WATER-SOLUBLE BORON IN SOIL AND RESPONSE OF ALFALFA AND GARDEN BEETS TO BORON FERTILIZATION

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INTRODUCTION

Boron is an essential element for plant growth. When the availability of boron in the soil is low and other plant nutrients are present in a sufficient quantity, many crops will not produce a maximum yield. When boron deficiency is severe, plant symptoms characterizing boron starvation may be observed in some crops.

Boron deficiencies have been found in many different soils which vary in texture from light sandy soils to acid mucks, but more commonly occur in sandy soils in high rainfall areas.

Crops which make a rapid growth and have a high boron requirement will exhibit deficiency symptoms where slow growing crops or those having a low boron requirement will produce a normal growth. Much information on boron deficiencies in soil has been collected in several parts of the United States but little information is available for Oklahoma.

Over forty different crops have been shown to exhibit boron deficiency symptoms. In many cases response to a borax application has been noted even though no plant deficiency symptoms were apparent. It is suspected that there is a large area in this country on the border line of a boron deficiency which would give responses to small applications of borax. These responses may take the form of increased vigor of the plant, increased yields, better quality crops, improved seed set, and longevity of stand.

A survey of "Water-Soluble Boron in Oklahoma Soils" was made by Byrnes (4), but no data were obtained to show when crops would respond to boron fertilization.

Since soil types should reflect the influence of parent material and soil development on plant nutrient availability, more chemical and plant studies are needed to identify and designate the soil types and the areas where boron fertilization will be needed for maximum production of many crops.

The purpose of this investigation is to analyze soils of known boron deficiencies and to study its relationship to certain types of disease, or failure of plants to make proper growth.

REVIEW OF LITERATURE

Boron occurs in nearly all soils in chemical combination with silica and other elements in a mineral called tourmaline $(Na_6Mg_3Al_2)_3(AlFe)_6(BOH)_4Si_8O_{38}$ (20)*, but due to some chemical, bacterial, or physical action, it may not be available to the plants. Naftel (21) found that boron was not as available in a high-lime soil as in a low-lime soil. He suggested that lime may stimulate bacterial action sufficiently so that there may be competition between plants and bacteria for available boron. Midgley and Dunklee (19) indicate that the fixation of boron in the soil is chemical rather than biological. They believe that the element is taken up by the soil organic matter instead of being fixed as calcium borate. Parks and Shaw (23) believe that boron may be fixed as a calcium aluminum-silicate product of synthesis as a substitution product for aluminum ions. Cook and Millar (8) observed an induced boron deficiency on hill tops and in sandy areas which was produced by leaching. They also found that calcium and magnesium carbonates fixed boron in forms not available to soybeans.

Drake, Sieling, and Scarseth (11) could not obtain any boron fixation with calcium or organic matter, but noted a change in the ratio between calcium and boron. When a boron deficiency occurred, they found a low boron content and a high calcium content in the leaves and stems. Normal plants were grown with the same level of boron but with a lower level of calcium. The ratio of calcium to boron in normal tobacco plants was 1340:1, while boron deficient plants showed a ratio of from 1500:1 to 2100:1. This ratio may vary with different plants. Shive (30) reported similar findings. He suggested the possibility that boron is,

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* Numbers in parentheses refer to references cited on pp. 35-37

in some way, involved in those processes in the plant in which calcium is a direct reactant. A definite correlation was found to exist between the boron content of the tissues and the soluble calcium content of the tissues. Shive concluded, therefore, that the proportional part of the total calcium which was present in the plant in a soluble state, was regulated by the supply of available boron and not by the total calcium in the plant.

White-Stevens (35) observed, on some Long Island plots in New York, that high potash applications increased boron deficiencies in cauliflower. Reeve and Shive (27) reported like findings in their New Jersey tests. They stated also, that the potassium concentration of the substrate has a definite influence on the accumulation of boron in the tissues of the tomato plant. In their tests with 1.0 part per million of boron in the substrate, the increase of boron in the leaves was nearly 91 percent when the potassium concentration was raised from 5 to 200 parts per million. Boron toxicity was also noted to be more severe at high boron levels when the potassium concentration was increased.

It has been noted by some investigators, that boron deficiency is more prevalent during dry years. Burrell (3) has observed that drought increased these deficiencies, probably by decreasing boron availability. This was confirmed when partial control was obtained by irrigation.

According to Shive (30) experimental evidence indicates that boron is an important factor in organic synthesis. He found that plants, grown in boron deficient media, yield strong positive tests for pectins and negative tests for fats. When boron toxicity occurred the results were reversed, while optimum concentrations produced both pectins and fats. Briggs (2) observed that plants grown in boron deficient solutions for six days, showed a decrease in nitrate absorption. The decrease was more pronounced with increasing severity of boron deficiency symptoms. He believes that boron is one of the esential factors in the utilization of nitrogen.

Both Shive (30) and Marsh (17) noted that the boron requirement in the substrate of dicots was higher than that of monocots. Marsh suggested one reason for this condition may be that, in monocots, a higher percentage of accumulated boron remains soluble and that this soluble boron acts directly or indirectly to keep calcium in a soluble, and therefore, an active state.

According to Purvis (25), areas in the United States showing boron-deficiency symptoms include states widely scattered over the country as a whole, but are centered principally along the Atlantic seaboard, around the Great Lakes, and in the Pacific Northwest. These states include Alabama, Connecticut, Florida, Idaho, Indiana, Maine, Maryland, Massachusetts, Michigan, North Carolina, Ohio, Oregon, Rhode Island, South Carolina, Vermont, Virginia, Washington, West Virginia, and Wisconsin.

The yellowing of alfalfa caused by a boron deficiency, has been noted in practically every state east of the Mississippi river. Willis and Piland (38) of North Carolina, in 1937, found that they could control alfalfa yellows by applying 5 to 10 pounds of borax per acre. McLarty, Wilcox and Woodbridge (18) in the same year, in British Columbia, recommended applications of 30 pounds of boric acid per acre. Boric acid has been the specific recommendation of McLarty because of an alkaline soil.

Potato leafhopper yellowing, according to Cowell and Lincoln (7) is difficult to distinguish from boron deficiency yellowing as far as color is concerned. The leaf hopper yellowing may give a streaked appearance in which the veins and mesophyll adjacent to them remain green. The yellowing may also take place at the distal portion of the leaf. In other cases the yellowing may be more uniform and from this standpoint alone cannot be differentiated from the uniform yellowing of boron deficiency.

The distribution of discoloration is one of the most important factors for distinguishing between the two types of yellows. The leaves injured by leafhoppers occur at various heights on a given shoot, while the yellowing or redding caused by boron deficiency is always confined to terminals. There is another type of yellowing associated with common leaf spot caused by Pseudopeziza medicaginis. This organism causes small, circular, dark brown spots to form on the lower leaves of the alfalfa and, if sufficiently severe, causes them to turn yellow and drop.

Recommendations as to rates of application of borax vary from 15 pounds per acre up to 40 pounds per acre. Colwell and Baker (6) found that around 40 pounds of borax gave good results on Idaho soils. In most of the eastern states 15 to 30 pounds of borax per acre seems to give the best results.

Boron deficiency has a definite bearing on the life of some alfalfa fields. In the western states it is not unusual for a field of alfalfa to produce for 8 to 10 years and even longer. This is not the case in most of the eastern states. Hendricks (15) found that alfalfa in Tennessee often seemed to have a hard time surviving the hot, dry periods of late June, July or August in the second or third year. The growth rate was slow, the leaves turned yellow, and summer showers seemed to encourage the growth of crab grass rather than alfalfa. Hutchenson and Cocke (16) found that in the eastern section of Virginia, high yields of alfalfa could be produced by using adapted seed, lime, and rather heavy applications of fertilizer. However a stand only persisted for short periods, usually being reduced to such an extent that low yields were obtained by the end of the second year. Borax was applied and its absence was found to be a limiting factor in growth and stand of alfalfa in this area. Applications as low as 10 pounds per acre corrected the yellowing and produced profitable stands for longer periods than previously experienced. In tests at Chatham, Virginia, by Grizzard and Matthews (13) it was found that borax treated areas gave a vigorous growth of alfalfa and good stands were maintained, while untreated areas were deteriorating and would appear to be too thin for profitable production during the third year of growth.

The physiological effect caused by a boron deficiency in the production of table beets is known by different names in different sections of the country. In Wisconsin (33) the term that describes the disease is "internal black spot". New York's authorities (26) classify it as "internal breakdown". Oregon (24) refers to it as "canker". In one area the external symptom may be pronounced, while in another area the internal symptom may be more noticeable.

Internal symptoms, according to Walker (32), take the form of black spots, irregular in size, shape and location, but uniform in the fact that they are black in color, and hard or corky in texture.

The external symptoms, as stated by Powers (24), appear as a dark spot on the root, usually on the part of the greatest circumference. Some roots may be very slightly affected with but one small spot of one-half to one inch in size. Other roots may have several spots which cause most of the root to be blackened. As affected roots increase in size, the black spots frequently develop into growth cracks and large open cankers, extending in extreme cases to a complete girdle of the root.

Leaf symptoms are often the best means of detecting a boron deficiency of beets in the field. A fairly close correlation occurs according to Walker, Jolivette, and McLean (34), between the top symptoms and internal black spot. The youngest leaves are distorted, often one-sided and are commonly longer and narrower than normal leaves. The affected leaves die early and drop off, while new leaves develop in a similar manner and finally die. In extreme cases the result is a rosette appearance with the tips of the small leaves dying back. Cook and Millar (9) found that in Michigan a color difference in foliage often appeared. The leaves of beets suffering from boron starvation may have a deeper red color. The color difference was noticeable when beets on borax-treated soils were compared with fields not treated with borax. According to Walker, Jolivette, and McLean (34), table beets are much more susceptible to boron deficiency than sugar beets. These investigators (33) suggest that, for Wisconsin conditions, about 40 pounds of borax per acre is a reasonable rate of application for beets, although in some cases 40 to 60 pounds was necessary to reduce a blackening of the roots to a minimum.

EXPERIMENTAL STUDIES

Source of Soils and Their Analyses

The soils used in this experiment were obtained from Craig County in North Eastern Oklahoma.

Byrnes (4) showed in his analysis of these soils that some were extremely low in water-soluble boron. This was a valuable aid in setting up the experiment for testing the effects of boron on boron deficient soils.

It was thought that there might be a substantial difference in the boron content of Parsons silt loam and Bates silt loam due to the clay pan which exists in the Parsons silt loam. Analysis of several samples of each soil type disaproved this idea. It seems, therefore, that the lack of boron in these soils is not a climatic factor but is due to the lack of tourmaline, a boron bearing mineral in the parent material on which these soils have developed.

The amount of boron extracted from a soil by boiling 20 grams of soil with 40 ml of water for a period of 5 minutes appears to correlate fairly well with crop responses to boron fertilization (1). Soils are classed as boron deficient when they contain less than 0.35 ppm of water-soluble boron (28). Table 1 contains information on the boron content of the various soils used in this experiment. Data from previous studies would indicate that the boron content of the soils used in this experiment are much too low for proper plant growth.

The soils used in this experiment were also analyzed for total nitrogen, easily soluble phosphorus, exchangeable potassium, and pH. The total nitrogen was determined by the Kjeldahl method (31), easily soluble phosphorus was determined by the Acetic Acid method (14),

Table 1. The Boron Content of Soils Collected From Craig County in North Eastern Oklahoma for Use in Pot Experiments.

Sample No.	Legal Description	Land Use	Surface and Subsoil	Soil Type	pH Value	ppm Boron *
1	NE$\frac{1}{4}$,NE$\frac{1}{4}$	Native Pasture	Surface	Bates silt loam	5.5	0.01
	34-26-21		Sub.		6.1	0.01
2	SE$\frac{1}{4}$,SE$\frac{1}{4}$,SW$\frac{1}{4}$	Native Meadow	Surface	Parsons silt loam	5.3	0.17
	30-29-21		Sub.		6.0	0.02
3	NE$\frac{1}{4}$,SE$\frac{1}{4}$	Cultivated	Surface	Hanceville fine	6.1	0.10
	3-28-21		Sub.	sandy loam	6.2	0.05
4	NW$\frac{1}{4}$,SW$\frac{1}{4}$	Cultivated	Surface	Bates silt loam	6.3	0.21
	16-26-20		Sub.		6.4	0.03
5	NW$\frac{1}{4}$,NW$\frac{1}{4}$,NE$\frac{1}{4}$	Cultivated	Surface	Bates silt loam	5.6	0.13
	31-29-21		Sub.		6.0	0.11
6	SW$\frac{1}{4}$,SE$\frac{1}{4}$	Native Pasture	Surface	Parsons silt loam	5.4	0.19
	26-28-20		Sub.		5.9	0.05
7	SE$\frac{1}{4}$,SE$\frac{1}{4}$,NE$\frac{1}{4}$	Native Pasture	Surface	Parsons silt loam	5.6	0.13
	34-26-20		Sub.		5.9	0.08
8	SE$\frac{1}{4}$,SE$\frac{1}{4}$,NE$\frac{1}{4}$	Native Pasture	Surface	Summit Clay	6.3	0.36
	16-25-21		Sub.		6.3	0.76

* Soluble in Water

exchangeable potassium by the Perkin Elmer flame photometer and pH by using the Line Beckman pH meter. The results are shown in table 2.

Table 2. The Chemical Analyses of Soils Collected From Craig County in North Eastern Oklahoma for use in the Pot Experiments

Soil Type	Total Nitrogen percent	Easily soluble phosphorus in lbs. per acre**	Exchangeable potassium in lbs. per acre	pH
Bates silt loam	0.16	0	49	5.5
Parsons silt loam	0.10	0	49	5.3
Hanceville f.s.l.*	0.05	1	43	6.1
Bates silt loam	0.14	0	43	6.3
Bates silt loam	0.13	0	46	5.6
Parsons silt loam	0.15	0	43	5.4
Parsons silt loam	0.18	4	43	5.6
Summit clay	0.33	2	134	6.3

* fine sandy loam ** soluble in N/10 acetic acid

These soils were extremely low in plant nutrients. Consequently fertilizers were applied as follows: P_2O_5 and K_2O, in the form of potassium monohydrogen phosphate, and magnesium sulfate at the rate of 100 pounds per acre. Ammonium nitrate was added only to the pots in which the beets were planted at the rate of 100 pounds per acre. Lime was added at the rate of one ton per acre to the Hanceville fine sandy loam, two tons per acre to the Summit clay and the Bates silt loam, each having a pH of 6.3, and three tons per acre to the remaining pots. The alfalfa seed was inoculated with legume culture "A" before planting.

Method for Determining Water-Soluble Boron in Soil

Boron-free glassware must be used in the determination of water-soluble boron in soil. A procedure described by Naftel (22) was studied and reproducible results were obtained.

Standard boric acid solutions with a range of 0.01 to 0.13 ppm* were prepared. The amount of boric acid needed to make a solution containing 1.0 ppm of boron was so small that it would be difficult to weigh; consequently, a solution containing 100 ppm of boron was made up by weighing out 0.5716 grams of boric acid and dissolving in one liter of distilled water. Ten ml** of this solution was diluted in one liter. This solution contains 1.0 ppm of boron.

To prepare a standard containing 0.1 ppm of boron, take 10 ml of the 1.0 ppm solution and dilute to a 100 ml volume with boron-free distilled water. For a standard containing 0.2 ppm take 20 ml of the solution containing 1.0 ppm and dilute to a 100 ml volume, etc.

A 5 ml aliquot of each standard is placed in a porcelain evaporating dish, made alkaline with 5 ml of a 0.10 N calcium hydroxide suspension, and then evaporated to dryness on a steam both. Remove the dish and allow to cool to room temperature then add 1 ml of a saturated solution of oxalic acid containing 20 percent concentrated hydrochloric acid and 2 ml of a 1.0 percent solution of turmeric dissolved in 95 percent ethyl alcohol. Rotate the dish so that the reagents come in contact with all residue, and evaporate to dryness over a water bath at a temperature of 55°C with no deviation greater than plus or minus 3°C.

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* ppm is parts per million
** ml is 1 cubic milliliter

On reaching the drying point, a reaction takes place between the oxalic acid, tumeric (5)*, and the boric acid forming a red dye called rosocyanine. The intensity of the color produced is directly proportional to the amount of boron present.

When the drying point is reached, continue heating for 30 minutes, remove and allow to cool to room temperature. The red colored residue is taken up in 95 percent ethyl alcohol, transferred to a centrifuge tube and centrifuged to obtain a clear solution. It is then made up to a suitable volume with ethyl alcohol (50 ml was used in this study) and the color intensity determined with a Fisher electrophotometer, with a setting at "B" using a green filter, number 525 B. These results were plotted on graph paper against the amounts of boron that were known to be present and a table was prepared from the curve which could be used to obtain the amount of boron in a soil sample. Figure 1 and table 3 contain information on the relation between the photometer readings and the boron content of the standard solutions.

The soil samples were treated in the same manner as the standards. They were prepared as follows: Twenty grams of air dry soil that has been pulverized to pass through a 60 mesh sieve was weighed out and placed in a 200 ml boron-free Erlenmeyer flask, 40 ml of distilled water was added and the flask connected to a reflux condenser. The soil suspension was boiled for 5 minutes, cooled, and transferred to a centrifuge tube and centrifuged to obtain a clear solution. A 20 ml aliquot of the soil extract was taken in this study, and from here on the sample received the same treatment as priviously described for the standard solutions.

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* $CH_2(CO\text{-}CH\text{-}C_6H_3(OH)OCH_3)_2$ is tumeric.

A 20 ml aliquot is equivalent to 10 grams of soil. When this solution is taken to dryness and then diluted to 50 ml with ethyl alcohol, a one to five dilution is obtained and the data obtained from the curve shown in figure 1 must be multiplied by 5 to obtain the amount of boron present in a soil.

Figure 1. Curve Showing the Relation Between Electrophotometer Readings and Quantity of Boron in Solution.

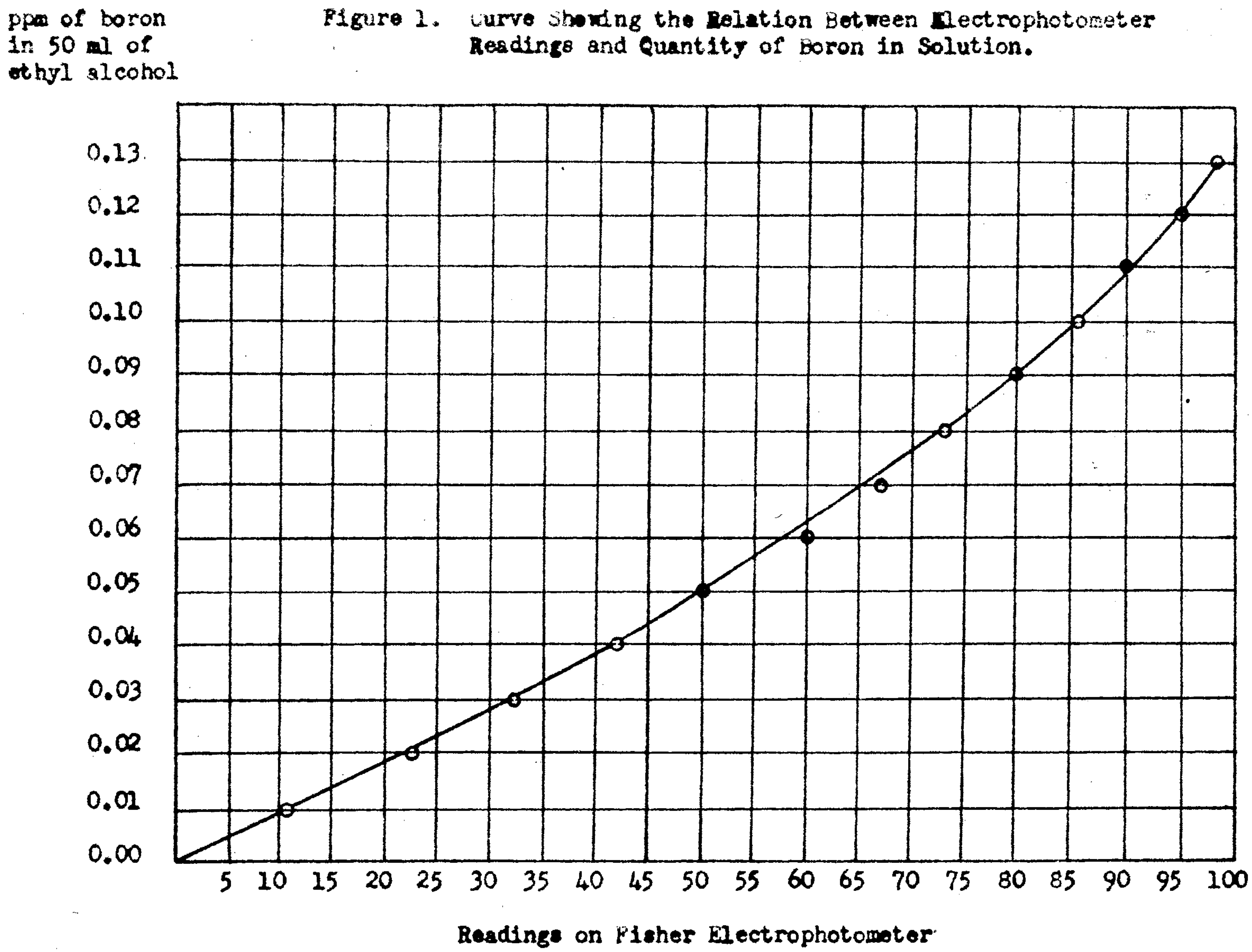

Table 3. Photometer Readings and Approximate Amounts of Boron as Prepared from the Calibration Curve Shown in Fig. 1.

Reading on Photometer	ppm.* Boron	Reading on Photometer	ppm. Boron
2.4	.01	65.5	.34
4.5	.02	67.0	.35
6.5	.03	68.5	.36
8.5	.04	70.0	.37
10.5	.05	71.5	.38
13.5	.06	73.0	.39
15.0	.07	74.0	.40
17.0	.08	75.5	.41
19.0	.09	76.5	.42
21.0	.10	78.0	.43
23.0	.11	79.0	.44
25.0	.12	80.0	.45
27.0	.13	81.5	.46
29.0	.14	82.5	.47
31.0	.15	83.5	.48
33.0	.16	84.5	.49
35.0	.17	85.5	.50
37.0	.18	86.5	.51
39.0	.19	87.5	.52
41.0	.20	88.5	.53
43.0	.21	89.0	.54
44.5	.22	90.0	.55
46.5	.23	91.0	.56
48.5	.24	92.0	.57
50.0	.25	93.0	.58
52.0	.26	94.0	.59
54.0	.27	94.5	.60
55.5	.28	95.5	.61
57.5	.29	96.0	.62
59.0	.30	97.0	.63
60.5	.31	97.5	.64
62.0	.32	98.0	.65
64.0	.33		

* ppm of boron in the above table are calculated for 10 grams of soil extracted with 20 ml of water.

Greenhouse Procedure

All greenhouse tests were conducted in two gallon, glazed earthenware pots. Field soils were collected, screened and placed in the pots in weighed amounts. One hundred pounds of P_2O_5, K_2O, and $MgSO_4$, derived from potassium monohydrogen phosphate and magnesium sulfate, was added to each pot. Lime was added at the rate necessary to bring the pH to neutrality. One hundred pounds of nitrogen, derived from ammonium nitrate, was added to the pots containing the beets. Borax was then added, in a solution, at the rate of 1 ppm to three of the six pots of each soil type. The moisture content of each soil was adjusted to one-third the water holding capacity, and the seed planted. Fifty plants of alfalfa were grown in each pot, and the beets were thinned to three plants per pot. All treatments were duplicated, except in the case of beets. A constant moisture content was maintained in the soil by weighing the pots at frequent intervals.

Three clippings, at monthly intervals, were made on the alfalfa and the beets were pulled at maturity. Chemical analyses were made separately on the leaves and the stems of the alfalfa and on the roots and the leaves of the beets.

Microtitration Method for Determining Total Boron in Plant Material

The method used to determine boron in plant material was a modification of the method recommended by Cook and Millar (10) for soil. The procedure is as follows: Weigh one gram of oven dried plant material into a porcelain evaporating dish or a platinum crucible and dry ash. Add 5 ml of methyl alcohol to the dried residue and rub loose as much of the residue as possible with a glass rod and rubber policeman. Transfer immediately to a soft glass test tube and take up the remaining residue with 7 ml of a 1:1 mixture of H_2SO_4 and methyl alcohol, transfer to the same test tube, then add 5 ml of methyl alcohol. Add 5 ml of distilled water and 5 drops of 2 N Na_2CO_3 to the test tube that will receive the distillate. (It is essential that the methyl borate be distilled into some solution which will render the boron non-volatile. Methyl borate is hydrolyzed to non-volatile sodium borate in a dilute solution of sodium carbonate). Place the test tube containing the plant ash into a water bath and bring the water slowly to boiling. Caution must be exercised in bringing the water to the boiling point. If this is done too rapidly, frothing may occur and some of the acid may pass into the receiving tube. When the water begins to boil and the methyl alcohol has ceased to distill from the tube containing the plant ash, distillation should be started from a third test tube, containing methyl alcohol, which is connected to the tube containing the plant ash. Continue the distillation until the volume of distillate in the receiving tube is approximately 50 ml. Most of the boron is distilled over as methyl borate in the first 25 ml.

The principle involved in the titration procedure for boron has been described by Foote (12) and applied electrometrically by Wilcox (36).

The procedure outlined below using the hydrogen ion meter is patterned with some modification after a method described by Wilcox and Hatcher (37). Boric acid, with an ionization constant of 5.5×10^{-10}, can not be titrated directly with a standard solution of sodium hydroxide, for no indicator is known that will give a sufficiently sharp color change at the neutralization point, which occurs at approximately pH 11. However, the addition of certain polyhydroxy organic compounds such as mannitol, glycerol, or dextrose, form complex acids with the boric acid which are much stronger than the boric acid alone. Foote (12) found that the quantity of alkali required to titrate the boric acid-mannitol complex back to the initial pH of the boric acid was an accurate measure of the boron present.

Apparatus

1. Beckman line-operated pH meter.
2. Microburette, capacity 5 ml, graduated in 0.01 ml.
3. Electric stirrer.
4. Boron-free beakers, 600 ml capacity.

Reagents

1. Bromthymol blue indicator solution; 1%.
2. Sulfuric acid solution; approximately 1 N.
3. Sulfuric acid solution; approximately 0.02 N.
4. Sodium hydroxide solution; carbonate-free; approximately 0.5 N.
5. Sodium hydroxide solution; carbonate-free; 0.0231 N, 1 ml = 0.25 mg boron.
6. Boric acid solution; 1 ml = 0.1 mg boron.
7. Mannitol, neutral. The blank titration for 5 grams of mannitol should not exceed 0.1 ml of the 0.0231 N NaOH.

Titration procedure for boron

Dilute the 50 ml of methyl alcohol containing the sodium borate to 250 ml with distilled water and place in a 600 ml boron-free beaker. Add 1 drop of bromthymol blue indicator and acidify with 1 N H_2SO_4 then add about 0.5 ml in excess. Bring to a boil, stir cautiously, then vigorously for one minute to expel carbon dioxide. Cover the sample with a watch glass and cool to room temperature in a water bath. Introduce the electrodes and the stirrer into the beaker containing the sample. Start the stirrer and add carbonate-free 0.5 N NaOH to approximate neutrality as shown by the bromthymol blue. Adjust sample to a pH of 7.0 with 0.0231 N NaOH or 0.02 N H_2SO_4 if necessary. The indicator needle of the hydrogen ion meter should be steady and should not drift from the reading of 7.0. At this point the microburette reading of the 0.0231 N NaOH should be recorded. Add approximately 5 grams of mannitol. If boron in the form of boric acid is present, the pH will drop to some value below 7.0. Titrate the sample back to the initial pH of 7.0 with standard base using caution near the end-point to permit a slight lag in the hydrogen ion meter. When the needle indicator remains steadily at pH 7.0 record the volume of standard base used to the nearest 0.01 ml. A blank correction for the reagents is subtracted from the total volume of standard 0.0231 N NaOH used.

Computations

If the standard base is exactly 0.0231 N, 1 ml is equivalent to 0.25 mg of boron.

$$\text{ppm B.} = \frac{1{,}000}{\text{wt. of sample}} \times (\text{ml NaOH} - \text{blank}) \times 0.25$$

Results of Present Work

Eight soils were included in this study. The purpose of the experiment was to study the growth of alfalfa and garden beets on soils of varying boron content when liberally fertilized with phosphorus, potassium, and magnesium and the acidity corrected by liming. Two weeks after emergence, the alfalfa was thinned to 50 plants per pot. No signs of boron deficiency or toxicity appeared on the three crops of alfalfa when boron was added to these soils. Plants which grew on soils not treated with boron were less vigorous than those on the treated pots. Alfalfa on some of the soils showed definite signs of boron deficiency symptoms after the second cutting. The beets showed boron deficiency symptoms on only one soil but the yield on most soils not receiving boron were less than on those soils receiving boron as shown in tables 4, 5, and 6.

It will be noted that there was an increase in the yield of alfalfa on all soils treated with boron but this increase was not consistant for the beets.

The Summit clay produced the greatest increase in the yield of alfalfa. This is attributed partially to the high exchangeable potassium and total nitrogen originally present in this soil.

The differences in yield might have been more pronounced if the experiment had been conducted with a less favorable moisture content in the soil. It has been noted by some investigators, that boron deficiency is more prevalent during dry years. Drought increases these deficiencies, probably by decreasing the solubility of boron in the soil solution.

Table 4. Yields of alfalfa from different soils used in greenhouse studies where boron was the limiting factor.

Oven-dry weights in grams. 50 plants per pot.

Soil Type	Crop 1				Crop 2				Crop 3			
	BORON		NO BORON		BORON		NO BORON		BORON		NO BORON	
	leaf	stem	leaf	stem	leaf	stem	leaf	stem	leaf	stem	leaf	stem
Bates silt loam	8.50	6.15	8.23	6.04	7.75	7.29	7.50	6.01	7.64	7.15	7.02	6.94
Parsons silt loam	10.34	8.12	10.32	7.36	8.78	8.17	8.45	8.10	8.67	8.05	7.89	7.43
Hanceville F.S.L.	8.91	6.59	8.64	6.86	6.90	6.01	7.03	6.65	6.94	6.14	6.25	5.98
Bates silt loam	9.55	7.38	8.94	7.01	5.05	4.21	6.01	6.00	8.34	7.81	7.53	7.02
Bates silt loam	8.46	6.93	8.36	6.56	5.48	5.00	6.00	6.23	6.32	6.13	5.73	5.48
Parsons silt loam	9.48	7.33	9.62	7.51	8.40	7.42	8.21	7.69	8.43	7.89	7.59	7.38
Parsons silt loam	10.27	8.17	9.31	7.10	8.35	7.30	8.09	7.63	8.29	7.86	7.52	7.03
Summit Clay	11.62	9.65	11.03	7.68	8.52	6.88	7.47	6.66	8.91	8.05	7.99	7.67

Table 5. Summary of alfalfa yields given in table 4.

Total alfalfa yield in grams

Soil Type	BORON		NO BORON		Increase due to Boron	
	leaf	stem	leaf	stem	leaf	stem
Bates silt loam	23.89	20.59	22.75	18.99	1.14	3.76
Parsons silt loam	27.79	24.34	26.66	22.89	1.13	3.77
Hanceville F.S.L.	22.75	18.74	21.92	19.49	0.83	2.43
Bates silt loam	22.94	19.40	21.48	20.03	1.46	1.45
Bates silt loam	20.26	18.06	20.09	18.27	0.17	1.82
Parsons silt loam	26.31	22.64	25.42	22.58	0.89	0.06
Parsons silt loam	26.91	23.33	24.92	21.76	1.99	1.57
Summit Clay	29.05	24.58	26.49	22.01	2.56	2.57

Table 6. Yields of garden beets on different soils in greenhouse studies where boron was the limiting factor.
Oven-dry weight in grams.

Soil type	Boron leaf	Boron root	No Boron leaf	No Boron root
Bates silt loam	12.904	17.723	12.897	2.123
Parsons silt loam	14.623	18.803	8.843	17.783
Hanceville F.S.L.	10.352	15.702	8.011	8.979
Bates silt loam	10.005	17.718	10.553	12.707
Bates silt loam	10.170	15.511	9.852	19.273
Parsons silt loam	11.830	22.614	13.156	13.733
Parsons silt loam	11.571	14.351	11.453	18.989
Summit Clay	11.362	18.615	10.489	22.252

Total nitrogen, phosphorus, potassium, calcium, and boron was determined on the leaves and stems of the alfalfa and on the leaves and roots of the beets. The nitrogen was determined by the Kjeldahl method (31), phosphorus by the Fisher electrophotometer (29), potassium and calcium on the Beckman flame photometer and boron by microtitration (10, 12, 33, 34). The results are shown in table 7 and 8.

Nitrogen was considerably higher in the alfalfa leaf material than in the stems. Phosphorus and calcium was slightly higher in the leaves than in the stems but potassium was found to be higher in the alfalfa stems. Boron was found to be consistantly higher in the alfalfa leaves.

It will be noted that the boron content of the alfalfa decreased successively through the three cuttings, while the other nutrients remained relatively constant.

Nitrogen, potassium, calcium and boron was considerably higher in the beet leaves than in the roots but phosphorus remained equally distributed in the leaves and roots.

It will also be noted that the beets were capable of absorbing a large amount of the boron that was applied. This indicates that applications of boron should be included in the fertilization of beets each year if they are expected to make proper growth on boron deficient soils.

Table 7. Percentages of nitrogen, phosphorus, potassium, calcium, and boron found in alfalfa leaves and stems obtained from greenhouse experiment.

	Crop 1					Crop 2					Crop 3				
No.	%N	%P	%K	%Ca	%B	%N	%P	%K	%Ca	%B	%N	%P	%K	%Ca	%B
Alfalfa Leaf Material															
1	4.67	0.300	2.13	2.65	0.00575	5.10	0.306	2.24	2.67	0.00425	4.83	0.302	2.03	2.54	0.00375
2	4.77	0.316	2.47	1.97	0.00850	5.06	0.334	2.56	2.08	0.00550	4.79	0.325	2.50	2.01	0.00415
3	4.86	0.309	2.91	1.44	0.00900	5.04	0.350	3.02	1.72	0.00850	4.87	0.321	2.90	1.52	0.00620
4	4.58	0.334	2.36	1.66	0.00800	5.63	0.373	2.54	1.74	0.00600	4.73	0.342	2.37	1.68	0.00515
5	4.56	0.319	2.78	2.44	0.00725	4.98	0.391	2.89	2.52	0.00575	4.43	0.326	2.65	2.39	0.00480
6	4.93	0.309	1.91	1.97	0.00850	4.85	0.327	1.98	2.02	0.00625	4.63	0.312	1.82	1.93	0.00515
7	4.87	0.366	2.25	2.00	0.00800	5.31	0.397	2.38	2.18	0.00575	4.80	0.356	2.19	1.98	0.00550
8	4.92	0.327	2.94	2.09	0.00675	5.00	0.344	2.96	2.13	0.00800	4.91	0.332	2.89	2.01	0.00675
A	4.82	0.352	2.44	2.56	0.00200	4.65	0.321	2.34	2.45	0.00150	4.63	0.305	2.19	2.43	0.00125
B	4.95	0.351	2.47	2.25	0.00450	4.95	0.332	2.31	2.15	0.00250	4.83	0.321	2.25	2.03	0.00215
C	4.75	0.359	3.06	2.00	0.00250	5.06	0.327	2.83	1.89	0.00325	4.79	0.319	2.79	1.86	0.00275
D	4.85	0.352	2.19	1.88	0.00425	5.13	0.344	2.08	1.78	0.00350	4.92	0.338	1.98	1.73	0.00300
E	5.08	0.312	2.50	2.25	0.00500	5.12	0.325	2.65	2.31	0.00250	5.03	0.311	2.43	2.16	0.00225
F	4.89	0.311	2.00	1.88	0.00275	5.17	0.344	2.09	1.96	0.00300	4.97	0.333	1.98	1.74	0.00200
G	4.74	0.342	2.06	2.09	0.00425	5.12	0.394	2.14	2.14	0.00300	5.01	0.351	1.98	1.89	0.00275
H	4.41	0.284	2.75	1.97	0.00575	5.28	0.325	2.83	2.07	0.00450	4.94	0.302	1.98	1.87	0.00425
Alfalfa Stem Material															
1	2.50	0.260	2.79	1.63	0.00200	2.00	0.272	2.81	1.60	0.00100	1.86	0.253	2.72	1.51	0.00085
2	2.39	0.340	3.38	1.33	0.00325	1.95	0.325	2.98	1.26	0.00225	1.78	0.297	2.78	1.19	0.00200
3	2.29	0.338	3.88	0.92	0.00325	2.04	0.319	3.76	1.03	0.00300	1.98	0.298	3.50	0.89	0.00275
4	2.36	0.307	2.96	0.92	0.00400	2.26	0.310	3.01	1.05	0.00275	2.10	0.301	2.87	0.91	0.00225
5	2.15	0.253	3.29	1.21	0.00575	2.20	0.270	3.36	1.23	0.00250	2.09	0.251	3.17	1.11	0.00175
6	2.37	0.353	2.29	1.16	0.00300	2.33	0.375	2.46	1.23	0.00350	2.28	0.364	2.31	1.20	0.00275
7	2.56	0.401	3.04	0.83	0.00325	2.36	0.381	2.98	0.80	0.00325	2.31	0.372	2.87	0.77	0.00300
8	2.18	0.260	3.54	1.08	0.00450	1.93	0.265	3.46	0.76	0.00300	1.87	0.251	3.29	0.78	0.00300
A	2.64	0.310	3.12	1.38	0.00100	2.23	0.306	2.98	1.31	0.00075	1.98	0.299	2.97	1.10	0.00050
B	2.45	0.333	3.21	1.04	0.00300	2.13	0.272	3.01	0.92	0.00115	1.98	0.264	2.89	0.87	0.00075
C	2.34	0.312	3.79	1.17	0.00125	2.13	0.334	3.87	1.21	0.00100	2.01	0.311	3.65	1.09	0.00075
D	2.33	0.292	3.33	0.96	0.00225	2.41	0.305	3.43	1.01	0.00150	2.27	0.294	3.27	0.98	0.00100
E	2.25	0.275	3.38	1.21	0.00275	2.20	0.270	3.34	1.12	0.00205	2.12	0.261	3.22	1.02	0.00150
F	2.25	0.275	2.79	1.00	0.00300	2.27	0.319	2.89	1.03	0.00265	2.23	0.271	2.77	0.96	0.00225
G	2.50	0.322	2.79	0.96	0.00275	2.27	0.363	2.61	0.92	0.00245	2.21	0.333	2.83	0.89	0.00225
H	2.32	0.242	3.54	1.17	0.00325	2.19	0.230	3.65	1.03	0.00305	2.10	0.227	3.66	0.99	0.00275

The numbered samples indicate the soils which received boron. The lettered samples did not receive boron.
1. Bates silt loam 2. Parsons silt loam 3. Hanceville f.s.l. 4. Bates silt loam 5. Bates silt loam 6. Parsons silt loam 7. Parsons silt loam 8. Summit Clay.

Table 8. Percentages of nitrogen, phosphorus, potassium, calcium, and boron in beet leaves and roots obtained from greenhouse experiment.

Soil	With Boron					Without Boron				
No.*	%N	%P	%K	%Ca	%B	%N	%P	%K	%Ca	%B
Beet Leaves										
1.	2.28	0.131	1.05	0.530	0.00500	3.20	0.145	1.23	0.410	0.00150
2.	2.27	0.145	1.23	0.410	0.00450	2.93	0.250	1.23	0.450	0.00200
3.	1.92	0.173	1.45	0.283	0.01000	1.56	0.219	1.53	0.400	0.00225
4.	1.73	0.195	1.18	0.331	0.00950	1.54	0.165	1.09	0.376	0.00500
5.	1.38	0.156	1.39	0.488	0.00950	1.46	0.156	1.25	0.450	0.00475
6.	1.92	0.169	0.98	0.396	0.01225	1.74	0.147	1.00	0.376	0.00450
7.	2.13	0.145	1.12	0.400	0.01050	1.86	0.164	1.03	0.402	0.00400
8.	1.62	0.117	1.47	0.402	0.00625	1.58	0.136	1.37	0.398	0.00525
Beet Roots										
1.	1.45	0.138	0.692	0.042	0.00350	1.51	0.167	0.780	0.034	0.00125
2.	1.71	0.160	0.841	0.033	0.00425	1.79	0.188	0.821	0.026	0.00150
3.	1.00	0.173	0.970	0.021	0.00725	1.30	0.222	0.951	0.029	0.00200
4.	1.35	0.200	0.741	0.021	0.00600	1.03	0.184	0.863	0.022	0.00200
5.	1.41	0.205	0.820	0.033	0.00575	1.05	0.175	0.844	0.040	0.00250
6.	1.43	0.191	0.572	0.020	0.00775	1.63	0.219	0.687	0.025	0.00225
7.	1.43	0.165	0.763	0.020	0.00700	1.42	0.216	0.687	0.024	0.00200
8.	1.44	0.127	0.885	0.027	0.00500	1.44	0.184	0.898	0.029	0.00225

*1. Bates silt loam 2. Parsons silt loam 3. Hanceville f.s.l. 5. Bates silt loam 6. Parsons silt loam 7. Parsons silt loam 8. Summit clay.

The variations in the relative content of calcium and boron in alfalfa and garden beets as a result of treatments with and without boron are wide, as shown in tables 9 and 10. The variations are most evident when expressed as a ratio between the calcium and boron. This ratio is narrower where boron was applied to the soil.

Since there was no symptoms of boron deficiency on the alfalfa produced on soils treated with boron, the conclusion is reached that the calcium-boron ratio of 679 in the alfalfa leaves and 1,780 in the alfalfa stems represents a range in which no deficiencies will occur. On soils not treated with boron, deficiencies were observed in the alfalfa after the second cutting. The calcium-boron ratio in the leaves was 1,940 and in the stems the ratio was 2,200. This ratio is considerably higher than that in the healthy alfalfa.

It has been found by other investigators that boron deficiencies are more likely to develop in limed soils than in the same soil type that has not received lime. This is in conformity with the well-established principle of the calcium-boron relationship in plants, which is that any increase in the rate of absorption of calcium results in a decrease in the intake of boron and in the availability of the boron within the plant.

No boron deficiency symptoms were observed on beets grown on soils treated with boron. Only one soil not treated with boron showed a definite boron-deficiency symptom when beets were grown on it. The beet showing the boron deficiency had a calcium-boron ratio of 273 in the leaves and 27 in the roots. The low ratio in the roots is attributed to the low calcium content in the root.

Table 9. Calcium and boron content of alfalfa leaves and stems from greenhouse studies on eight soils from Craig County where boron was the variable factor.

	Crop 1			Crop 2			Crop 3		
Soil	Ca	B	Ratio	Ca	B	Ratio	Ca	B	Ratio
No.	ppm	ppm	Ca/B	ppm	ppm	Ca/B	ppm	ppm	Ca/B
Alfalfa Leaf Material									
1	26,500	57.5	460	26,700	42.5	629	25,400	37.5	679
2	19,700	85.0	232	20,800	55.0	378	20,100	41.5	484
3	14,100	90.0	157	17,200	85.0	202	15,200	62.0	245
4	16,600	80.0	208	17,400	60.0	290	16,800	51.5	326
5	24,400	72.5	337	25,200	57.5	438	23,900	48.0	498
6	19,700	85.0	232	20,200	62.5	323	19,300	51.5	375
7	20,000	80.0	250	21,800	57.5	379	19,800	55.0	360
8	20,900	67.5	310	21,300	80.0	266	20,100	67.5	298
A	25,600	20.0	1,280	24,500	15.0	1,630	24,300	12.5	1,940
B	22,500	45.0	500	21,500	25.0	860	20,300	21.5	944
C	20,000	25.0	800	18,900	32.5	582	18,600	27.5	676
D	18,800	42.5	442	17,800	35.0	509	17,300	30.0	577
E	22,500	50.0	450	23,100	25.0	924	21,600	22.5	960
F	18,800	27.5	684	19,600	30.0	653	17,400	20.0	870
G	20,900	42.5	492	21,400	30.0	713	18,900	27.5	687
H	19,700	57.5	343	20,700	45.0	460	18,700	42.5	440
Alfalfa Stem Material									
1	16,300	20.0	815	16,000	10.0	1,600	15,100	8.5	1,780
2	13,300	32.5	409	12,600	22.5	560	11,900	20.0	595
3	9,200	32.5	283	10,300	30.0	343	8,900	27.5	324
4	9,200	40.0	230	10,500	27.5	382	9,100	22.5	404
5	12,100	57.5	210	12,300	25.0	492	11,100	17.5	634
6	11,600	30.0	387	12,300	35.0	351	12,000	27.5	436
7	8,300	32.5	255	8,000	32.5	246	7,700	30.0	257
8	10,800	45.0	400	7,600	30.0	253	7,800	30.0	260
A	13,800	10.0	1,380	13,100	7.5	1,750	11,000	5.0	2,200
B	10,400	30.0	467	9,200	11.5	800	8,700	7.5	1,160
C	11,700	12.5	936	12,100	10.0	1,210	10,900	7.5	1,450
D	9,600	22.5	427	10,100	15.0	673	9,800	10.0	980
E	12,100	27.5	440	11,200	20.5	546	10,200	15.0	680
F	10,000	30.0	333	10,300	26.5	389	9,600	22.5	427
G	9,600	27.5	349	9,200	24.5	376	8,900	22.5	396
H	11,700	32.5	360	10,300	30.5	338	9,900	27.5	360

Numbered samples 1 to 8 are the same as lettered samples A to H except that the lettered samples did not receive boron. 1. Bates silt loam 2. Parsons silt loam 3. Hanceville f.s.l. 4. Bates silt loam 5. Bates silt loam 6. Parsons silt loam 7. Parsons silt loam 8. Summit clay.

Table 10. Calcium and boron content of beet leaves and roots from greenhouse studies on eight soils from Craig County, Oklahoma where boron was the variable factor.

	With Boron			Without Boron		
Soil No.*	Ca ppm	B ppm	Ratio Ca/B	Ca ppm	B ppm	Ratio Ca/B
Beet Leaves						
1.	5,300	50.0	106	4,100	15.0	273
2.	4,100	45.0	91	4,500	20.0	225
3.	2,830	100.0	28	4,000	22.5	178
4.	3,310	95.0	35	3,760	50.0	75
5.	4,880	95.0	51	4,500	47.5	95
6.	3,960	122.5	32	3,760	45.0	84
7.	4,000	105.0	38	4,020	40.0	101
8.	4,020	62.5	64	3,980	52.5	76
Beet Roots						
1.	420	35.0	12	340	12.5	27
2.	330	42.5	8	260	15.0	17
3.	210	72.5	3	290	20.0	15
4.	210	60.0	4	220	20.0	11
5.	330	57.5	6	400	25.0	16
6.	200	77.5	3	250	22.5	11
7.	200	70.0	3	240	20.0	12
8.	270	50.0	5	290	22.5	13

* 1. Bates silt loam 2. Parsons silt loam 3. Hanceville f.s.l. 4. Bates silt loam 5. Bates silt loam 6. Parsons silt loam 7. Parsons silt loam 8. Summit clay.

The yellowing caused by boron deficiency is generally confined to the upper portion of the plant (Fig. 2). The appearance of the yellows depends on the severity of the deficiency. In most cases the first cutting is green with yellows or bronzing showing in the second and latter cuttings.

Soil types do not determine the occurrence of alfalfa yellows or response of alfalfa to boron applications. In general, however, the sandier soil types give a more consistent response to boron fertilization.

Fig. 2 Alfalfa yellows. Shortage of boron causes the terminal leaves to turn yellow.

Leaf symptoms are often the best means of detecting a severe boron deficiency in garden beets. Boron deficient beet leaves have a glistening, darker green color, are down curled, and brittle. Eventually, the older leaves become yellowed and wilt, and the entire center of the growing point turns black (Fig. 3).

Fig. 3 Photo by Horace J. Harper. Oklahoma Agriculture and Mechanical College. Garden beets in boron deficient soil from Craig County, Oklahoma. Unhealthy terminal bud in pot without boron.

No visible symptoms of boron deficiencies were observed on the beet roots but noticeable differences in yield were observed (Fig. 4).

Fig. 4 Photo by Horace J. Harper. Oklahoma Agricultural and Mechanical College. Garden beets grown in boron deficient soil from Craig County, Oklahoma.

DISCUSSION

This study concerning the water soluble boron in soils and the response of alfalfa and garden beets to boron fertilization has shown that boron must be present in the soil in certain quantities for the plant to make proper growth. It is also apparent, from a review of the literature on the needs of certain crops for boron, that all crops are not alike with reference to the amount of available boron needed to make good growth. In view of these facts one would hesitate to say that a certain soil is deficient in available boron even though an analysis shows that it contains less water-soluble boron than another.

After reviewing the literature on recommendations for boron fertilization for alfalfa and garden beets, it seems that a recommendation for one area may not apply in another area due to amount of rainfall, soil texture, and soil reaction. Analysis of soils to determine the amount of available boron should be conducted before any recommendation or application of boron is made. In view of the foregoing statement as to the amount of available boron in soils, it is apparent that field experiments should be conducted in areas that are found to be low in available boron using crops that are usually grown in those areas. Until this is done, no definite recommendations can be given. A plant composition study may be the best approach to the problem of determining the relative availability of boron in different soils.

SUMMARY

A study was made of the water-soluble boron in eight soils from Craig County, Oklahoma.

Analytical methods for determining water-soluble boron in soils and total boron in plant material were studied and the methods described.

About 200 pounds of each of the 8 soils was collected and placed in six pots. Alfalfa was planted in four pots and garden beets in two pots. The alfalfa pots received P_2O_5 and K_2O in the form of potassium monohydrogen phosphate and magnesium sulfate each at the rate of 100 pounds per acre. Lime was applied at the rate necessary to bring the pH to neutrality. The alfalfa seed was inoculated with legume culture "A". The beet pots received the same fertilizer treatment except that nitrogen was applied as ammonium nitrate at the rate of 100 pounds of N per acre. Boron was applied as sodium borate at the rate of 1 ppm to two of the alfalfa pots and to one of the beet pots in each group of soils. The moisture content of each soil was adjusted to one-third of the water holding capacity and maintained at this value by frequent weighing.

Three cuttings of alfalfa were made. The beets were pulled at maturity. Chemical analyses were determined on the leaves and the stems of the alfalfa and on the leaves and the roots of the beets for total nitrogen, phosphorus, potassium, calcium and boron. The results indicate that a plant with a high calcium-boron ratio will exhibit boron deficiency symptoms.

Soils containing less than 0.35 ppm of available boron would be considered deficient for alfalfa and soils containing less than 0.10 ppm of available boron would be considered deficient for garden beets.

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