17
Thunbergia	0.72	2.10	0.09	0.30	0.04	0.08
Snapdragon	2.46	0.63	0.32	0.08	0.15	0.02
Sweet Corn	6.67	3.25	0.64	0.39	0.39	0.24
Marigold	0.00	1.83	0.00	0.32	0.00	0.05
Zinnia	----	2.80	----	0.22	----	0.12

due to lack of sufficient moisture. A gravelly ridge ran diagonally across the North Corn Acre and it was occasionally impossible to obtain leaf samples from plants that had not suffered a great deal from lack of moisture during certain periods of the summer. The writer had planned to take leaf samples for the 1948 season from some of these same plants plus additional ones, but unfortunately for this work the North Corn Acre plots were destroyed in May 1948 after fifty-seven years of history in order to make room for a campus building.

To find if a certain gain in chlorophyll was accompanied by a proportionate gain in xanthophyll or carotene, correlations were made between increases in any two of the three pigments. A coefficient of correlation of 0.848 was found between the various gains of chlorophyll and xanthophyll. The corresponding values for gains in chlorophyll and carotene and xanthophyll and carotene were 0.900 and 0.776, respectively.

It is not possible to compare these correlation coefficients with any published results since apparently no one has made a similar study of the effects of magnesium deficiency. As indicated in the section dealing with literature review, Oserkowsky (28) obtained a coefficient of correlation between chlorophyll and total carotinoid pigments of 0.934 in a study of the effects of iron deficiency on pear leaves. From his results it would seem that iron and magnesium deficiency behave somewhat alike as far as

their effects upon the chloroplast pigments are concerned. It is possible that the deficiency of any elements which affect chlorophyll will also affect the yellow pigments. Such elements would be iron, magnesium, nitrogen, manganese and, possibly, potassium.

Figure 2 shows the results obtained when the gain in chlorophyll content of the various plants, which is given in Table 5, is plotted against the corresponding gain in xanthophyll. It will be noted that in sixteen of the twenty plants investigated the magnesium sulfate treatment caused an increase in leaf pigment. Twenty-five comparisons were available for plotting since some were made on both limed and unlimed plots. Figures 3 and 4 give similar results for chlorophyll and carotene and xanthophyll and carotene.

The question no doubt will arise in the reader's mind as to why there should be a greater correlation between chlorophyll and carotene than between chlorophyll and xanthophyll. The reason, of course, is not known but speculations on possible reasons are interesting.

Carotene is a hydrocarbon of the empirical formula $C_{40}H_{56}$. There are several isomers of carotene, but leaf carotene is evidently almost entirely beta-carotene. On the other hand, xanthophyll is represented in leaves by no less than twelve isomers of the general formula of $C_{40}H_{56}O_2$. This fact in itself might indicate that one could expect more variation among different plants in xanthophyll than in carotene.

Fig. 2. Effect of Magnesium Sulfate on the Amount of Increase of Chlorophyll and Xanthophyll in Plant Leaves

Fig. 3. Effect of Magnesium Sulfate on the Amount of Increase of Chlorophyll and Carotene in Plant Leaves

Fig. 4. Effect of Magnesium Sulfate on the Amount of Increase of Xanthophyll and Carotene in Plant Leaves

Other research workers who have investigated various phases of pigment relationships in plants have laid considerable stress on certain ratios between the various pigments. As mentioned previously, Mackinney (27) found that chlorosis due to iron deficiency caused the ratio of carotene to xanthophyll to be much lower in barley, but that the ratio of chlorophyll to total carotinoids fluctuated together. On the other hand, the data of Oserkowsky (30) showed that the ratio of chlorophyll to total carotinoids was less in chlorotic pear leaves than in non-chlorotic leaves.

Table 6 gives the average for the various pigment ratios of all the plants studied.

Table 6. Effect of Deficiency of Magnesium on the Average Pigment Ratios

Ratios	Magnesium Deficient Plots	Magnesium Treated Plots
<u>Chlorophyll</u> <u>Xanthophyll</u>	8.25	8.56
<u>Chlorophyll</u> <u>Carotene</u>	20.80	19.43
<u>Xanthophyll</u> <u>Carotene</u>	2.58	2.80
<u>Chlorophyll</u> <u>Carotinoids</u>	5.72	5.85

Since the ratios are practically the same whether they are calculated from samples taken from the magnesium deficient plots or from the magnesium treated plots, the only

conclusion that can be drawn is that a deficiency of magnesium does not disturb the proportions of the various pigments one to another. In other words, the ratios are the same in chlorotic and non-chlorotic leaves. In this respect magnesium deficiency differs from iron deficiency.

Results of Analysis for Total Pigments
in Leaves of Tomato Plants Treated with the
Alkali Cations

As noted in the literature survey, several of the recent studies which have been made on magnesium deficiency have shown that potassium apparently represses the intake of magnesium by the plant. Soils which are moderately low in available magnesium can be made deficient simply by supplying an abundance of potassium. Magnesium deficiency symptoms have been induced in tomatoes grown in soil containing an abundance of magnesium by supplying an over-abundance of potassium. Leaf samples of apples, tomatoes, potatoes, currants, gooseberries and tung trees have all shown lower magnesium content and higher potassium content upon analysis (5, 11, 31, 33, 34, 35, 36). Whether potassium represses the absorption of magnesium by the plant or represses its movement into the leaf is not known, as all the analyses to date have been on leaves rather than on the entire plant.

No one has made a study of the influence of other cations appearing in the same group in the periodic table with potassium upon absorption of magnesium by the plant.

With the view in mind of attempting to throw a little

light on this problem of ion absorption as related to magnesium, the following greenhouse experiment was performed. Soil from Plots A and D was brought into the greenhouse and mixed by shovelling back and forth on a cement floor. The purpose behind the idea of mixing the two soils in equal proportion was to obtain a magnesium deficient soil with a favorable soil reaction for plant growth. Soil A had a pH of 4.2 and B of 6.5 when taken from the field. The mixed soil had a pH of 5.6.

The mixed soil was placed in twenty-eight one-gallon pots and given the following treatments in quadruplicate: (1) 75 ppm. Mg, (2) 30 ppm. Li, (3) 300 ppm. Na, (4) 300 ppm. K, (5) 125 ppm. Rb, (6) 125 ppm. Cs, (7) Check - nothing added.

Two tomato plants which had previously been started in the same soil were transplanted into each pot. All pots were given an initial treatment of ammonium nitrate in solution in order not to have a deficiency of nitrogen. Since the soil had been fertilized in the field each year with a mixed fertilizer having a grade approximately equal to a 5-8-7, and since potassium and phosphorus are not so readily leached from the soil as nitrogen, no potassium or phosphorus were given, except, of course, the potassium to the pots in treatment No. 4.

Tomato seeds were planted December 2, 1947, the plants were transplanted into the pots January 2, 1948 and the plants were harvested about the first of March. Thus the

cultural period lasted about three months during which the plants grew very well considering that part of the period was rather unfavorable due to short days.

The total amount of the alkali cations was not given all at once. One-third was supplied the first week after transplanting, one-third the second week, and the remainder the third week. This was done to prevent exceeding the toxic limits of any of the cations used. However, in spite of this precaution, lithium toxicity symptoms developed a few days after the last increment of treatment was added. The edges of the lower leaves died by drying as though this part of the leaf had been placed in a stream of warm, dry air. The dead part remained a bright green, but would crumble easily if touched. This dying process spread from the edges toward the midrib and finally the entire leaf was affected, and it gradually spread from lower leaves to those higher up until it had reached about half way up to the terminal growth by the time the plants were harvested. Moisture determinations showed that the unaffected top leaves were about ten per cent lower in moisture than corresponding leaves on plants in the other treatments. Since the lithium treated plants made only about one-half the growth, as measured by dry weight, as compared to the other plants, and since the lower leaves were destroyed from which samples for pigment analysis are normally taken, they were discarded.

Before the plants from the other treatments were harvested, all the lower leaves were removed from the lower-most

five branches and these served as samples for pigment analysis. None of the lower leaves were chlorotic on those plants to which magnesium was added, but all the others were, and from observation it appeared that those plants receiving potassium and rubidium were more chlorotic than either the check treatment or the sodium and caesium treatments. Only slight amount of chlorosis was noted on sodium treated plants. A visual comparison of the plants may be obtained by examining the colored photographs in the Appendix (see Plates 1, 2 and 3).

In Table 7 are recorded the results of the pigment analyses and the amount of magnesium absorbed. The absorbed magnesium was obtained by multiplying the dry weight of the aerial portion by the amount of magnesium it contained as determined by chemical analysis, using the 8-hydroxyquinolate method. There were no significant differences in the dry weight of the plants due to treatment, nor in the amount of magnesium absorbed, except that the magnesium treated plants absorbed significantly more than those not receiving magnesium.

It is interesting to note that the plants which received sodium were not significantly lower in any of the pigments as compared to those which received magnesium. The amount of magnesium absorbed was greater, but not significantly greater, than treatments involving other alkali cations.

Pigment concentrations in the lower leaves of plants which received potassium, rubidium and caesium were all

Table 7. Effect of Certain Alkali Cations on Pigment Content of the Lower Leaves of Tomatoes and their Influence on Magnesium Absorption when Grown in Magnesium Deficient Soil

Treatments	Chloro- phyll (1)	Xantho- phyll (2)	Carotene (3)	Magnesium Absorbed (4)
	mg./gm.	mg./gm.	mg./gm.	gm.
75 ppm. Mg.	8.64	1.05	0.43	0.1579
300 ppm. Na	8.16	1.03	0.42	0.0469
300 ppm. K	6.55	0.93	0.37	0.0436
125 ppm. Rb	5.80	0.78	0.31	0.0436
125 ppm. Cs	6.64	0.86	0.36	0.0431
Check	6.99	0.88	0.36	0.0430

(1) Differences significant to 1% point; least significant difference is 1.32 milligrams per gram.

(2) Differences significant to 1% point; least significant difference is 0.12 milligrams per gram.

(3) Differences significant to 1% point; least significant difference is 0.05 milligrams per gram.

(4) Differences significant to 1% point; least significant difference is 0.0420 grams.

significantly lower than those which received magnesium and sodium, but not significantly different from the check treatment. It must be concluded, therefore, in the case of these cations under the conditions of this experiment that they had no effect upon pigment concentrations in the lower leaves of the tomato plants, nor on the absorption of magnesium by the aerial portions of the plants. It appears that the differences are due to magnesium deficiency alone

and would have appeared as great had these cations not been added.

In view of the many recent reports to the effect that potassium decreases the absorption of magnesium by plants, it seems strange that the potassium treatment in this experiment did not repress absorption of magnesium by tomatoes and greatly increase chlorosis. There is a possibility that different results would have been obtained had a soil been used that contained at least a moderate amount of magnesium. The soil used in this experiment was extremely deficient in magnesium, as is indicated by the fact that addition of magnesium increased the magnesium content of the tomato plants nearly 300 per cent.

Order of Plant Development in Relation to Degree of Chlorosis

By studying the flowering structures of seed plants botanists have shown that present day plants are related to each other, some having been evolved from others. It is believed that all seed plants have had common ancestors. Seed plants apparently arose during Jurassic times, although there were flowering fern-like forms before that time.

A comparison of fossil remains of early seed plants with present day plants reveals that some of our present plants have not evolved to any great extent, their flowering structures remaining similar to the early forms. On the other hand, many have evolved new and more efficient structures. The latter group are said to have reached a high

level of evolution.

Grouping or arranging the various plant families and orders into so-called "family trees" which are supposed to show in a diagramatic fashion which families have evolved one from another has occupied the time of many botanists. For this type of study the flower is usually used since it is the one structure in which variation among individuals of the same family is small. Leaf and stem structures vary a great deal even among individuals of the same specie. Serum and pollen studies have also been used as a basis for phylogentic classifications. These studies have in general agreed with taxonomic studies.

The actual points used for basis of classification have been fairly well agreed upon by botanists, but the actual classifications as given by different workers naturally differ somewhat. In this study Pools (29) modification of Bessey's arrangement will be used.

As was noted in the survey of literature dealing with the effects of magnesium deficiency on different plants, Eisenmenger and Kucinski (12) have observed that plants belonging to low orders of development are generally more chlorotic than those of higher order. This was interpreted as meaning that the low plants need magnesium more than the high ones.

It might be thought that the difference of chlorophyll concentration in the leaves of two plants of the same specie, one suffering from magnesium deficiency and one supplied with

magnesium, should be a measure of the degree that that plant suffers from magnesium deficiency. This cannot be entirely true because it has been shown that magnesium deficiency affects the rate of photosynthesis and thus the rate of growth long before it causes a reduction in chlorophyll concentration of the foliage. When it is realized that only about ten per cent of the total magnesium in the plant is contained in the chlorophyll it becomes clear that magnesium plays other roles in nutrition and growth besides synthesis of this pigment.

In Table 8 all the plants grown in the North Corn Acre from which leaf samples were obtained are arranged in order from those of low to high development. Percentage gains of chlorophyll in the leaves resulting from the addition of magnesium sulfate to limed and unlimed plots are given.

Table 8. Increase of Leaf Chlorophyll in the Plants When Arranged According to Order of Development

Plant	Family	Gain of Chlorophyll over control on --	
		Unlimed Plots	Limed Plots
		per cent	per cent
Meadow Rue	Ranunculaceae	264.1	---
Raspberry	Rosaceae	47.8	99.0
Apple	Rosaceae	0.0	0.0
Hibiscus	Malvaceae	126.7	75.7
Lavatera	Malvaceae	---	171.9
Bean	Leguminosae	304.4	36.5

Table 8. Increase of Leaf Chlorophyll in the Plants
When Arranged According to Order of Development (Con't.)

Plant	Family	Gain of Chlorophyll over control on --	
		Unlimed Plots per cent	Limed Plots per cent
Mock Orange	Saxifragaceae	36.4	---
Sedum	Crassulaceae	9.8	0.0
*Coral Bell	Saxifragaceae	0.0	0.0
*Pinks	Caryophyllaceae	116.8	108.2
*Maltese Cross	Caryophyllaceae	---	50.7
*Rhubarb	Polygonaceae	11.5	25.7
*Red maple	Aceraceae	27.0	19.5
Carrot	Umbelliferae	---	167.4
Petunia	Solanaceae	32.3	132.5
Tomato	Solanaceae	498.0	122.6
Tobacco	Solanaceae	147.9	157.6
Egg Plant	Solanaceae	233.6	115.3
Pepper	Solanaceae	---	6.1
*Iris	Iridaceae	0.0	0.0
*Bush honey- suckle	Caprifoliaceae	---	42.1
Oak	Fagaceae	0.0	0.0
Thunbergia	Acanthaceae	12.2	35.3
*Veronica	Scrophulariaceae	22.5	0.0
Snapdragon	Scrophulariaceae	36.1	7.2
Sunflower	Compositae	---	1.0
Zinnia	Compositae	---	41.0
Marigolds	Compositae	0.0	27.0
Sweet Corn	Gramineae	142.5	43.2

*Calculated from Table 2.

The percentage gain of chlorophyll for the plants when arranged in ascending order of development shows a great deal of variation, even between comparisons on limed and unlimed plots for the same plant. As stated previously, the 1947 season was unusually dry. Only 8.26 inches of rain was recorded by the Official Weather Station of the Massachusetts Agricultural Experiment Station for the months of July, August and September as compared to the normal of 12.42 inches. Some of the plants on the magnesium deficient limed plot (Plot D) suffered a great deal for lack of moisture. This was particularly true for zinnias, marigolds, and petunias.

For the purpose of comparison of the behavior of the plant families in Table 8 to magnesium deficiency, the low group was considered as being from Ranunculaceae through Leguminosae, the intermediate group from Saxifragaceae through Solonaceae, and all the rest were considered high.

The average gain in leaf chlorophyll for the low group was 112.3 per cent. Apple in this group did not show any chlorosis during the season. Evidently the degree of magnesium deficiency in these plots was not sufficiently great to bring about chlorosis in this plant.

The average gain in leaf chlorophyll for the intermediate group was 91.3 per cent. The Solonaceae family, which was included in this group, while being rather highly developed, behaved more like a lowly developed plant. It was unfortunate that so many representatives of this family were included in the samples taken.

The average gain for the high group was 24.1 per cent. Thus it would appear that as we ascend the scale of evolution for seed plants the degree of chlorosis becomes less and less.

If plants of low development suffer more from magnesium deficiency than those of high development, it would seem reasonable to expect that those of low development would make a greater gain of chlorophyll with the addition of magnesium. The data recorded in Table 8 would seem to indicate that this assumption is correct, although there were a number of exceptions. It seems reasonable to assume that if a very large number of plants of different orders were grown under conditions of magnesium deficiency and magnesium sufficiency, the amount of chlorophyll synthesized would follow a general pattern. With the increase of magnesium the plants of the lower orders would in general show the greatest percentage gain; the intermediate orders, a medium percentage gain; those of the highest orders, the least percentage gain.

It should be emphasized that throughout this report the term "gain of chlorophyll" is used advisedly in the sense that we are dealing with the comparison of individuals of the same species under the two conditions of soil magnesium supply. A plant that has become chlorotic can make no gain of chlorophyll with the addition of magnesium. This is one respect in which chlorosis due to magnesium deficiency differs markedly from chlorosis due to nitrogen, iron, or manganese deficiency. When once the chlorophyll in

a given leaf has been destroyed by lack of magnesium, it cannot be restored by adding soluble magnesium compounds to the roots of the plant or as a spray to the leaf surface. The physiological reason for this is not known.

Since the field plots from which the data in Table 8 was obtained were destroyed, it was impossible to further check any of the plants or to add new ones to the list. In an attempt to gain further information on the relation of magnesium deficiency to order of plant development a greenhouse experiment was performed.

Soil from Plot A was used to fill forty-two three-gallon pots. These were all given a treatment of chemically pure calcium oxide in amount equivalent to a field application of 1300 pounds per acre of limestone. In addition, one-half of the pots were treated with $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ in amount equivalent to a field application of 225 pounds per acre, or 22.2 pounds of magnesium per acre.

Velvet leaf, mustard and string beans, which are respectively members of the Malvaceae, Cruciferae and Leguminosae families, were selected as representatives of the lowly developed group. The intermediate group was represented by buckwheat, a Polygonaceae, and spinach, a Chenopodiaceae. Two Compositae, ragweed and aster, were used for the high group.

The various plants and two soil treatments, one with magnesium and one without, were replicated at random three times. Nitrogen was supplied in the form of ammonium nitrate,

but no additional phosphorus or potassium was added since the field plots had received eighty and seventy pounds per acre of the elements annually. The pots were planted May 13 and the various plants were harvested when they had reached the blooming stage, except in the case of the velvet leaf plants, which were harvested in a pre-blooming stage, since it became evident that the magnesium deficient plants would not reach the blooming stage without losing too many leaves.

It was noted that the magnesium-treated series of buckwheat, beans, mustard and spinach reached the blooming stage before the corresponding magnesium-deficient plants. It is probable that the capacity of magnesium to act as a carrier of phosphorus was responsible for this effect. Hastening of maturity is a well-known function of phosphorus.

Buckwheat reached the blooming stage of growth first, and was closely followed by the string beans and spinach. Before these plants were harvested the entire lot of plants was photographed, using 35 mm. color-sensitive film. Reproductions in color of these pictures may be found in the Appendix (See Plates 4 to 10, inclusive).

Table 9 gives the dry weight of the plants for the two soil treatments and the percentage increase in dry weight which resulted from the use of magnesium sulfate. Velvet leaf and spinach were most severely affected and mustard the least. There was little if any relation between the percentage increase in dry weight, or yield, and order of plant development.

Table 9. Influence of Magnesium Sulfate on Dry Weight of Plants of Different Order of Development Grown in Magnesium-Deficient Merrimac Sandy Loam

Plants	Plants per pot	Dry Weight (1)		Per cent Increase
		No.	Mg.	
		Mg. mg.	Added mg.	
Low Order:				
Velvet Leaf	8	5.91	13.27	124.5
Mustard	10	17.00	17.29	1.7
Beans	4	10.91	13.97	28.0
Intermediate Order:				
Buckwheat	8	19.88	26.83	34.9
Spinach	8	2.29	5.04	120.1
High Order:				
Ragweed	3	21.47	31.15	45.1
Aster	3	10.65	14.70	38.0

(1) Least significant difference between mean dry weights is 2.56 grams.

In Table 10 the chlorophyll content of the leaves of the various plants and the location on the plant at which the samples were taken are recorded. The percentage increase in chlorophyll which resulted from the use of magnesium sulfate is also given.

Evidently the difference in the chlorophyll content of plant leaves is a better indicator of the need of magnesium than the difference in dry weight. Even with chlorophyll many precautions should be taken. The sample must be taken

Table 10. Influence of Magnesium Sulfate on Chlorophyll Content of Leaves of Plants of Different Order of Development Grown in Magnesium-Deficient Merrimac Sandy Loam

Plants	Leaf Position	Chlorophyll (1)		Per cent Increase
		No Mg.	Mg. Added	
		mg./gm.	mg./gm.	
Low Order:				
Velvet Leaf	Top	8.23	15.08	83.2
Mustard	Bottom	3.67	14.14	285.3
Beans	Middle	8.19	13.74	67.8
Intermediate Order:				
Buckwheat	Bottom	4.26	9.95	133.5
Spinach	Middle	13.52	13.14	0.0
High Order:				
Ragweed	Bottom	11.56	12.62	9.1
Aster	Bottom	11.63	10.88	0.0

(1) Least significant difference between the means is 1.11 milligrams per gram of dry weight.

at the correct stage of growth and at the same location on the plant. Also plants which behave normally toward magnesium deficiency should be used in making the comparisons. The term normal behavior means the gradual fading of chlorophyll between the veins on the lower leaves and the spreading of this condition to higher locations on the plant. Spinach is an example of a plant that does not behave normally. In this experiment it was the first one to show

symptoms of deficiency. White necrotic areas developed on the cotyledons and on some of the first true leaves, which soon died. Later leaves appeared normal but the plants made very slow growth. The magnesium-deficient plants had just as much chlorophyll in their leaves as the magnesium-treated ones. Thus in spinach, plant yield would be a better criterion for judging the degree of magnesium deficiency than chlorophyll concentration.

Since magnesium deficiency affects the lower leaves first, it is possible to find on the same plant all gradations from severely chlorotic to normal leaves. It is evident from this fact that location on the plant at which samples of the leaves are taken is of prime importance when different plants are to be compared. In this experiment velvet leaf and bean plants dropped their lower leaves as soon as most of the chlorophyll was gone. In velvet leaves only three or four top leaves remained on the magnesium-deficient plants by the time they were harvested, which was about two weeks after the photographs were taken. Had it been possible to use bottom leaves in these plants the percentage gain of chlorophyll would have been several times that given in Table 10.

In order to determine if the plants of low development, as a group, could be expected to make a greater gain in magnesium from supplying ample amounts to the soil, the records of analyses of plants grown on the North Corn Acre were examined. Table 11 is a selection from some of the data which were available.

Table 11. Magnesium Content of Leaves of Plants Grown
on Magnesium-Deficient and Magnesium-Treated
Plots and Arranged According to
Order of Development (1)

Plant	Year	Family	Plot A	Plot B	Per cent
			No Mg. per cent	Mg. per cent	Increase per cent
Low Order:					
Rose	1946	Rosaceae	0.15	0.20	33.3
Raspberry	1946	"	0.14	0.22	57.1
Apple	1943	"	0.15	0.32	113.3
Peach	1943	"	0.23	0.32	39.1
Kale	1938	Cruciferae	0.12	0.25	108.3
Broccoli	1938	"	0.09	0.28	211.1
Radish	1938	"	0.30	0.53	76.6
Meadow Rue	1946	Ranunculaceae	0.13	0.21	61.5
Mallow	1945	Malvaceae	0.12	0.49	308.3
Elm	1946	Ulmaceae	0.20	0.34	70.0
Larch	1946	Coniferae	0.09	0.13	44.4
Cleome	1945	Capparidaceae	0.22	0.36	63.6
Intermediate Order:					
Buckwheat	1944	Polygonaceae	0.35	0.81	131.4
Dock	1944	"	0.42	0.69	64.3
Globe Amaranth	1944	Amarantaceae	0.26	0.57	119.2
Cockscomb	1944	"	0.59	0.84	42.4
Tobacco	1944	Solanaceae	0.53	0.95	79.2
Potato	1936	"	0.15	0.18	20.0
Blueberry	1946	Ericaceae	0.15	0.17	13.3

Table 11. Magnesium Content of Leaves of Plants Grown
on Magnesium-Deficient and Magnesium-Treated
Plots and Arranged According to
Order of Development (1) - (Cont'd.)

Plant	Year	Family	Plot A	Plot B	Per cent
			No Mg. per cent	Mg. per cent	Increase per cent
High Order:					
Aster	1942	Compositae	.41	0.46	12.2
Cosmos	1944	"	.27	0.26	0.0
Calendula	1944	"	0.23	0.27	17.4
Sunflower	1940	"	0.36	0.49	36.1
Calliopsis	1944	"	0.16	0.13	0.0
Love grass	1944	Gramineae	0.13	0.14	7.7
Sweet Corn	1940	"	0.24	0.28	16.6
Popcorn	1940	"	0.23	0.28	21.7

(1) The data in this table was taken from unpublished analyses on file in the Agronomy Department of the Massachusetts Agricultural Experiment Station.

The average percentage gain in magnesium for the low group was 98.9 per cent. This can be compared to 67.1 per cent for the intermediate group and 14.0 per cent for the high group.

DISCUSSION OF RESULTS

The results of the pigment analysis of chlorotic and non-chlorotic leaves of plants grown in the field on magnesium-deficient soil and treated with 150 pounds of magnesium sulfate per acre annually indicate that chlorophyll, xanthophyll and carotene concentration in the leaves is reduced under conditions of magnesium deficiency.

The extent of loss of the three pigments varies with different plants when grown under the same condition of deficiency. Seed plants of low order of development lose more pigments than do plants of high order of development.

The amounts of chlorophyll, xanthophyll and carotene lost as a result of magnesium deficiency are proportional to one another. This conclusion follows from the high coefficients of correlation obtained between the gains of any two of them as a result of correcting the deficiency. It is further substantiated by the more or less constant ratio of the three pigments in chlorotic and non-chlorotic leaves. It is known that the three pigments, possibly along with others, are located in the chloroplasts of leaves. It would seem from these results that they are not only located together, but are associated by some chemical or physical means. If this is not the case it would be difficult to explain why a deficiency of magnesium could affect xanthophyll and carotene, since neither contain this element as an integral part of their molecules. There is

the possibility that loss of chlorophyll has an indirect effect on the carotinoid pigments and other leaf constituents. Michael (24) advanced the theory that magnesium deficiency upsets the protein-carbohydrate ratio in the leaf and the loss of xanthophyll and carotene was a result of this disturbance. The explanation of the relation between the green and yellow pigments and their behavior to mineral deficiencies must wait until more is known concerning their functions in the economy of the plant.

The lack of a depressing effect of potassium on the intake of magnesium in the experiment dealing with the alkali cations is at variance with the results obtained by others. After the results of the chemical analysis for magnesium were completed the reason for the failure of any influence became clear. This particular soil was so low in magnesium that apparently there was not enough present to be affected. It was unfortunate that time did not permit repeating the experiment with a soil containing more magnesium.

It will be remembered that in this experiment the sodium treatment prevented to a certain degree the loss of pigments as compared with the check treatment. This might have been accomplished by replacing magnesium in some of its functions, exclusive of chlorophyll synthesis, or by aiding in the intake of magnesium to a very small, yet sufficient, degree to increase pigment synthesis above that in the check plants. The sodium-treated plants were slightly

higher in magnesium.

Eisenmenger and Kucinski (12) advanced the theory that primitive plants may have had to deal with more magnesium in proportion to calcium than plants of today. This was based on some results which they cited which showed that magnesium in dolomite dissolves and is removed more rapidly than calcium. Thus it was concluded that the first soils were higher in magnesium than present day soils.

Another theory which may account for the behavior of plants of different order of development with respect to magnesium deficiency, or any factor contributing to adverse growing conditions, is that the highly developed plants are the ones now developing new forms as a result of competition. They are naturally husky plants and are able to survive under adverse conditions. The low plants are now declining and in time will become extinct. They are naturally weak plants.

SUMMARY

1. Plant leaves were collected from thirty different plants grown on four field plots which had been treated for thirteen years as follows: one was left as a check; to a second, 150 pounds per acre of magnesium sulfate was added; to a third, the same amount of magnesium was added and in addition 1000 pounds per acre of non-dolomitic limestone; and to the fourth 1000 pounds per acre of limestone but no magnesium. All four plots received annual applications of a complete fertilizer free of magnesium. The soil is a sandy loam type which belongs to the Merrimac series.

2. Chlorophyll, xanthophyll and carotene contents of the leaf samples were determined. Methods approved by the Association of Official Agricultural Chemists were used for chlorophyll and carotene, and a modification of the latter was made to include xanthophyll.

3. Results indicate that a deficiency of magnesium in the soil causes a loss of chlorophyll, xanthophyll and carotene in the lower leaves of some plants and not in others.

4. Plants which suffered the greatest loss of pigments are those seed plants which belong to low order of development from an evolutionary standpoint. Plants of high order of development either did not become chlorotic or reacted only to a minor extent under the conditions of magnesium deficiency which existed in these plots.

5. The leaves of fifteen out of twenty plants for which chlorophyll, xanthophyll and carotene determinations were made were higher in these pigments when grown on plots treated with magnesium sulfate as compared with check plots.
6. A positive coefficient of correlation of 0.848 was found relating the gain of chlorophyll to the gain of xanthophyll for the fifteen plants for which a total of twenty-five comparisons was made on both unlimed and limed plots. The correlation coefficient for chlorophyll and carotene was higher, being 0.900. That for xanthophyll and carotene was lowest of all, being 0.776.
7. Lime had no consistent effect on the degree of chlorosis suffered by any of the plants tested.
8. Greenhouse experiments indicated that chlorophyll concentration in the lower leaves of plants is a better single criterion for judging the degree to which most plants need magnesium than the dry weight.
9. Rubidium, cesium and potassium failed to influence the pigment concentration in the lower leaves of tomatoes or the intake of magnesium when grown in soil very low in available magnesium. Sodium had a beneficial effect in preventing chlorosis and lithium proved toxic in the concentration used (30 ppm.).

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APPENDIX

Table 1. Common, Family and Scientific Names of Plants Used

Common Name	Family Name	Scientific Name
Apple	Rosaceae	Malus sylvestris
Aster	Compositae	Aster spp.
Bean	Leguminosae	Phaseolus vulgaris
Buckwheat	Polygonaceae	Fagopyrum esculentum
Bush honey-suckle	Caprifoliaceae	Lonicera spp.
Carrot	Umbelliferae	Daucus carota
Coral bell	Saxifragaceae	Huechera sanguinea
Egg plant	Solanaceae	Solanum melongena
Hibiscus	Malvaceae	Hibiscus syriacus
Iris	Iridaceae	Iris spp.
Lavatera	Malvaceae	Lavatera trimestris
Maltese cross	Caryophyllaceae	Lychnis chalcedonica
Marigold	Compositae	Tagetes patula
Meadow rue	Ranunculaceae	Thalictrum aquilegifolium
Mock orange	Saxifragaceae	Philadelphus coronarius
Mustard	Cruciferae	Brassica juncea
Pepper	Solanaceae	Capsicum frutescens
Petunia	Solanaceae	Petunia hybrida
Pinks	Caryophyllaceae	Dianthus spp.
Ragweed	Compositae	Ambrosia artemisiifolia
Raspberry	Rosaceae	Rubus idaeus
Red oak	Fagaceae	Quercus borealis

Table 1. Common, Family and Scientific Names of Plants Used
(con't.)

Common Name	Family Name	Scientific Name
Red maple	Aceraceae	Acer rubrum
Rhubarb	Polygonaceae	Rheum rhaponticum
Sedum	Crassulaceae	Sedum telephium
Spinach	Chenopodiaceae	Spinacia oleracea
Snapdragon	Scrophulariaceae	Antirrhinum majus
Sunflower	Compositae	Helianthus annuus
Sweet corn	Gramineae	Zea mays, var. rugosa
Thunbergia	Acanthaceae	Thunbergia alata
Tobacco	Solonaceae	Nicotianna tobacum
Tomato	Solonaceae	Lycopersicon esculentum
Velvet leaf	Malvaceae	Abutilon Avicennae
Veronica	Scrophulariaceae	Veronica subsessilis
Zinnia	Compositae	Zinnia eligans

Plate 1. Tomato Plants Grown in Magnesium Deficient Soil:
A was treated with 75 ppm. magnesium,
B with 30 ppm. lithium and
C with 300 ppm. sodium

Plate 2. Tomato Plants Grown in Magnesium Deficient Soil:
A was treated with 75 ppm. magnesium,
D with 300 ppm. potassium and
E with 125 ppm. rubidium

Plate 3. Tomato Plants Grown in Magnesium Deficient Soil:
A was treated with 75 ppm. magnesium,
F with 125 ppm. cesium and
G was left untreated

Plate 4. Velvet Leaf Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 5. Buckwheat Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 6. Beans Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 7. Spinach Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 8. Mustard Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 9. Ragweeds Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

Plate 10. Asters Grown in Magnesium Deficient Soil:
A was untreated and B treated
with 11.1 ppm. magnesium

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