YIELD AND NUTRITIVE STATUS OF ALFALFA AS INFLUENCED BY THE BORON CONTENT OF SOILS WITH SPECIAL EMPHASIS ON A PRACTICAL AND RELIABLE BIOLOGICAL TESTING PROCEDURE

By

AARON SIDNEY BAKER

AN ABSTRACT

Submitted to the School of Graduate Studies of Michigan
State University of Agriculture and Applied Science
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Soil Science

1955

Approved: R. L. Cook
J. Ty.

ABSTRACT

The primary purposes of this work were to determine the extent of boron deficiency on alfalfa meadows in the lower peninsula of Michigan and to discover a reliable and practical testing procedure for predicting whether a soil was likely to produce boron deficient alfalfa. A secondary objective was to investigate the influence of the boron level in the soils and plants on the yield and nutrient composition of alfalfa.

The studies took the form of a brief field survey, field plot experiments, greenhouse pot experiments and analytical laboratory work.

It was observed that boron deficiency on alfalfa was quite prevalent on droughty, coarse textured soils and also occurred to a lesser extent on soils of intermediate texture. No boron deficiency was observed on the very fine textured soils. It was also noted that boron deficiency did not occur in the spring or early summer but was restricted entirely to the second and third crops on alfalfa meadows.

Yield and quality responses to borax applications were demonstrated in the field and greenhouse when the check plots were boron deficient. The apical portions of deficient alfalfa were found to be lower in boron, calcium, potassium, and magnesium, than were the more mature nondeficient portions of

the plant. The boron, calcium, potassium, magnesium and protein contents of the boron deficient portions of alfalfa were found to be lower than in the same portions of healthy plants.

Soil tests were found to be unreliable for predicting the boron supplying power of the soil especially when two or more soils were compared. This is true because of inherent limitations of present soil testing procedures and due to the fact that plants may not be absorbing their nutrients from the surface layer when this soil horizon is very dry. For these reasons it was suggested that the apical portions of plants be sampled and tested for boron during an extended hot dry period. If the boron level is 20 p.p.m. or less in this portion of the plant then it is likely that boron deficiency will occur when the surface soil becomes very dry.

It was shown that the boron associated with soil organic matter under alkaline conditions is in a different chemical form than boron associated with mineral soils. The former is much more soluble in hot distilled water than is the latter. It was also demonstrated that soluble boron compounds are "fixed" much more rapidly by alkaline organic soils than by the two mineral soils used in the greenhouse experiments.

Variations of the boron levels in the soils and plants above deficient levels were found to produce no significant differences in the yield and composition of alfalfa. Toxic levels of boron were not attained.

It was demonstrated that boron deficiency can occur on alfalfa grown on acid soils if relatively thrifty plants are produced.

The so-called "fixed" forms of soil boron were found to be readily available to alfalfa although not as available as the original soluble form which was applied to the soil.

An extensive review of the literature and a summary of the practical applications of the conclusions from this work are presented.

YIELD AND NUTRITIVE STATUS OF ALFALFA AS INFLUENCED BY THE BORON CONTENT OF SOILS WITH SPECIAL EMPHASIS ON A PRACTICAL AND RELIABLE BIOLOGICAL TESTING PROCEDURE

By

AARON SIDNEY BAKER

A THESIS

Submitted to the School of Graduate Studies of Michigan
State University of Agriculture and Applied Science
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Soil Science

1955

4/30/57
g989

ACKNOWLEDGMENTS

The author wishes to express his gratitude to Dr. R. L. Cook for serving as chairman of his committee and for the ready assistance which was forthcoming whenever a problem arose. Dr. Cook's advice and guidance were invaluable in the formulation of experiments and editing of this thesis.

He is indebted also to Drs. K. Lawton, A. E. Erickson, G. P. Steinbauer, and L. M. Turk for serving on his special committee.

The writer also wishes to express his appreciation to Mr. Grant Davis and Mr. Edward Kitchen of the Pacific Coast Borax Company for their assistance in the performance of field studies.

Gratitude is also extended to the Mid-West Soil Improvement Association for the funds that were provided for this work and the special graduate research assistantship.

VITA

Aaron Sidney Baker

candidate for the degree of

Doctor of Philosophy

Final examination, July 26, 1955, 10:00 A.M., Room 210, Agriculture Hall.

Thesis:

Yield and Nutritive Status of Alfalfa as Influenced by the Boron Content of Soils with Speical Emphasis on a Practical and Reliable Biological Testing Procedure

Outline of Studies:

Major subject: Soil Science

Minor subjects: Chemistry, Plant Physiology

Biographical Items:

Born; January 27, 1924, St. Thomas, Ontario, Canada

Undergraduate studies; New York State College of Agriculture, Cornell University, Ithaca, N.Y., 1946-1950, B.S. degree with major in Agronomy

Graduate studies; New York State College of Agriculture, Cornell University, Ithaca, N. Y., 1950-1951, M.S. degree with major in Agronomy and minor in physical chemistry

Michigan State University of Agriculture and Applied Science, East Lansing, Michigan., 1951-1955.

Experience:

Work on New York State Agricultural Experiment Station during summer of 1950

Special Graduate Research Assistant, Michigan State University, 1951-1955

Member of Sigma Xi, American Society of Agronomy, and Soil Science Society of America.

TABLE OF CONTENTS

TABLE OF CONTENTS-- Continued

LIST OF TABLES

TABLE Page

LIST OF FIGURES

INTRODUCTION

In recent years symptoms of boron deficiency have been noted on alfalfa meadows in the state of Michigan. Surrounding states with soils and climate quite similar to those of Michigan have been making extensive studies of the problem and are making regular recommendations for applications of borax on alfalfa meadows. For these reasons a study was begun in September of 1951 with the specific purpose of determining the extent of boron deficiency on alfalfa in the lower peninsula of Michigan and whether top dressings with fertilizers containing borax would be worthwhile. Special emphasis was placed on a search for a reliable and practical method for predicting whether a soil was likely to produce boron deficient alfalfa. The work involved a brief field survey, field plot experiments, greenhouse pot experiments and analytical laboratory work.

REVIEW OF THE LITERATURE

Introduction

There is a very large store of literature on boron as a factor in plant growth but there is still much to be learned for a complete understanding of the subject. By reviewing some of this literature it should be possible to glean some useful generalizations and explain some of the experimental results obtained in the present work.

Economic Value of Borax Applications on Deficient Soils

It is reasonable to expect a decrease in yield if the metabolism of a plant is upset due to a deficiency of one of the essential elements. In the case of boron however, the deficiency, as will be discussed later, quite often occurs late in the growth cycle of the alfalfa. It also tends to occur during droughty periods when growth would be slow even in non-deficient plants. It is uncommon, therefore, to obtain noticeable yield increases in alfalfa due to boron applications, although large increases in seed production have often been reported (4, 23, 28, 51, 57, 63). Russel (57) found that forage yields from the first, second and third cuttings of alfalfa were not significantly increased by the removal of the deficiency symptoms through borax fertilizer treatment. However, some investigators have reported vegetative responses to borax applications on alfalfa (28, 51, 56, 59, 63).

It is still questionable as to whether alfalfa will respond to boron where the element is plentiful enough to produce plants without deficiency symptoms. Wadleigh (65) noted a marked decrease in the pH of scattered cells in the meristematic tissue of boron deficient plants even before deficiency symptoms were apparent. Walker (66) stated that the influence of boron deficiency may be noted microscopically before it is seen macroscopically. Hutcheson (28) refers to some instances of alfalfa responding favorably to boron even when there were no outward signs of a deficiency, but Dawson (16) found in his work that boron deficiency symptoms appeared before the yield of alfalfa was limited. Working with red beets and sugar beets Berger (6) obtained responses to boron applications where deficiencies were, heretofore, unnoticed. Smith (62) found that orange trees grown in nutrient cultures showed no differences in growth or yield and quality of fruit when boron was supplied at three different levels between, but not including, deficiency and toxicity levels.

Another factor that warrants discussion when considering the economics of boron applications to deficient soils is crop quality. Barber (4) noted that in Indiana alfalfa response was mainly in quality of hay. Yield responses were very small since yellowing occurred but stunting did not. Russel (57), in an attempt to test the quality of alfalfa due to elimination of boron deficiency, found no correlation either with leaf-to-stem weight ratios or total protein content. Cook (15) working with spinach and sugar beets found that borax treatments increased yields and eliminated deficiency symptoms but decreased the nitrogen content of these plants. Perhaps this decrease was simply a result of dilution. On the other hand there is evidence that the protein or nitrogen content of plants is increased by boron applications. Kochler (33) stated that plants produced under conditions of adequate supplies of all essential nutrients including trace elements had a better balance of amino acids and required less dry matter to produce equal gains in rabbits. Experimenting with alfalfa and soybeans, Sheldon (60) found very marked decreases in the tryptophane content of these plants when boron was reduced or withheld from the nutrient medium. He believed that the quality of a plant may well be lowered before it actually shows visable deficiency symptoms. Investigating the nitrogen nutrition of pea plants in nutrient solutions and soils, Mulder (40) discovered that nodulation did not occur when boron was omitted from the nutrient medium. Peas grown in nutrient solutions required more boron than did the nodules, but the reverse was true in a soil experiment. Jordan (30) found that boron treatments in soils and pure cultures increased nitrogen fixation by azotobacter chroococcum, a nonsymbiotic type of nitrogen fixing bacteria.

There is also some evidence that top dressing deficient meadows with borax tends to increase the longevity of alfalfa stands especially when the deficiency is severe (10, 28, 63).

Physiological Effects of Boron on Alfalfa

A. Symptoms of boron deficiency in alfalfa.

The visual symptoms of boron deficiency have been quite clearly defined in the literature (4, 13). The terminal leaves yellow and redden while the internodes shorten, forming a rosette, followed by the death of the terminal bud. Little or no flowering or seed set occurs. Walker (66) described symptoms of boron deficiency that can been seen microscopically before visual symptoms can be noted. The first effect is a more rapid cell division and growth of meristematic and cambium tissue concurrent with less cell wall formation and less differentiation of the cells. Thus the development of xylem and phloem is interrupted. This causes the visual symptoms mentioned above because of less efficient conduction of plant nutrients to the growing portions.

B. Functions of boron in plants.

Boron apparently is functional in some way in the young, rapidly growing tissue of plants. As mentioned above Walker (66) observed this microscopically. Wadleigh (65) found that not only was the meristematic tissue of the above ground portions of plants affected, but the root tips of cotton seedlings grown on boron deficient soils were dead. Haynes (26), using a split root technique, showed that boron was a necessary component of the soil solution wherever the roots of tomato plants were in contact with water. Leggatt (35) found that even such rapidly growing tissue as germinating seeds was adversely affected when boron was absent from the medium and seeds came from boron deficient plants. Struckmeyer (64) discovered that reducing the cambium activity of many plants by controlling the photoperiod eliminated boron deficiency symptoms, although the boron content of the tissue was not altered.

The question remains as to just what function boron performs in rapidly growing tissue. Many workers (2, 5, 6, 65) are of the opinion that boron is active in carbohydrate oxidation since sugars tend to accumulate in deficient plants. Boron deficient plants also tend to accumulate ammonium nitrogen and are lower in protein and amino acids. This latter condition is explained by Beckenback (5), Berger (6), and Wadleigh (65) as a secondary effect of a lack of carbohydrate oxidation because the by-products of this process are required by plants to form amino acids from ammonium nitrogen. As evidence of the fact that boron is active in carbohydrate metabolism Bailey (2) showed that in alfalfa plants the activity of the enzyme invertase was increased 100 % over the check when boron was supplied at the highest level. He also found that the activities of catalase, peroxidase and oxidase were increased slightly by increased boron supply but this was interpreted as being due to improvement of the general metabolic condition within the plant.

C. The relationship between boron uptake and the absorption of other ions by plants.

There is a large store of literature concerning the interrelationships of boron and other ions so far as absorption is concerned but some of the results are quite contradictory. Parks (48) pointed out the noteworthy lack of agreement among investigators as to the specific effect of boron on the accumulation of any given element.

Calcium has been most frequently investigated with respect to boron metabolism in plants. In 1937 Naftel (42) noted that the overliming injury of alfalfa grown on a Norfolk loamy sand could be entirely eliminated with borax applications. At that time it was not known whether it was the increased demand for boron due to growth increments from liming, the higher concentration of calcium ions in the soil and plant or the higher soil pH that caused this deficiency. At present there is evidence that all three factors may be directly or indirectly involved. Rogers (56) concluded that alfalfa requires only very small amounts of boron on soils of low calcium supply and low base exchange capacity. Berger (6) stated that the only elements that tend to influence boron directly or be influenced directly by boron are calcium and nitrogen. Several investigators (9, 29, 39, 55) have indicated that increased calcium in plants reduced the boron uptake. Purvis (52) stated that there is a functional relationship between calcium and boron. That is, as one element is taken up in larger quantities the requirement for the other increases. This does not mean, of course, that this requirement is fulfilled and therefore does not contradict workers who found increased calcium uptake intensified boron deficiency symptoms or reduced the boron content of the plant. The above statement by Purvis does, however, bring up the question as to whether an increased boron concentration in plants influences the calcium content. Some investigators (9, 15, 37, 55) found no correlation between the boron content of plant tissue and the percent of calcium. However, Marsh (55) noted that soluble calcium in corn tissue was determined, not by the total calcium of the plant, but by the boron content. Jones (29) and Smith (62) found that increased boron in the tissue resulted in increased calcium content. Parks (48) discovered that normal plants were higher in calcium than were either boron deficient plants or plants showing boron toxicity, but he only tested one level of boron in the normal plant range. It is thus apparent that there is still some question as to the influence of boron levels on the uptake of calcium by plants.

An alternate means for investigating the relationship between calcium and boron in plants has been the consideration of Ca/B ratios. Drake (20) stated that the Ca/B ratio in the tobacco plant was important to the formation of boron deficiency symptoms, although he did not attempt to discover if this ratio were the same when other conditions of the environment were varied. Schaller (58) found that he could produce boron deficiency in alfalfa when the Ca/B ratio ranged from 667 to 1,250. Jones (29) obtained no boron deficiency or toxicity symptoms in alfalfa when the Ca/B ratio was varied from 80 to 600. One would not be justified in stating from the results of Schaller, op. cit., that the Ca/B ratio is not relatively constant at a critical level for the reason that boron in the plants may well have decreased below a critical level before samples were taken. If this value is relatively constant at some critical level, the results of Jones, op. cit., would eliminate any value below 600 and the lowest possible value from Schaller's data would put the critical level at about 700.

Another element which has been given some attention in connection with boron uptake by plants is potassium. Berger (6) pointed out that potassium influences or is influenced by boron only indirectly. If it is true that boron influences the uptake of calcium and the well known reciprocal relationship between calcium and potassium in alfalfa is considered, it is obvious that boron may well have an indirect influence on potassium uptake. On the other hand when calcium is increased in the plant tissue, then potassium is reduced by the afore mentioned reciprocal relationship and boron may also be reduced due also to the increased concentration of calcium in the plant. These types of relationships make it difficult to investigate boron, calcium, potassium and magnesium relationships in plants. Reeves (55) noted that both boron deficiency and toxicity in tomato plants grown in nutrient solutions were accentuated by increased uptake of potassium. He also found that boron was increased in the plant tissue when potassium was increased even through boron deficiency symptoms were intensified. These results are impossible to explain on the basis of a reciprocal relationship between potassium and calcium and therefore suggest a functional relation between potassium and boron. Wallace (67) working with alfalfa in nutrient solutions, also observed that increments of potassium in the solution and plant tissue intensified boron deficiency symptoms but also increased yields. In this case it may well have been that the increment in growth resulted in an increased demand on the already short supply of boron. Parks (48) showed that normal tomato plants grown in nutrient solution were lower in potassium than were plants exhibiting boron deficiency or toxicity symptoms. This may well have been due to a reciprocal relationship between calcium and potassium since, as mentioned previously, the opposite relation was found for calcium.

Nitrogen also appears to influence or to be influenced by boron uptake by plants (6). As mentioned previously this may be due to an indirect functional relationship. Evidence of this was presented by Wadleigh (65) who observed an increase in sugar and ammonium nitrogen in boron deficient cotton seedlings. This, he believed, was due to a breakdown in the carbohydrate metabolism, a process in which boron is somehow involved. He also observed a decrease in the nitrate content of boron deficient plants which he thought might be due to reduced uptake as the root tips were dead or necrotic. Parks (48) found that there was a general rise in the nitrogen level in tomato leaflets from deficient levels to toxic levels of boron. He thought that although plants affected by boron toxicity were very proteinacious, this may have been due to stunting. Smith (62) could find no difference in the nitrogen content of orange leaves when boron was supplied at three different levels, none of which caused deficiency or toxicity. Mulder (40) discovered that the protein of pea plants was raised when boron was supplied to deficient soils, due to increased nitrogen fixation by symbiotic nitrogen fixing bacteria.

Cook (15) found that NH_4NO_3 applications decreased the boron content of dried sugar beet root tissue. Bechenback (5) was able to show that tomato plants grown in nutrient solutions containing ample nitrates required many times more boron than did nitrogen starved plants.

Other nutrient elements such as phosphorus, magnesium, sodium, iron, molybdenum, and sulfur have been investigated in connection with boron in various plant species. Cook (15) observed an increase in the percent of magnesium in sugar beet roots but not in spinach when borax was applied to the soil. Using the soybean as an indicator plant, Muhr (39)found that $MgCO_3$ and to a lesser extent $MgSO_4$ treatments on the soil caused a decrease in the boron content of the plant tissue. Parks (48) noticed that magnesium was higher in normal tomato leaflets than in either those showing boron deficiency or toxicity. Magnesium was found by Smith (62) to be in highest concentration in orange leaves at the lowest boron levels. As mentioned previously, his treatments were such that neither boron deficiency nor toxicity occured at any of the boron levels of the nutrient solutions. Smith also noted that phosphorus was slightly higher at the lowest boron level. Generally, phosphorus is found to be in higher concentration in boron deficient plants (5, 48).

Cook (15) found less iron in sugar beet roots and spinach tops when the soil was treated with borax. Parks (48) found that sulfur and sodium were higher in normal tomato plants than in plants showing boron deficiency or toxicity symptoms.

In contrast molybdenum increased in the plant tissue with increased boron in the nutrient solution and decreased only at the most toxic level. Muhr (39) observed that $NaCO_3$ and $NaSO_4$ applications on soils caused no changes in the boron concentration in soybean tissue.

D. The critical level of boron in alfalfa.

An evaluation of the critical level of boron in plants, involves consideration of the following important facts:

1. Boron is not translocated from the older to the newer growing portions of plants when the supply in the soil becomes limiting.
2. Leaves are much higher in boron than are stems.

These facts have been well confirmed by many workers (1, 9, 18-174 pp., 19, 63). For these reasons the meristematic portions of boron deficient alfalfa are found to be lower in boron than are the more mature parts. Conversely, in normal alfalfa plants the tops tend to be higher in boron than are the basal portions due to the higher leaf to stem ratio at the top (19, 63).

Another factor that should be taken into account when the critical level of boron in plants is considered is that the requirement for boron may vary with the environment. Purvis (52) pointed out that as calcium is taken up in larger quantities the requirements for boron increase. The investigations performed by Reeves (55) indicated that increasing the potassium supplied to tomato plants increased the boron in plants although it intensified boron deficiency symptoms. On the other hand Wallace (67) observed that the boron concentration of leaves of boron deficient alfalfa grown in a nutrient solution of a low potassium level was 26 p.p.m. while it was only 10 p.p.m. when grown in high potassium nutrient solution. He also presented evidence from the literature that boron requirement and uptake are dependent on many factors such as, temperature, osmotic pressure of the nutrient media, and amounts and ratios of many elements as calcium, magnesium, nitrogen and potassium. Other workers (5, 40) discovered that plants required much more boron when supplied with adequate nitrogen than when they were nitrogen deficient. Bechenback (5) noted the opposite relation between boron and phosphorus. Struckmeyer (64) observed that by shortening and lengthening the photoperiod he prevented or enhanced the onset of boron deficiency in many plant species without altering the boron content of the plants.

In view of the many factors affecting boron uptake and requirement it is little wonder that the critical level for sufficiency in alfalfa varies so widely in the reports of different investigators. Rogers (56) found no yield increase from borax applications if alfalfa contained more than 10 p.p.m. Strickly speaking, this is not a critical level for boron deficiency but for a yield response. He pointed out that this figure was, in all probability, low for fine textured soils and only applied to the coarse textured, red and yellow podzolic soils of Alabama that have a low calcium supply and low base exchange capacity. He also published the following list of critical values for alfalfa reported by other workers.

Investigators	Amount of B reported in deficient plants (p.p.m.)
McLarty, Wilcox and Woodbridge	6.9
Berger and Truog	8.0
Haddock and Vandecaveye	10.0
Powers	10.0
Dregne and Powers	7.0 to 11.5 deficient plants 12.0 to 22.5 normal plants
Jordan and Powers	12.0
Dunklee and Midgley	15.0
Brown, Munsell and King	17.0 also 17.0 with no response to B
Whetstone, Robinson and Byers	12.0 to 17.0 response to B 13.0 to 19.0 no response to B
Dawson and Gustafson	20.0
Munsell and Brown	23.0 in leaves that were yellowed

The following are added by the author and were obtained from more recent papers.

Investigators	Amount of B reported in deficient plants (p.p.m.)
Barber (4)	20.0
Dible and Berger (19)	9.0 in apical portions
Schaller (58)	19.0
Stinson (63)	20.0

Several of these investigators qualified their reported critical levels with statements to the effect that they may be higher or lower in certain instances (16, 31, 56). The critical value of 9 p.p.m. of boron in the apical portions of deficient plants obtained by Dible (19) was derived from a few field trials and a nutrient solution experiment, in which the composition of the solution was not changed except for boron. Perhaps this value would be different under a variety of environmental conditions. It should be noted that the critical levels reported by all the above mentioned workers fall below 20 p.p.m. of boron except in the one case in which only the leaves were analyzed.

Boron in the Soil

A. Methods of extraction of boron from soils.

Most of the investigators experimenting with boron in soils have used a method of extraction similar to the one described by Berger (7). This method consists mainly of refluxing a 1:2 soil-water mixture for five minutes, separating the water from the soil by filtering or centrifuging and determining the boron in the water fraction by one of several colorimetric methods. Although Berger (7) found that little or no extra boron was dissolved from soils after five minutes of boiling, Rogers (56) found that this did not hold true for all soils. Haas (24) noted that the boron extracted was generally increased by decreasing the soil to water ratio. This indicates that a solid phase-liquid phase equilibrium is in operation in the hot water extraction technique. McClung (36) and Baird (3) essentially decreased the soil to water ratio to a very small figure by using a soxhlet extraction method which increased the boron released by soils and accounted for the boron removed by sunflowers much more accurately than did the hot water extraction technique of Berger (7). Page (46) also found that sunflowers released more boron than could be accounted for by the decrease in hot water soluble boron. Others (11, 22) maintain that biological tests are superior to chemical tests of the boron supplying power of soils. Many workers (4, 14, 41) were unable to correlate boron uptake or boron deficiency with the boron extracted from soils by the five minute boiling procedure. McClung (36) noted that the boron released by the soxhlet method was approximately four times greater than that obtained by the five minute boiling technique. He worked with only three acid, course textured New York soils while Baird (3) working with more soils and a greater variety of soils found no such correlation between the two methods.

Because the extractant, hot distilled water, is the same in both methods, the boron extracted must come from similar solid phase compounds. The soxhlet extractable boron therefore should be a measure of the capacity of the soil to supply boron to the plant and the five minute boiling technique ought to provide a measure of the equilibrium concentration in the soil solution. It is obvious that a knowledge of both values is necessary for predicting how much boron can be removed by a plant during a growing season when other conditions of growth are controlled. Baird (3) found that he got the best correlation between sunflower yield and soil tests when both soxhlet extractable boron and boron extracted by the five minute boiling procedure were considered.

Another method for extracting boron from soils is described by Whetstone (69). It consists essentially of digesting the soil in concentrated phosphoric acid and distilling off the boron with methyl alcohol. He found that tourmaline was not acid soluble and suggested acid soluble boron be considered as all the available boron there is in soil organic matter, precipitated borates and in clays.

B. Fixation and availability of boron in soils.

The fixation and availability of boron in soils has been found to be related to many soil properties and constituents such as reaction, various cation and anions, texture and organic matter.

Many investigators have found that increased hydrogen ion concentration in the soil caused increased availability of boron or a decrease in the rate of fixation of boron (22, 39, 44, 61, 69). However Drake (20) and Reeve (54) noted that variations in pH had little or no effect on fixation although the latter worker stated that crops grown on well limed soils were found to be more responsive to boron than were those grown on acid soils.

A large number of workers have shown that the cations associated with changes in pH are as important, if not more so, than the hydrogen ion concentration in affecting the availability of soil boron. Dregne (21), Parks (47), Purvis (52), and Reeve (54), to mention only a few, found that additions of lime caused decreases in the availability of soil boron. Parks (47) believed that additions of lime caused fixation both by raising the pH and by the effect of the calcium ion in mixtures being precipitated. The majority of workers (14, 34, 49, 50, 61, 74) found that ions such as calcium and magnesium fixed more boron than did sodium and potassium although a few investigators (20, 43) were unable to notice any difference in boron fixation with the addition of various bases to the soil. Neutral salts of calcium such as $CaCl_2$ or $CaSO_4$ were not found to decrease boron availability as much as did $Ca(OH)_4$ or $CaCO_3$ and even appeared to increase the availability in some cases (14, 34, 39, 61, 74). It would appear from these investigations that high pH values must coincide with high concentrations of calcium and magnesium in soils in order to obtain maximum fixation of boron.

It has been fairly well authenticated that the texture of soils has a profound effect on the fixation and availability of boron. Cook (14) found that excessive leaching conditions leads to boron deficiency. Page (46) found a low correlation between hot water soluble boron and the silt and clay fractions of the soil. Although Baird (3) obtained the same type of results between boron extracted by the five minute boiling procedure and the specific surface of soils, his soxhlet extraction method gave a very good correlation. This indicates that the boron supplying power of soils may be a function of the clay content while the relative equilibrium concentration of boron in the soil solution may not. In an earlier paper Page (45) stated that finer textured and high organic matter soils can stand higher boron applications without causing boron toxicity than can coarse textured soils which are low in organic matter. This latter indicates that finer textured soils are able to fix more boron even though texture does not appear to be correlated with the boron extracted by the hot water extraction technique. As if to corroborate this point Whetstone (69) found that acid soluble boron was directly related to the colloid content of the soil. Moreover he found that within the separate of two microns or less, the finer fractions had the highest acid soluble boron content. Whetstone's acid soluble boron might well be analogous to Baird's (3) soxhlet extractable boron. Kubota (34) and Olson (44) both noted that the rate of fixation of boron, as measured by Berger's (7) five minute boiling technique, was highest in soils with the greatest clay content.

Many investigators found that boron was more concentrated in the surface along with soil organic matter than in the lower horizons (19, 21, 27, 44). Page (46) was able to get a good correlation between hot water soluble boron and soil organic matter while he found only a low correlation with silt and clay content and soil pH. Conversely, Berger (8) observed that pH exerts a greater influence on hot water extractable boron in alkaline soils than does organic matter. The reverse was found to be true in acid soils. Parks (50) in confirmation of Berger's results was able to show that a hydrogen saturated humus extract fixed more boron than did a calcium saturated one. Parks (50) also proposed a mechanism for boron fixation by organic matter. It is well known that a "favorable" diol or an alpha hydroxy acid will react with boric acid in water. He cited evidence that these types of compounds are present in decomposing organic matter. However he offers no evidence as to the stability of these compounds in the soil and they appear to be simple and readily metabolizable compounds. Perhaps they would be more stable when complexed with the boric acid. Olson (44) found that removal of soil organic matter resulted in a slight decrease in the fixation of boron while oxidation of organic matter resulted in an increase in hot water soluble boron.

Some investigators (42, 53) suggested that boron might be fixed by the increased microbiological activity brought about by liming but others (10, 31, 54) found that additions of fresh organic matter tended to increase the availability of soil boron. Rogers (56) found that sterilization of soils with toluene had no effect on boron fixations.

Although the literature is in conflict as to the specific effect of organic matter on fixation and availability of boron it appears to play only a secondary role in most mineral soils. In organic soils boron often appears to be a limiting factor for growth (21, 69).

Another property of soils that should be considered is age. As mentioned previously, Cook (14) stated that excessive leaching conditions leads to boron deficiency. It follows that the relative amount of time of leaching should also influence the boron status of soils. In this connection many workers found that the older soils tended to show boron deficiency more than did the younger ones (4, 21, 69). Hutcheson (28) stated that boron deficiency was not as prevalent on naturally alkaline soils as on acid ones. This latter may well be due to the fact that alkaline soils usually do not have a history of extensive leaching but also may be because boron is more soluble in acid soils and therefore is depleted more rapidly.

Another factor that appears to be important to the availability of soil boron to plants is the moisture status of the soil. It is quite well authenticated that boron deficiency is much more common in humid regions during long periods of hot dry weather when the surface of the soil is dryed out (6, 16, 21, 22, 59, 63). Using a split root technique, Hobbs (27) showed that plants were boron deficient when the surface soil was allowed to dry out and the subsoil was kept moist. This even occured when the surface received a borax application. Drying has been found to decrease hot water extractable boron in soils and clay separates (47,50). On the other hand Winsor (72) observed that hot water soluble boron was as high or higher during dry summer seasons as during the wet summer seasons. This latter may be explained by the higher rate of crop removal and leaching during the wet seasons. Jordan (31) working in Oregon noted that irrigation with water low in boron intensified boron deficiency symptoms. Again this might be explained on the basis of leaching loses and greater plant growth creating a greater demand on the soil for boron. The bulk of the literature points to the fact that dry soil conditions occuring in humid regions often bring on boron deficiency symptoms. This deficiency is probably caused in one of two ways or a combination of both.

1. Fixation of boron in a form relatively insoluble in water.
2. Lack of water to move readily soluble boron compounds into the plant.

C. Possible chemical forms of soil boron.

In spite of the fact that the solid phase of soil boron may exist in complex forms it is quite probable that boron in the soil solution exists as simple molecules and ions such as boric acid (H_3BO_4) or tetraborate ions ($B_4O_7^{=}$) (3, 12, 22).

Eaton (22) classified the solid phase of soil boron into three possible classes.

1. Molecularly adsorbed boron
2. Ionically adsorbed boron
3. Boron precipitated in relatively insoluble compounds.

After extensive investigations on many soils of quite different properties Baird (3) decided that the major source of available boron in soils is associated with silicon. It appeared that dissolution or hydrolysis of silicon was necessary for the release of boron. In order to distinguish boron associated with silicon from simple borates and boric acid, acetone was used as an extractant in the soxhlet procedure and results compared to data obtained when water was used as the extractant. Acetone will dissolve these simple molecules without being able to hydrolyse and dissolve silicon. Only extremely small amounts of boron were dissolved in acetone indicating that very little of the solid phase boron was adsorbed by the soil in molecular or ionic form. It should be mentioned that the ratio of silicon to boron increased with successive extractions of the same soil with distilled water. Baird (3) accounted for this by stating that boron was selectively removed from the surfaces of particles during the first soxhlet extraction. Ground pyrex glass, a calcium boro silicate, was found to release silicon and boron in the same manner as does the soil except for the fact that the ratios were of a much lower order of magnitude. Another point that ought to be brought out here is that although the before and after cropping determinations of boron by the five minute boiling technique did not account for all the boron removed by the sunflowers, these values did decrease. This indicates that either a less soluble compound was releasing boron to the soil solution after cropping or that the same compound was being dissolved but that the equilibrium condition was not attained in the five minute boiling technique. This latter is probable if the selective dissolution of boron referred to previously takes place during crop growth. Fixation of boron in soils associated with alkaline conditions in the presence of calcium ions was considered by Baird (3) to involve formation of relatively insoluble calcium silicates.

It should be emphasized that Baird's (3) work does not eliminate the possibility of a portion of the available soil boron existing as long chain calcium metaborates. Colwell (12) stated that the formation of metaborates is favored by high concentrations of hydroxyl ions, low moisture and the presence of suitable cations. Calcium causes the condensation of very long chain metaborates while high concentrations of sodium and potassium cause the condensation of metaborates of more discrete size. The calcium metaborates are much more slowly soluble than sodium or potassium metaborates. Indications are that a chemical change takes place so that calcium metaborates do not dissolve as such. This change may well be a hydrolysis reaction and hence would not release boron to the acetone extractant used by Baird (3). It is quite probable however that all of the calcium metaborate would be removed during a six hour soxhlet extraction with water. Wear (68) experimented with three boron compounds; fertilizer borate (sodium metaborate), colemanite (calcium metaborate) and howlite (borosilicate). Using Berger's (7) hot water extraction method he found that the ratio of solubility of fertilizer borate: colemanite: howlite was 25:5:1 but only twice as much colemanite as fertilizer borate and two to three times as much howlite as colemanite was required to produce the same degree of toxicity on turnips in the greenhouse. Wear's work indicates that compounds like calcium metaborate and borosilicates can be used as sources of available boron and still not be leached too rapidly. More important is the fact that the five minute boiling technique for extracting boron from these compounds gave a very poor indication of their actual boron supplying power.

Parks (49) made curves of boron fixation against the pH of various systems. He found that the curve for a Ca + Si + Al + B system most nearly matched that of a Ca + Bentonite + B system which indicates that at least one means of fixation of boron in soils may be by precipitation in complex alumino silicates. The greatest amount of fixation was found to take place at pH 8 which is similar to the situation in soils. In a later paper (47) he stated that his data tend to support the mechanism of boron fixation brought about by wetting and drying, as the entrance of boron into the clay crystal lattice rather than by fixation by chemical precipitation, adsorption by clays or organic matter or microbiological fixation. That isomorphous substitution of boron for aluminium in alumino silicates may take place in soils, gains credence when one considers the chemical similarity of the two elements and the small atomic radius of boron (49).

Eaton (22) considered that increased fixation of boron caused by grinding kaolinite was proof of molecular or ionic adsorption. In view of Baird's (3) results from acetone extractions this appears to be improbable. The increased fixation could well have been due to increased release of aluminum and silica to the soil solution and subsequent precipitation with boron and calcium or increased isomorphous substitution.

Evidence that some form of available boron is constantly being replenished in soils from unavailable boron sources was presented by McClung (36) and Baird (3).

D. Critical level of boron in soils for alfalfa.

It is obvious from the foregoing presentation that the boron level of soils as measured by the five minute boiling technique is subject to criticism and even if this method were an accurate index of boron availability other factors in the environment profoundly affect the amount of boron absorbed and the amount required by plants. Nevertheless many investigators have attempted to correlate boron deficiency in alfalfa with the hot water soluble boron level of soils. As one would expect, these levels vary greatly but all fall below 0.75 to 1.00 p.p.m. (16, 18-81 pp., 21, 51, 54, 56, 63).

E. The distribution of boron in soils.

The types of soils which most commonly produce boron deficient alfalfa tend to fall into the following catagories (4, 8, 21, 28, 44, 54, 69):

1. Leached coarse textured soils.
2. Organic soils.

3. Old residual soils.

4. Naturally acid soils which have been limed.

Whetstone (69) presented a fairly complete set of generalizations concerning the distribution of boron in soils. He differentiated between acid soluble boron and acid insoluble boron. The former was considered to be that boron which was dissolved when the soil was digested in concentrated phosphoric acid. The acid insoluble boron was the difference between acid soluble and total boron. Whetstone considered the acid soluble boron a measure of the available soil boron. He observed that the acid soluble boron was directly related to the clay content of the various horizons of many soils. The total amount dependedon the parent material and extent of leaching. The kind of colloid was not significantly related to acid soluble boron content. Acid soluble boron increased regularly with increasing pH of the virgin soil. Soils derived from alluvium, limestone, shale and glacial drift were high in acid soluble boron. Those derived from igneous rock and unconsolidated sediment were low in acid soluble boron. Podzol, half bog, muck and red and yellow Podzolic soils were low in acid soluble boron with a higher percentage of acid insoluble boron. Alluvial, grey brown Podzolic, Prairie, Chestnut, Brown and Chernozem soils were high in boron most of which was acid soluble.

F. Recommended treatments for soils which produce boron deficient alfalfa.

Generally recommendations for fertilizer borate are 20 to 30 pounds per acre, broadcast on established stands of alfalfa (4, 10, 59). Banding with the seed may prove injurous especially if a nurse crop is planted with the seeding because grains are easily injured by relatively small amounts of borax (4). Barber (4) stated that the rate should never exceed 80 pounds per acre. Brown (10) stated that a 20 pound application was sufficient to prevent boron deficiency symptoms from occurring over a six year period while Barber (4) recommended repeating the application every two or three years. Both Brown and Barber made their recommendations for humid, temperate states. Workers in the southeastern states found that boron was leached much faster than in the North (17). Simmons (59) of Alabama recommended 20 to 30 pounds per acre of borax before planting followed by annual or biennial applications of 15 to 25 pounds per acre. In Florida, Winsor (70, 71, 73) recommended the use of less soluble sources of boron, such as colemanite, to prevent lose by leaching. In order to prevent boron deficiency from occurring during an extended hot dry period when the surface soil dries out, Hobbs (27) recommended that borax applications be made during the late Fall or early Spring in order to have the borax leach down to the deeper soil horizons.

ANALYTICAL METHODS

Plant material was dried at 70° to 80° C. and ground in a Wiley mill to pass through a 20 mesh screen. This material was again oven dried and placed in air tight bottles previous to weighing 2.5 grams into porcelain crucibles. The ground plant material was then ashed in a muffle furnace at 550° C. for four hours. The ash was taken up in 3 ml. of 6 normal HCl and brought to volume with distilled water in 25 ml. volumetric flasks. This latter was labeled solution A. When the plant material was to be analysed for bases and phosphorus, a 5 ml. aliquot of solution A was diluted to 50 ml. with distilled water and this latter labeled solution B. Boron determinations were made on solution A by the carmine method (25). This method is less sensitive than the curcumin procedure described below and consequently allows for determination of more concentrated solutions and reduces the possibility of significant amounts of contamination. Nevertheless boron free glassware (Corning 728) was used where possible and soft glass volumetric equipment (Kimble exax glass) was used when boron free glassware was not available. Phosphorus in solution B was run by the colorimetric procedure described by Kitson (32). Calcium, potassium and magnesium concentrations were determined in solution B with a Beckman D. U. flamephotometer with a photomultiplier attachment. An oxygen-hydrogen flame was used and the standards were made up to approximate the concentrations of calcium, magnesium, potassium, phosphorus, iron, aluminium, manganese and chloride in the unknown B solutions.

Boron was extracted from soils by a five minute boiling procedure described by Berger (7). Since the concentration of boron in this extract was too low for determination by the carmine technique, the more sensitive curcumin dye was used. The procedure for estimating boron using the curcumin dye was developed by Naftel (43) and modified by W. T. Dible at Wisconsin. The latter modification is not published and therefore the color development procedure is described here. A 1 ml. aliquot of the extract is transferred to a 250 ml. boron free beaker. To this is added 4 ml. of a 95% ethyl alcohol solution containing .04 gm. of curcumin and 5 gm. of oxalic acid per 100 ml. This is mixed thoroughly by rotating the beaker and evaporated on a water bath at $55^{\circ} \pm 3^{\circ}$ C. Dryness is insured by leaving the beaker on the bath for 15 minutes after it appears to be dry. The beaker is then cooled and the contents dissolved in 25 ml. of 95% ethyl alcohol. This is then filtered or centrifuged and the transmission determined at 540 millimicrons.

The pH of the soils was determined on a 2:1 water to soil suspension with a Beckman potentiometric pH meter. The texture was approximately determined by feel. The moisture equivalent of greenhouse soils was determined by centrifuging for thirty minutes at 1000 g.

A determination of the amount of CaO required to bring the pH of an Oshtemo loamy sand from 5.5 to 6.5 and 7.5 was made by titrating with a saturated solution of $Ca(OH)_2$. Twenty-five grams of this soil was weighed into a series of 100 ml. Erlenmeyer flasks and increments of saturated $Ca(OH)_2$ solution added. Each one was brought up to the same moisture level with distilled water, stoppered and placed on a shaker for 12 hours. The pH was then determined on these suspensions potentiometrically.

Ammoniacal nitrogen was determined on some of the plant material through the courtesy of Dr. Benne of the Michigan State College Agricultural Chemistry Department. The plant material was digested in sulfuric acid and the ammonia determined by a Kjeldahl procedure.

Because of the large number of analyses required, only single determinations were made on most samples. Duplicate analyses were made only to verify data that appeared to be out of line and to check on the precision of the procedures used.

FIELD SURVEY IN THE LOWER PENINSULA OF MICHIGAN

Methods

During the last week of July and continuing through the second week of August 1952, a trip was made through the lower peninsula of Michigan for the purpose of collecting alfalfa and surface soil samples for laboratory analyses and to locate sites for possible field plot experiments. Samples were procured from eighty-nine locations. At twenty-five of the locations both boron deficient samples and samples showing no apparent symptoms of boron deficiency were collected. Separate soil samples were procured from directly beneath the deficient and non-deficient plants. At eighteen other locations only boron deficient samples of alfalfa were collected along with soil samples. Alfalfa at forty-six locations showed no boron deficiency. Alfalfa and soil samples were collected from these meadows also.

The top inch or two of the plants were separated from the remainder and placed in separate paper bags. On boron deficient plants the top portions were the boron deficient fractions of the alfalfa. Henceforth these portions of the plants will be referred to as tops and bottoms.

The plant material was analysed for boron, calcium, potassium and magnesium. Ammoniacal nitrogen was determined on the plant material from the twenty-five locations at which both boron deficient and nondeficient samples were collected. These were the only samples that were at comparable stages of maturity. Nitrogen values were converted to protein percentages by multiplying by the factor 6.25.

Only pH and hot water extractableboron were determined on the soils. As many of the soils as possible were tentatively classified by the use of available soil maps and a few extra properties like texture, color, and topography. The legal descriptions of all the locations sampled during this survey and the locations used in field experiment II are listed in Appendix II.

Results and Discussion

A. Soil data.

In any attempt to correlate boron deficiency with soil test data, the following two factors must be considered:

1. Soil moisture content.

   When the soil was sampled plants may have been obtaining their nutrients from a lower horizon due to the dry condition of the surface layer.

2. Reliability of the extraction procedure.

> Although the five minute boiling technique is the fastest and most convenient method available for the extraction of boron from soils for analysis, it has certain serious defects which were discussed in the review of literature. Probably its most serious fault is the failure to give an accurate picture of the soil's capacity to supply boron during the entire growing period of the plant.

Boron extracted by the five minute boiling procedure will henceforth be referred to as extractable boron.

The averages for soil reaction and extractable boron are presented in Table 1. These data include only the results obtained from locations at which a sample was procured from beneath both boron deficient and nondeficient alfalfa plants. There is apparently no significant difference between the averages for extractable boron or the pH values. It should be noted here that only the surface soils were tested and that many soils had reactions well below that at which it is generally possible to obtain even fair alfalfa seedings (Appendix 1). This indicates that the subsoil horizons of these acid soils must have been more alkaline. Therefore the picture presented here probably does not tell the entire story.

Figure 1 illustrates diagramatically that there is apparently no consistent relationship between boron deficiency and the extractable boron content of the surface soil. The

TABLE 1

EXTRACTABLE BORON AND pH ON SOILS PROCURED FROM BENEATH BORON DEFICIENT AND NONDEFICIENT ALFALFA PLANTS[1/]

	pH	B p.p.m.
Deficient	6.3	.72
Nondeficient	6.1	.76
t test[2/]	N.S.	N.S.

1/ The pH values were converted to H ion concentration for averaging and performing t test. Data are averages from 24 samplings from locations where both deficient and non-deficient samples were procured.

2/ N. S. -- Not significant at the 5% level.

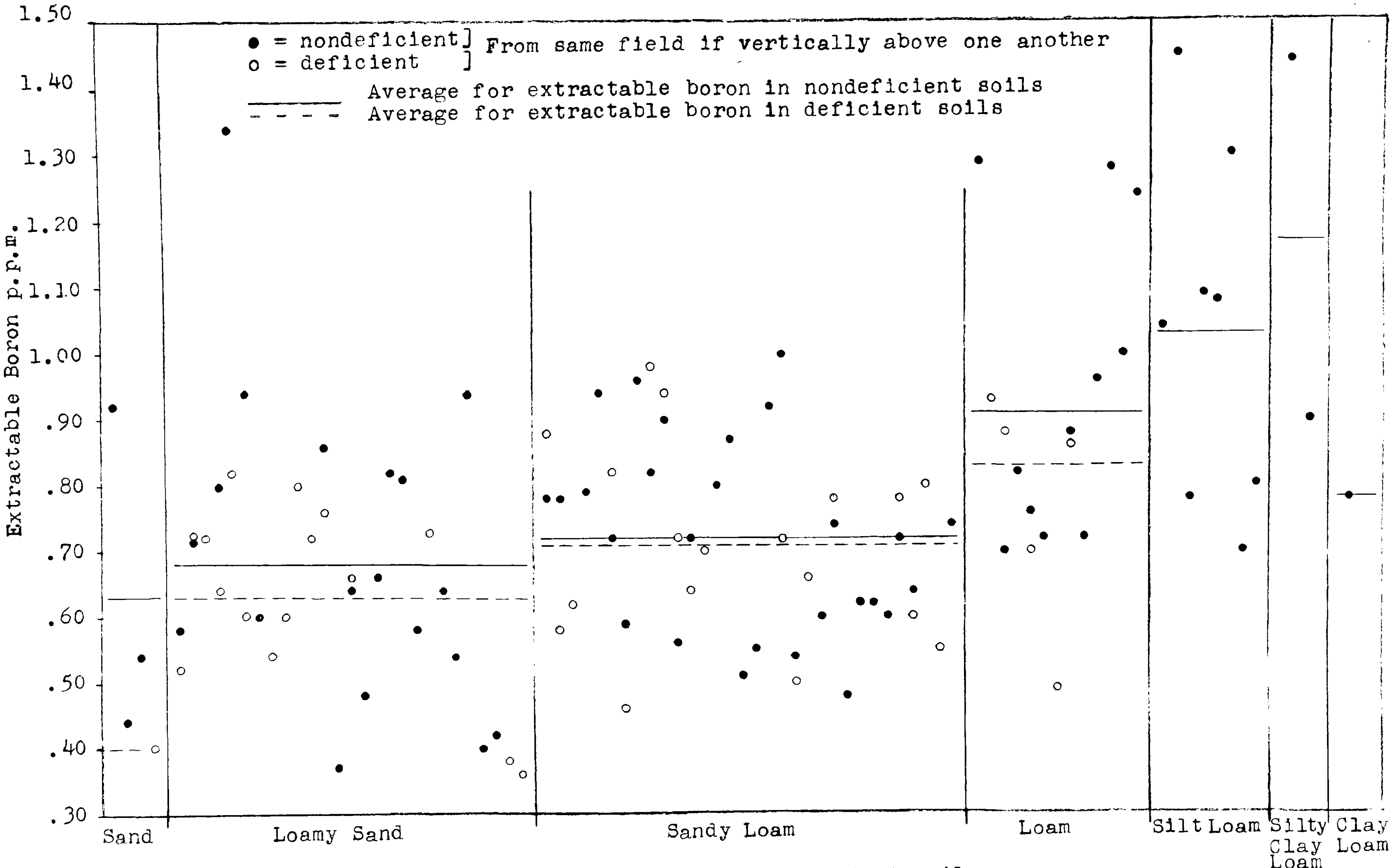

Fig. 1. Extractable boron in deficient and nondeficient soils from 89 locations.

data obtained do indicate, however, that the ratio of deficient to nondeficient soils becomes smaller as the texture shifts from coarse to fine. This is even more evident because an overt effort was made to locate finer textured soils on which boron deficient alfalfa was growing. The average value for extractable boron in each textural grouping increased as the texture shifted from coarse to fine.

As can be observed from an examination of the data in Appendix 1, the range of extractable boron in the soils was from .36 to 1.45 p.p.m., approximately a 4 fold difference. The range in the plant tops was from 3.6 to 43.5 p.p.m., a little more than a 12 fold difference.

B. Plant data.

In agreement with the findings of other investigators it was found that the apical portions of boron deficient plants were considerably lower in boron than were the more mature plant parts (Table 2). The reverse was found to be true for plants which appeared normal. The data in Table 2 show that the same relationships existed with respect to the bases calcium, potassium, and magnesium. Although the protein in the tops of deficient plants was significantly higher than in the more mature portions, the disparity was not as large as that in the nondeficient plants. As was pointed out earlier, boron deficiency causes a break down in the vascular tissue of

TABLE 2

BORON, CALCIUM, POTASSIUM, MAGNESIUM AND PROTEIN IN THE TOP AND BOTTOM PORTIONS OF BORON DEFICIENT AND NONDEFICIENT ALFALFA PLANTS

B Deficient Plants	B p.p.m.	Ca %	K %	Mg %	Protein %
top	9.6	1.81	1.21	0.28	21.23
bottom	13.3	1.91	1.36	0.36	18.78
t test	1 %	1 %	1 %	1 %	1 %
number of comparisons	43	42	42	42	25
Nondeficient Plants					
top	28.8	1.82	1.93	0.35	27.09
bottom	23.7	1.72	1.72	0.32	19.09
t test	1 %	1 %	1 %	1 %	1 %
number of comparisons	70	65	65	65	25

the plant. In all probability this causes a hindrance to the flow of nutrients to the meristematic tissue and may be the cause of the lower percentages of calcium, potassium, magnesium and protein in the apical portions of boron deficient alfalfa.

The data reported in Table 3 show that all the constituents determined were lower in the tops of boron deficient plants than they were in the tops of nondeficient plants. This, however, was not true of the bottom portion of the plants. Only the boron concentration was significantly lower in the more mature portion of deficient alfalfa than in the same fraction of nondeficient alfalfa.

The results compiled in Tables 2 and 3 are to be expected when one considers the fact that only the apical portion of alfalfa actually showed deficiency. Apparently boron in the more mature parts of the plants that showed boron deficiency was sufficient to allow normal functioning of the metabolic processes. It should be noted that in trends cited above, calcium showed the least amount of constancy while the trends for potassium, magnesium and protein were very consistant (Appendix 1). This may lead one to suspect that the reason for the decrease of these constituents in meristematic tissue of boron deficient plants may not be due solely to malfunctioning of the vascular tissue but also to a functional relationship between boron and potassium, magnesium and nitrogen.

TABLE 3

BORON, CALCIUM, POTASSIUM, MAGNESIUM AND PROTEIN IN THE TOP AND BOTTOM PORTIONS OF BORON DEFICIENT AND NONDEFICIENT ALFALFA PLANTS1/

Top portions	B p.p.m.	Ca %	K %	Mg %	Protein %
B deficient	10.0	1.80	1.31	0.29	21.23
Nondeficient	25.7	1.97	1.76	0.36	27.09
t test	1 %	5 %	1 %	1 %	1 %
Number of comparisons	25	24	24	24	25
Bottom portions					
B deficient	13.7	1.87	1.43	.35	18.78
Nondeficient	20.9	1.92	1.49	.32	19.09
t test2/	1 %	N.S.	N.S.	N.S.	N.S.
Number of comparisons	25	23	23	23	24

1/ Deficient and nondeficient plants were selected from the same location.

2/ N.S. -- Not significant at the 5% level

One might conclude by an examination of Tables 2 and 3 that 10 p.p.m. would be a good figure to quote for a critical level for boron in the apical portions. It must be remembered, however, that this figure was derived from averages and that individual plants varied widely from the mean. As shown in Table 4 the lower limit of boron in nondeficient tops was well below the upper limit attained for the boron content of deficient tops. This variation is to be expected when one considers the rather wide range of critical levels found for boron in alfalfa by other investigators. As pointed out in the literature review, all of the critical levels published by other investigators were 20 p.p.m. or lower. The highest value obtained for the more mature portion of the plants in Table 4 was 20.8. This value would be slightly lower if the entire plant had been analysed for boron and is in good agreement with the results of other workers.

The boron values obtained on boron deficient alfalfa from locations 12 and 44 were not considered in Table 4 because of the unusually high boron levels in these plants (Appendix 1). Typical symptoms of boron deficiency were seen at these locations at sampling time. The following summer they were again observed at location 44, but not at location 12. However, there was no blossoming and the buds appeared to be dying at location 12. This condition also suggests boron deficiency. Furthermore, the analytical data tend to bear out the fact

TABLE 4

THE RANGE OF BORON LEVELS IN THE TOP AND BOTTOM PORTIONS OF BORON DEFICIENT AND NONDEFICIENT ALFALFA

	Lower Limit B p.p.m.	Upper Limit B p.p.m.
Boron deficient tops[1/]	3.6	17.7
Boron deficient bottoms[1/]	8.5	20.8
Nondeficient tops	13.5	43.5
Nondeficient bottoms	11.7	38.2

1/ Locations 12 and 44 were not included in these data.

that these plants were boron deficient (Appendix 1). The apical portions of deficient plants were lower in boron, bases, and percent protein than the more mature parts. The possibility exists that some other factor may cause typical symptoms of boron deficiency by interfering with the flow of nutrients to the apical meristem. Perhaps a sucking insect could do this.

Due to the emphasis in past years on Ca:B ratios, calculations were made on the data from this study in order to present these ratios in Table 5. It is obvious that this ratio is anything but a constant, even in the nondeficient plants. Almost without exception the tops of deficient plants had a higher calcium : boron ratio than did the bottom portions. The reverse situation was found in the nondeficient plants. This indicates that the boron content of plants was more influential in producing boron deficiency symptoms than was the calcium level. A decrease of calcium, which was noted previously, in the boron deficient tops, would cause a lower ratio while the lower boron content causes the ratio to rise.

In attempting to determine a critical calcium : boron level in plants, if such is possible consideration must be given to the fact that the critical level has probably been exceeded in most of the plants sampled. Therefore the boron deficient sample with the smallest ratio should be judged the one most closely approaching a critical level. In this study the deficient sample procured at location 41 was found to have

TABLE 5

THE Ca/B RATIOS[1/] IN THE TOP AND BOTTOM PORTIONS OF BORON DEFICIENT AND NONDEFICIENT ALFALFA PLANTS

Location	Deficient Plants		Nondeficient Plants	
	Top	Bottom	Top	Bottom
1	1194	1588	803	862
2	1898	1466	677	809
3			393	498
4	1011	773	767	942
5	2303	2042		
6	1958	1369	1356	1362
7	2243	1185	713	952
9			260	326
10			548	599
11			532	704
12	672	603	563	619
13	2306	2092	966	1129
14	2074	1683	824	1053
15			802	1031
16			454	
17	3861	1765	940	1029
18			375	498
19			572	871

1/Ratios were calculated on a weight basis.

TABLE 5--Continued

Location	Deficient Plants		Nondeficient Plants	
	Top	Bottom	Top	Bottom
20	3083	2064	609	811
21			366	508
22			511	619
23			398	648
24			476	771
25	1758	1368	422	659
26	1557	1387		
27			596	848
28	4080	2024	646	
29	3760	2298	1539	
30	2827	1592	764	1127
31	3482	1743		
32	4000	1835		
33	1851	1278	771	1081
34	1761	1149	937	1202
35		1487	774	914
36	1503	1049		
37	2820	2560		
38	2857	1728		
39	2770	1268		

TABLE 5--*Continued*

Location	Deficient Plants		Nondeficient Plants	
	Top	Bottom	Top	Bottom
40	2535	1636	855	893
41	902	838		
42			555	652
43			856	817
44	408	528	417	461
45			572	523
46			680	788
47			476	569
48			782	582
49			435	464
50			379	457
51	982	856	613	673
52			516	594
53			592	
54			830	703
55			741	784
56			587	721
57	3225	2163		
58	2973	2630	2015	1341
59	1759	1172	591	684

TABLE 5--Continued

Location	Deficient Plants		Nondeficient Plants	
	Top	Bottom	Top	Bottom
60	4220	1413		
61			407	411
62			450	564
63			402	485
64			664	781
65	3370	1900	796	1050
66			707	563
67			471	508
68			713	833
69			730	812
70			666	632
71			678	699
72	1309	969		
73			1091	1357
74			556	592
75			869	1112
76			675	771
77			943	886
78	3015	2095	629	748
79			636	591

TABLE 5--Continued

Location	Deficient Plants		Nondeficient Plants	
	Top	Bottom	Top	Bottom
80			810	
81			420	425
82			634	629
83	1967	1982		
84	4385	2160	950	1917
85	2235	2056		
86	1187	1149		
87	2354	1870		
88	1292	1173		
89	2932	2402		

the smallest calcium : boron ratio in the apical portion of the plant if the data from locations 12 and 44 are not considered (Table 5). This value was 902. It was concluded in the literature review by comparing the data of other investigators that this critical level should be about 700. Considering the fact that this latter value was calculated from analyses on the whole plant, the two figures are not greatly different. It should be noted, however, that in nine cases, the plants showing no apparent symptoms of boron deficiency had calcium :boron ratios higher than 902 in the apical portions (Table 5). This indicates that other factors besides the calcium level affect the boron requirements of alfalfa.

Conclusions

Because of the inadequacies of the five minute boiling procedure for extracting boron from soils and the fact that plants may not obtain their nutrients from the surface horizon when the soil is dry, it can not be expected that boron determinations on surface soils would be a very reliable means of predicting whether boron deficiency will appear on alfalfa. The data of this study tend to bear this out. The reaction of the surface soil was not significantly lower on deficient soils than on nondeficient soils. Generally more boron deficiency was noted on the coarse than on the fine textured soils. There was also a larger amount of extractable boron in the finer textured soils.

The apical portions of boron deficient alfalfa were lower in boron, calcium, potassium and magnesium than were the more mature parts. The reverse was true of nondeficient plants. The tops of deficient plants were found to also be lower in boron, calcium, potassium, magnesium and protein than were the tops of nondeficient plants. When the analytical data from the more mature portions of alfalfa are compared, it is found that the boron was the only constituent that was significantly lower in deficient plants. These results should be expected when it is realized that only the apical portions of boron deficient alfalfa actually show the symptoms. It is probable that the lower base and protein content is due to a combination of a vascular breakdown in the plant and a direct or indirect functional relationship between boron and these other nutrients.

It must be concluded that no one critical value for boron or for a calcium : boron ratio in alfalfa can be quoted because this level may vary widely with different environmental conditions. However, the data from this study and from others cited in the literature review indicate that the boron content of alfalfa is 20 p.p.m. or less, the soil probably has a low boron supplying capacity and boron deficiency is likely, especially during an extended hot, dry period or when the pH of an acid soil is raised by lime applications.

Analysis of the apical portions of the plants is probably more useful for verifying visual symptoms of boron deficiency, especially if the boron content of the top portions of the plant is compared with the concentration of this element in the more mature fractions.

FIELD EXPERIMENT I

Methods

During April of 1953 an attempt was made to place experimental plots at as many as possible of the locations that had been sampled the previous summer. The treatments consisted of 0-20-20 applied at the rate of 300 pounds per acre and 0-20-20 applied at the same rate plus 40 pounds of borax. The plots were 20 by 100 feet placed adjacent to each other. Because of the large number of locations the treatments were not replicated. The fertilizer was broadcast as a top dressing on established stands of alfalfa.

Although plots were located at 65 locations, first crop yields were obtained at only 29 locations and second crop yields at 39. Observations were recorded at 52 locations during the period when the second crop was about ready for harvest. The remaining 13 locations on which observations were not recorded had been either plowed, pastured, cut for hay, or were in such a state of degeneration because of dry soil conditions, extensive insect damage, or wilt that no observation could intelligently be made.

Results and Discussion

The yield data from this experiment are summarized in Table 6 and the bulk data from the second crop are presented in Table 7. It is obvious that yields were not increased by borax applications on the first crop of alfalfa where no deficiency had been noted the year before. However significant increments in growth were obtained on the second crop from borax applications at sites where there had been a history of boron deficiency the previous year and where boron deficiency was noticable at harvest time (Table 6). These responses were not large; the former being an 8 percent and the latter a 12 percent increase in growth.

Symptoms of boron deficiency were not observed on the first crop of alfalfa at any of the plot sites nor on any alfalfa meadows encountered when traveling to the various locations. On the second crop, however, symptoms were much more prevalent at the plot sites and throughout the state. This is a common trend in areas where there is usually sufficient moisture for the first crop but not for the second crop. Boron deficiency symptoms were not observed on any of the plots that were treated with fertilizer borate.

As is illustrated in Table 7, most of the boron deficiency observed was at sites that had previously shown symptoms of boron deficiency. It was pointed out in the previous section

TABLE 6

AFFECT OF BORON FERTILIZATION ON YIELDS

A. Yield Averages From Locations that Showed Boron Deficiency the Previous Year

First Crop		Second Crop	
Treatment	Yield in Lbs.4/	Treatment	Yield in Lbs.
Check1/	93.2	Check	36.1
Borax2/	91.5	Borax	38.9
t test3/	N.S.	t test	5 %
Number of Comparisons	25	Number of Comparisons	26

B. Yield Averages From Locations that Showed No Boron Deficiency the Previous Year

First Crop		Second Crop	
Treatment	Yield in lbs.	Treatment	Yield in lbs.
Check	52.4	Check	64.1
Borax	56.6	Borax	68.7
t test	N. S.	t test	N.S.
Number of Comparisons	4	Number of Comparisons	13

C. Yield Averages From Locations Containing Plots That Showed Boron Deficiency on or Near Them During the Second Harvest5/

	Second Crop	
	Treatment	Yield in Lbs.
	Check	41.4
	Borax	46.5
	t test	1 %
	Number of Comparisons	22

1/ 300 pounds of 0-20-20 per acre.
2/ 300 pounds of 0-20-20 plus 40 pounds of borax per acre.
3/ N.S. stands for not significant at the 5% level.
4/ Green weight of hay harvested from 0.576% of an acre.
5/ Location 78 was not included in these averages because deficiency was observed some distance from plots.

that deficient plants collected from these locations all had boron contents of 20 p.p.m. or less. However, in six instances boron deficiency was found where it was not seen the previous year. At three of these sites (55, 66, 80) the boron concentration was around 20 p.p.m. in plants sampled the previous year but at the other three sites (48, 53, 54) this concentration was appreciably higher (Appendix 1). That plants may have a high content of boron one year and be deficient the next should not be surprising. A plant may obtain most of its nutrients from the surface layer when the moisture level is high but should the surface become dry it is probable that the subsoil will become the main source of plant nutrients. Another point which should be brought out is that a small sample taken from a field may not be representative of all parts of the field. This is illustrated by the data presented in Appendix 1. At many of the locations where boron deficiency was observed, nondeficient samples were also procured and although the boron content of these plants was often lower than average, some of them contained more than 30 p.p.m. of boron. It was also pointed out in the previous section that sometimes apparent symptoms of boron deficiency appear in plants that contain a relatively high concentration of boron (Appendix 1, locations 12 and 44). Apparent boron deficiency symptoms again occurred at location 44 and were eliminated by a borax application (Table 7).

TABLE 7

VISUAL OBSERVATIONS AND YIELDS OF THE SECOND CROP OF ALFALFA OF FIELD EXPERIMENT I

Location	Deficiency Serious on Check Plot	Deficiency Weak on Check Plot	Deficiency on Field but not On Check Plot	No deficiency Observed on Field	Green wts.2/ in Pounds on Second Crop	
					Check	Borax
1			x		45.9	47.8
2		x			60.0	61.6
3*				x	54.7	47.9
4				x	45.2	50.5
5		x			36.2	61.4
7		x			36.4	39.4
12				x	24.3	25.3
21*				x		
22*				x		
23*				x		
25				x	24.2	27.0
28			x		50.8	60.2
29				x	29.7	33.8
30				x	73.0	88.6
31				x	26.9	25.7
32				x		
34		x				
35				x1/		
36	x				9.3	10.6
37	x				29.0	29.8
38		x			46.0	48.0
39		x			44.2	41.2
40				x	25.9	18.2
41		x			20.7	18.8
44		x			17.1	18.5
45*				x	45.0	54.6
47*				x	47.4	49.8
48*		x			27.8	28.8

1/ Plants in early vegetative stage.

2/ Green weight of hay harvested from 0.576 % of an acre.
* No deficiency observed the previous year.

TABLE 7-- Continued

Location	Deficiency Serious on Check Plot	Deficiency Weak on Check Plot	Deficiency on Field but not On Check Plot	No deficiency Observed on Field	Green wts.[2] in Pounds on Second Crop	
					Check	Borax
52*				x		
53*		x				
54*		x			56.2	75.7
55*		x			108.7	113.0
57		x			28.4	23.6
58	x				24.8	26.9
59		x			36.2	41.1
63*				x	132.7	128.2
65				x1/		
66*		x			53.8	74.2
69*				x1/		
71*				x	46.4	44.5
72				x1/		
74*				x	73.3	81.6
75*				x	71.6	65.0
76*				x1/		
78			x3/		87.3	87.0
79*				x	24.5	24.5
80*		x			91.5	104.7
83				x	30.6	31.0
84			x			
86			x		18.7	18.0
87		x			47.3	60.0
88		x			20.4	19.1

1/ Plants in early vegetative stage.

2/ Green weight of hay harvested from 0.576% of an acre.

3/ Deficient plants observed at same place sampled the previous year but plots were some distance away.

* No deficiency observed the previous year.

It is possible to observe from an examination of Table 7 that in twelve cases, locations which had grown boron deficient plants the previous year did not show it again when observations were made at the plot sites. This is to be expected with seasonal climatic variations in different areas, but it should also be noted that in three of these cases the plants were in an early vegetative state at the time observations were recorded. Generally, boron deficiency appears on more mature alfalfa.

It was pointed out in the review of literature section that many investigators find it difficult to demonstrate a vegetative yield response to borax applications. One reason for this apparent lack of response may be that the deficiency occurs predominently during extended,hot, dry periods when even the growth of nondeficient plants is slowed by a limited moisture supply. Another factor to be considered is that the deficiency often appears late in the life cycle of the plant when vegetative growth is slower. Defects in the experimental setup might also be a cause for not being able to detect small growth responses. Three such defects were apparent in this experiment only after the work was completed. These are listed below.

1. Because the treatments were made in the spring when it was impossible to observe the deficient areas, many of the plots did not cross deficient portions of the fields.

2. The plant populations were not always uniform on both plots. In the spring the plant population is not always apparent.

3. Insect damage was often so severe as to practically mask any response to borax especially on excessively drained, coarse textured soils. This was particularly true of the three sites that showed serious boron deficiency (Table 7).

Conclusions

Small vegetative responses were obtained on the second crop where there was a history of boron deficiency and where deficiency symptoms were observed at harvest time. No responses were procured on the first crop or where there was no deficiency noted the previous year.

The data confirm the fact that boron deficiency symptoms are likely to occur in alfalfa, especially during extended periods of hot, dry weather, when the boron content of the apical portions of the plants is below 20 p.p.m. Some exceptions occur, however, when one predicts that no boron deficiency will occur when the boron content of alfalfa sampled the previous year is considerably higher than 20 p.p.m. Three explanations are advanced for this dilemma.

1. A sample may not be representative of all parts of a field.

2. Plants may obtain most of their nutrients from the surface horizon when this layer is moist and have a relatively

high content of boron. When the surface becomes dry, the subsoil horizons may be the main source of plant nutrients and deficiency may occur.

3. In exceptional cases boron deficiency may occur when plants have a relatively high boron content.

FIELD EXPERIMENT II

Methods

During the last two weeks of August, 1953, fourteen field plot experiments were layed out. The treatments were made at this time in order to overcome the already mentioned objectional features of making treatments in the spring. In most cases top dressings were made directly on boron deficient portions of established stands of uniformly pure alfalfa. Yield data were procured from only ten of the fourteen sites. Of the ten, seven showed boron deficiency at the time of application (Table 8) and it appeared probable that the other three, which were new seedings, might produce boron deficient alfalfa from consideration of the coarse texture and high pH of the soils. The sites were selected at widely separated locations throughout the lower peninsula of Michigan in order to take advantage of any dry weather that might occur in one section or another previous to harvest time.

There were two treatments broadcast as a top dressing and these consisted of 0-20-20 applied at the rate of 600 pounds per acre and the same amount of 0-20-20 plus 30 pounds of borax. The treatments were randomized in four blocks at each location but the randomization was modified at times by consideration of the slope. At locations where a block was placed lengthwise to the perpendicular of a slope the check plot was always placed at the top in order to prevent the possibility of runnoff contaminating this treatment with the borax. Each plot was 20 feet long by 6 feet wide and there were 3 feet between the two treatments in each block. A strip, 32 inches by 20 feet, was cut through the center of each treatment in order to obtain the yield data. Only the second crop was cut for yields; the first being cut by the farmer cooperator during his regular haying operation. Samples of the top one or two inches of alfalfa plants were collected along with surface soil samples from each treatment at harvest time. The plant samples were analysed for boron while pH and boron determinations were made on the soils. The soils were tentatively classified from soil maps and consideration of such properties as pH,texture, and topography.

An attempt was made to control insect damage by spraying the plots and a small area surrounding them with insecticides. Lindane was applied during the last week in May to control spittle bug and chlorodane was sprayed during the third week in July to control the potato leaf hopper and grasshoppers.

Results and Discussion

All of the data from this experiment are condensed in Table 8. In no case was boron deficiency noted on the treatments

TABLE 8

YIELD DATA[1], ANALYTICAL DATA[1], AND VISUAL OBSERVATIONS ON THE PLANTS AND SOILS OF FIELD EXPERIMENT II

Location	Soil Type	Treatment	Yield[5] Lbs.	B in Plant tops p.p.m.	B in Soil p.p.m.	Soil[6] pH	B Deficiency[7] Symptoms: At Application Time	B Deficiency[7] Symptoms: At Harvest Time
I	Macomb	check[2]	7.75	38.5	1.00	6.9		
	fine sandy	borax[3]	8.93	48.0	1.80	7.0		
	loam	significance[4]	N.S.	N.S.	5 %	N.S.		
III	Montcalm	check	8.15	29.4	0.57	5.8	x	
	loamy sand	borax	7.00	61.5	0.78	6.0		
		significance	N.S.	1 %	N.S.	N.S.		
IV	Grayling	check	5.23	16.4	0.59	6.7	x	x
	sand	borax	6.40	59.0	0.72	6.6		
		significance	1 %	1 %	1 %	N.S.		
VI	Kalkaska	check	13.95	19.4	0.43	6.5	x	
	loamy sand	borax	13.63	50.5	0.55	6.6		
		significance	N.S.	1 %	N.S.	N.S.		
VIII	Coloma	check	11.55	13.3	0.65	7.3		x
	loamy sand	borax	12.30	31.8	1.19	7.3		
		significance	N. S.	5 %	N.S.	N.S.		
X	Coloma	check	6.88	19.4	0.49	7.2		
	loamy sand	borax	6.63	76.7	0.60	7.2		
		significance	N.S.	5 %	1 %	N.S.		
XI	Coloma	check	11.38	11.7	0.44	6.8	x	
	loamy sand	borax	11.03	51.5	0.53	6.8		
		significance	N.S.	5 %	5 %	N.S.		

1/ All data are the averages of four replications.
2/ 0-20-20 applied at the rate of 600 pounds per acre.
3/ Same as check plus borax fertilizer applied at the rate of 30 pounds per acre.
4/ Analysis of variance. N.S. stands for not significant at the 5% level.

TABLE 8--Continued

Location	Soil Type	Treatment	Yield 5/ Lbs.	B in Plant tops p.p.m.	B in Soil p.p.m.	Soil 6/ pH	B Deficiency 7/ Symptoms At Application Time	B Deficiency 7/ Symptoms At Harvest Time
XII	Oshtemo loamy sand	check	8.20	21.3	0.84	7.2	x	
		borax	7.70	44.3	1.07	7.2		
		significance	N. S.	1 %	1 %	N.S.		
XIII	Fox sandy loam	check	14.20	7.0	0.75	7.2	x	x
		borax	14.88	24.0	0.92	7.2		
		significance	N.S.	1 %	N.S.	N.S.		
XIV	Fox sandy loam	check	11.45	10.6	0.46	7.0	x	x
		borax	12.05	27.1	0.55	7.0		
		significance	N.S.	1 %	N.S.	N.S.		

5/ Green weight of alfalfa cut from a strip of dimensions 32 inches by 20 feet.
6/ pH was converted to hydrogen ion concentration for the purpose of averaging and making the analysis of variance calculations.
7/ The x indicates that boron deficiency symptoms were seen at the time indicated.

that received an application of borax fertilizer at the rate of 30 pounds per acre. In only one case out of the ten was there a vegetative response noted (Table 8, location IV). In this instance a 22 percent increase in yield was obtained and the response was very apparent by visual observation. The other three plot sites where deficiency was seen in the check plots at harvest time only showed a tendency toward a yield response (Table 8).

The boron applications increased the boron content of the plant material and also increased the extractable boron in the soil. However, the relationship between applied and extractable boron was not too consistent as is indicated by the number of cases that showed no significance at the five percent level.

The data presented in Table 8 again illustrate the difficulty involved in establishing a critical level for boron in alfalfa. The plant tops sampled from the check plots at location IV had a higher boron content than plant tops from the other three locations which showed boron deficiency even though the deficiency was much more severe at location IV. More important is the fact that the plant tops from location XI, which showed no symptoms of boron deficiency at harvest time, were actually lower in boron than the plant tops sampled at location IV.

In Figure 2 the extractable boron values from the check treatments are plotted against the boron content of the apical portions of the plants sampled from these plots. The very wide scatter of points again illustrates the impracticality of attempting to use soil test data for the purpose of predicting the boron concentration in alfalfa or, by inference, whether or not boron deficiency symptoms will appear.

The fact that boron deficiency may appear during one season and not the next, is to be noted in Table 8. This was also observed to be the case in the preceding experiment.

Conclusions

The main conclusion that can be drawn from this experiment is that it is possible to demonstrate a vegetative response to borax applications on deficient soils with careful experimental procedures. The data once again illustrate the impracticality of the use of soil test data for predicting whether or not boron deficiency will appear. They also demonstrate that it is not possible to find one critical level for boron in alfalfa that will be the same for all environmental conditions.

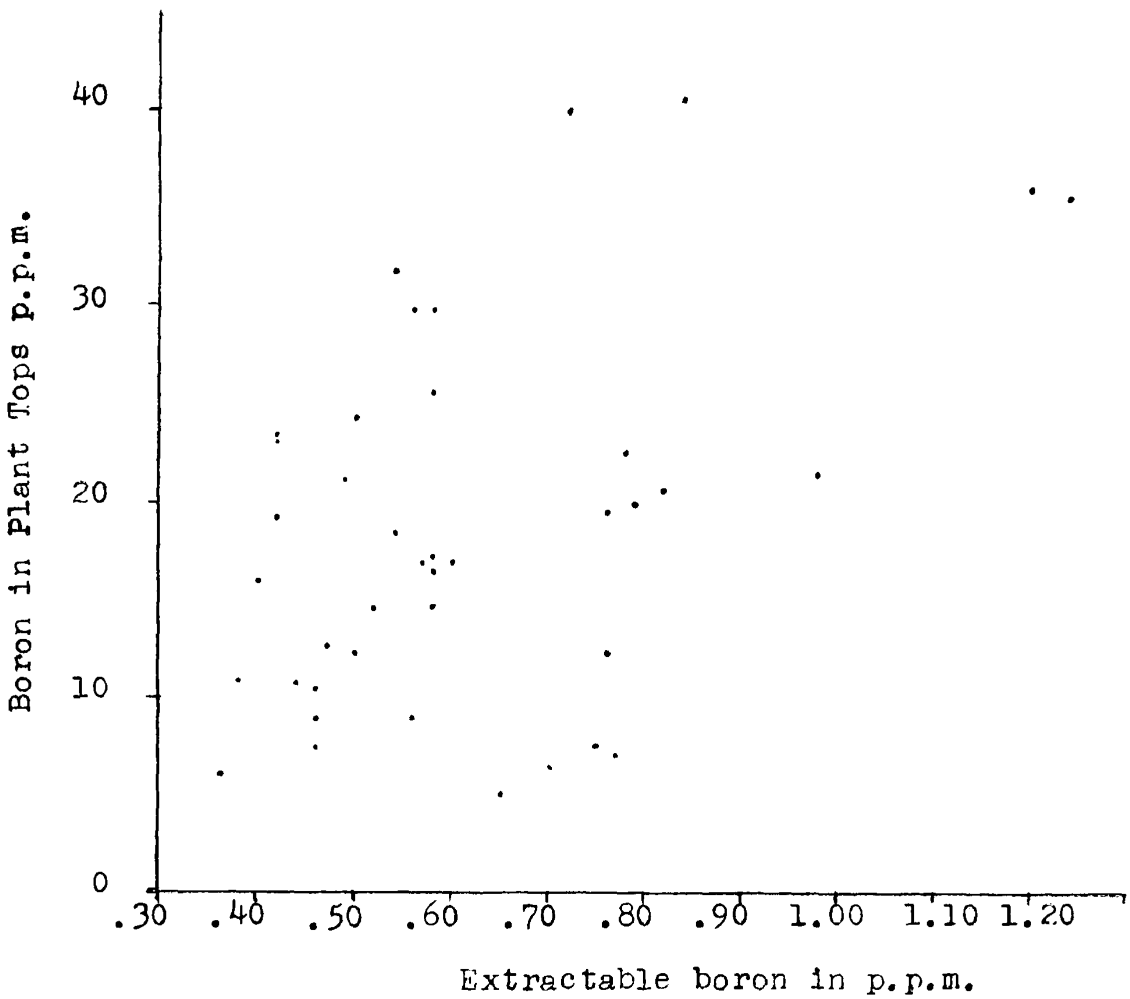

Fig. 2. Comparison of extractable boron from check plots of Field Experiment II with the boron content of apical portions of alfalfa harvested from the same plots.

GREENHOUSE EXPERIMENTS

Introduction

The use of the greenhouse for experimental work in soils has several advantages. Although field conditions can seldom be strictly duplicated in the greenhouse, environmental conditions can be controlled with more facility and work can be hastened by carrying on experiments throughout all four seasons. For these reasons, it was decided to attempt to investigate a few factors related to boron deficiency in alfalfa in the greenhouse. In the main, these factors are as follows.

1. The determination of a critical level for boron in alfalfa and perhaps in the soil in which it was grown.
2. To demonstrate, if possible, a response to borax applications before a deficiency becomes apparent.
3. To determine the effect of pH or the calcium level in the soil on boron availability and requirement.

Methods

Three soils were selected for the experiments. Two of these soils were a Wisner clay loam and a Thomas sandy loam;

both of which are calcareous at the surface. The third was an Oshtemo loamy sand with a pH of 5.5. The Thomas is a lake bog soil which is drained and used extensively for sugar beet production in Michigan. It contains about eight percent organic matter. Sugar beets grown on the Wisner and Thomas soils are often affected with heart rot, a symptom of boron deficiency.

The soils were placed in one gallon glazed pots without drainage openings. These pots were tared by placing coarse, acid washed gravel at the bottom of the pots in varying amounts. Because of the differences in the volume weights of the three soils, the pots held 3.5 kilograms of the Thomas, 4.0 kilograms of the Wisner and 4.5 kilograms of the Oshtemo soils. The lime, fertilizer and boron treatments were made on the premise that each pot held 4.4092×10^{-4} percent of an acre furrow slice. Stated another way, it was assumed that an acre furrow slice of the Thomas soil weighed 1,750,000 pounds; Wisner 2,000,000 pounds and Oshtemo 2,250,000 pounds.

The treatments on the Thomas and Wisner soils consisted of borax added at the rates of zero, twenty and forty pounds per acre furrow slice. On these two soils the treatments were replicated eight times.

The same boron treatments were made on the Oshtemo soil but at three different pH levels; 5.5, 6.5, and 7.5. These latter treatments were replicated four times.

A blanket fertilizer application was made on all three soils. This application was equivalent to 1000 pounds of

0-20-20, 100 pounds of $MgSO_4$ and 50 pounds of $MnSO_4$ per acre furrow slice. The phosphorus and potassium sources were KH_2PO_4 and K_2SO_4. Both the borax and the fertilizer applications were applied in solution form to the air dry soils in the pots.

The Oshtemo soil was brought up to the required pH levels by thoroughly mixing weighed amounts of CaO with the air dry soil and immediately bringing the soil up to slightly higher than the moisture equivalent point with a solution containing the fertilizer and borax treatments. The technique for determining the lime requirements for the two pH levels was discussed in the section on analytical methods. This was found to be 0.903 m.e. of Ca per 100 gms. of soil to attain a pH of 6.5 and 1.653 m.e. per 100 gms. to bring the pH to 7.5. This would amount to 1,027 and 1,693 pounds of $CaCO_3$ equivalent per acre furrow slice.

Seedings were started on the Thomas and Wisner experiments on March 10 of 1952 and on the Oshtemo experiment on May 17 of 1952. Certified Ranger alfalfa seed was used for all three experiments. Ten seeds were planted in each pot and thinned to four plants per pot when the seedings were about two weeks old. The Thomas experiment had to be reseeded after the tenth crop because the plants had been allowed to desiccate. Actually eighteen crops of alfalfa were harvested from the Wisner and seventeen crops from the Thomas soil before these experiments were terminated on the tenth of January of 1955.

Seven crops of alfalfa were harvested from the Oshtemo soil, that had been limed with CaO, before the experiment was terminated on July 8 of 1953. It was not possible to include the yields from the unlimed Oshtemo soil (pH 5.5) because of the extremely feeble growth of alfalfa at this pH level. However, on March 30 of 1954 a solution of NH_4NO_3 was applied to the unlimed Oshtemo soil which was not discarded with the rest of the experiment. This was applied at the rate of 60 pounds of N per acre furrow slice and an alfalfa harvest was made from this acid soil on May 4 of 1954. These treatments were then discarded.

Watering was done with distilled water. The soils were brought up to the moisture equivalent point by weighing when the moisture contents became too variable.

Applications of 0-20-20 in solution were made periodically at the rate of 1000 pounds per acre furrow slice. The following is a list of the dates on which these extra fertilizer applications were made on the various soils.

Oshtemo (Initial pH 5.5)	Oshtemo (Initial pH 6.5 and 7.5)	Thomas	Wisner
1-5-54	8-13-52	5-19-52	5-19-52
	10-17-52	7-21-52	7-23-52
	4-8-53	10-17-52	10-31-52
		1- 7-54	1- 7-54
		9-16-54	9-17-54

Soil samples were taken from all pots at the time the experiments were discarded. This sampling was done with a cork borer of 3/8 inch diameter; five borings being taken in each pot. Extractable boron and pH were determined on all of these samples.

Because of the large number of plant samples involved in these experiments, not all of them were selected for weighing and chemical analyses. The data from those selected are presented in the following section. All the data were analysed statistically by means of the analysis of variance procedure.

Results and Discussion

A. The Wisner and Thomas soil experiments.

No boron deficiency symptoms appeared on any of the alfalfa grown on the Thomas soil but scattered symptoms appeared on some of the check treatments of the eleventh crop of alfalfa grown on the Wisner soil. These symptoms continued to appear intermittently until the end of the experiments but were never very severe. Actually only one or two out of about twenty shoots showed deficiency symptoms and quite often a check pot showed no deficiency at all on one cutting and then did show it on the next. The apical portions of a few shoots that showed boron deficiency were collected from three of the check treatments from the eighteenth crop of alfalfa on the Wisner soil. At the same time samples were taken of apparently non-deficient shoots from these same treatments. These were analysed for boron by the curcumin procedure because of the small amount of plant material available. These data are presented in Table 9. Apparently the critical level for boron was between 9 and 10 p.p.m. in alfalfa grown on this soil and under the particular environmental conditions that existed when the crop was grown.

The tendency for the top portions of boron deficient plants to be lower in boron than were the more mature plant parts was noted on the sixteenth crop on the Wisner soil (Table 11). This tendency was observed only on the check treatments and was reversed on the borax treatments. It should also be pointed out that the top portions of the fifteenth crop of alfalfa grown on the Thomas soil were higher in boron than were the more mature parts on all treatments (Table 10). As mentioned previously, no deficiency was seen on any of the seventeen crops of alfalfa grown on the Thomas soil.

The question of whether alfalfa will respond to boron before deficiency actually appears is apparently answered by the data from these two experiments. The data in Tables 10, 11, and 12 show that significant differences in yield were not obtained in spite of the wide variations in boron contents above deficient levels. More important is the fact that no

TABLE 9

THE BORON CONTENT OF BORON DEFICIENT AND NONDEFICIENT APICAL PORTIONS OF ALFALFA SAMPLED FROM THE 18TH CROP GROWN ON THE WISNER SOIL

Replication	Borax Treatment Lbs/Acre	B in Tops Showing B Deficiency p.p.m.	B in Tops Showing No B Deficiency p.p.m.
1	0	8.75	19.42
3	0	8.63	10.02
7	0	6.78	18.17

significant difference was found in yield due to borax applications when weak symptoms of boron deficiency were seen (Table 11, 16th crop; Table 12, 18th crop). It also appears that variations in the boron content of alfalfa, above deficient levels were not accompanied by differences in the percentages of calcium, potassium, magnesium or phosphorus in the plants (Table 12). Toxic levels of boron were not attained.

The boron content of the alfalfa grown on the Wisner clay loam was relatively high in the first and second crops but fell off considerably in the eighteenth crop (Tables 11 and 12). In contrast, the boron content of the plants grown on the Thomas sandy loam was lower in the first crop and did not decrease as much by the time the seventeenth crop was harvested (Tables 10 and 12). Perhaps the reason boron availability remained quite constant on the Thomas soil was that boron was released as the soil organic matter was decomposed. If it can be assumed that the boron content of the plant is a good indication of availability of boron in the soil, then these data indicate that organic matter fixed boron more readily than did clay at a high pH level. This latter statement is made for the following additional reasons:

1. Probably the main active constituent of the Thomas soil is organic matter (8% organic matter) and that of the Wisner is clay.
2. According to the plant analysis data more of the added borax remained available to the early crops

TABLE 10

THE YIELD AND BORON CONTENT OF ALFALFA GROWN ON THE THOMAS SANDY LOAM EXPERIMENT

	Borax Treatment Lbs./Acre	Dry Matter[1/] Yields gms.	B p.p.m.[2/]			
			reps. 1 thru 4		reps. 5 thru 8	
1st crop	0	3.99	30.4		27.4	
	20	4.16	52.4		50.0	
	40	3.84	61.0		61.4	
ck. vs. B[3/]		N.S.[5/]				
rates of B[4/]		N.S.				
2nd crop	0	5.17	24.5		22.3	
	20	5.55	48.0		49.0	
	40	5.27	57.5		59.5	
ck. vs. B		N.S.				
rates of B		N.S.				
7th crop	0	16.44	21.0		23.0	
	20	17.54	45.0		41.5	
	40	18.29	61.0		54.0	
ck. vs. B		N.S.				
rates of B		N.S.				
			Tops	Bottoms	Tops	Bottoms
15th crop	0	7.82	26.0	21.0	25.5	23.5
	20	8.29	32.0	25.0	35.0	27.0
	40	8.27	46.5	34.5	43.5	33.5
ck. vs. B		N.S.				
rates of B		N.S.				

1/ The averages of eight replications.

2/ Replications 1 thru 4 and 5 thru 8 were composited for boron determinations. The alfalfa from the 15th crop was divided into tops and bottoms; the tops consisting of the apical one or two inches and the bottoms consisting of the more mature portions.

3/ The average for the eight replications of zero borax treatments is compared with the average of the sixteen borax treatments.

4/ The averages for the two rates of the borax treatments are compared.

5/ N. S.--Not significant at the 5% level.

TABLE 11

THE YIELD AND BORON CONTENT OF ALFALFA GROWN ON THE WISNER CLAY LOAM EXPERIMENT

	Borax Treatment Lbs./Acre	Dry Matter[1/] Yields gms.	B p.p.m.[2/] reps. 1 thru 4		B p.p.m.[2/] reps. 5 thru 8	
1st crop	0	3.51	56.0		61.4	
	20	2.77	75.0		76.6	
	40	3.20	114.0		126.2	
ck. vs. B[3/]		N.S.[5/]				
rates of B		N.S.				
2nd crop	0	4.36	47.0		45.0	
	20	3.84	80.0		68.3	
	40	3.95	105.5		94.5	
ck.vs. B		N.S.				
rates of B		N.S.				
7th crop	0	10.83	28.3		29.0	
	20	10.26	45.5		45.3	
	40	11.13	53.0		59.7	
ck. vs. B		N.S.				
rates of B		N.S.				
			Tops	Bottoms	Tops	Bottoms
16th crop	0	7.41	15.0	18.5	16.5	17.5
	20	6.77	39.0	32.0	32.0	30.5
	40	6.98	55.5	45.0	59.5	50.5
ck. vs. B		N.S.				
rates of B		N.S.				

1/ The averages of eight replications.

2/ Replications 1 thru 4 and 5 thru 8 were composited for boron determinations. The alfalfa from the 16th crop was divided into tops and bottoms; the tops consisting of the apical one or two inches and the bottoms consisting of the more mature portions.

3/ The average for the eight replications of zero borax treatments is compared with the average of the sixteen borax treatments.

4/ The averages for the two rates of the borax treatments are compared.

5/ N.S. -- Not significant at the 5% level.

TABLE 12

THE YIELD AND COMPOSITION OF THE SEVENTEENTH CROP OF ALFALFA FROM THE THOMAS EXPERIMENT AND THE EIGHTEENTH CROP FROM THE WISNER EXPERIMENT AND THE pH AND EXTRACTABLE BORON DATA OBTAINED ON THE SOILS AFTER THESE CROPS WERE HARVESTED

		Plant Material[1]						Soils[2]	
	Borax Treatment Lbs./Acre	Dry Matter Yields gms.	B p.p.m.	Ca %	K %	Mg %	P %	B p.p.m.	pH
17th crop on Thomas soil	0	14.59	29.0	2.05	1.03	0.61	.472	.74	7.6
	20	14.13	35.1	1.83	1.04	0.56	.491	.81	7.6
	40	14.42	47.9	1.85	1.04	0.58	.488	1.01	7.5
ck. vs. B[2]		N.S.[4]	1%	N.S.	N.S.	N.S.	N.S.	1%	N.S.
rates of B[3]		N.S.	1%	N.S.	N.S.	N.S.	N.S.	1%	N.S.
18th crop on Wisner soil	0	13.60	15.2	3.11	0.98	0.53	.276	.52	8.0
	20	12.93	25.4	2.90	1.11	0.48	.288	.56	8.0
	40	12.47	40.6	3.16	1.13	0.52	.277	.59	8.0
ck. vs. B		N.S.	1%	N.S.	N.S.	N.S.	N.S.	5%	N.S.
rates of B		N.S.	1%	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

1/ The averages of eight replications. For pH, replicate values were first converted to H ion concentration.

2/ The average for the eight replications of zero borax treatments is compared with the average of the sixteen borax treatments.

3/ The averages for the two rates of borax treatments are compared.

4/ N.S. -- Not significant at the 5% level.

on Wisner soil than to alfalfa grown on the Thomas soil.

3. The reduction in availability of boron, according to plant tests, that took place on the Wisner soil probably is due at least in part, to the greater removal of boron by the plants in the early cuttings.

These results are in agreement with those obtained by Muhr (39), who found that the severity of boron toxicity was reduced by delaying the planting of soybeans after borax applications on a Thomas sandy loam of pH 7.5.

Even though the Thomas soil had a higher content of boron than did the Wisner before treatments were made, according to both the soxhlet and five minute boiling techniques (Table 19), the check treatments on the first and second crops had a higher content of boron in the alfalfa on the Wisner than on the Thomas (Tables 10 and 11). Apparently neither procedure is a good test for readily available boron when two different soil types are compared.

The extractable boron content of the soils, sampled at the time the seventeenth crop was harvested from the Thomas and the eighteenth crop was harvested from the Wisner soil, is presented in Table 12 along with the boron concentration in the alfalfa from these crops. It can be observed that both the extractable boron in the soil and the boron content of the plant material rise significantly with increments of added borax in the Thomas experiment. In the Wisner experiment, however, the extractable boron values of the soils are not appreciably different, either statistically or actually, although there is a considerable difference in the boron content of the alfalfa due to the treatments. Perhaps the boron associated with organic matter is in a different chemical form than that associated with clays or mineral soils and therefore is subject to better correlation between the extractable boron in the soil and the boron content of the plants.

The data do illustrate, once again, the impracticality of using extractable boron values to predict the amount of boron that plants can remove from soils. Both the seventeenth crop of alfalfa on the Thomas soil and the eighteenth crop on the Wisner soil were grown in the same 71 day period. It is therefore possible to calculate the average number of micrograms of boron removed per day by the plants. The Wisner soil (Table 12), with an extractable boron level of .59 p.p.m., released an average of 7.13 micrograms of boron per day while the Thomas soil, with an extractable boron level of .74 p.p.m., released an average of only 5.96 micrograms of boron per day.

B. The Oshtemo soil experiment.

The data from this experiment have been compiled in three different ways in order to make comparisons of the effects of the two variables, pH and rates of borax, more convenient.

First, the data are presented for the three levels of borax at pH 6.5 and 7.5 (Tables 13 and 16A)and then the results from the two pH levels are thrown together in order to present the data at the three borax levels only (Tables 14 and 16B). Finally the results from the three borax levels are averaged in order to present the data at the two pH levels (Tables 15 and 16C). As mentioned in the section on methods, the results from the unlimed soil treatments are not included with the comparisons at the two higher pH levels.

Weak symptoms of boron deficiency were observed for the first time on the third crop of alfalfa and continued to appear more severely on succeeding crops. These symptoms appeared only on the check treatments at pH 6.5 and 7.5 and never appeared on the alfalfa grown in borax treated soil during the entire experiment. Although no yield response to the borax applications occurred on the third crop, increments in yield resulted on the fourth, fifth and seventh crops (Table 14). The data of the sixth crop were not obtained because the samples were lost. It should be noted that yield response decreased with succeeding crops after the fourth crop of alfalfa (Table 14). Apparently this latter was due to several changes in the soil that were taking place simultaneously.

The calcium level in the soil and the pH were decreasing because of the depletory effect of crop removal of bases. As was noted in the section on methods, very little lime was required to raise the pH to 6.5 and 7.5 on this soil. This, of course, is indicative of a low base exchange capacity or low base supplying ability. It was also observed that the pH was reduced considerably from the initial reaction by the time the seventh crop was removed (Table 16). The plant analysis data in Table 16 indicate that the seventh crop was dangerously low in calcium and magnesium. Apparently the plants from the fourth crop on were progressively suffering the maleffects of high acidity and deficiencies of calcium and magnesium. These limiting factors were strong enough to eliminate all response to borax on the seventh crop grown on the soil of initial pH 6.5 (Table 13).

Since it is generally agreed that the availability of soil boron is increased when the soil reaction is lowered, this may be another factor worth considering. The boron contents of the plants were generally higher in all treatments on the seventh crop of alfalfa than on the preceding fourth and fifth crops (Table 13). This too may account for some of the decrease in response to boron, at least, in the seventh crop. A clue to this was noted before the plants were analysed for boron when it was observed that boron deficiency symptoms were not as prevalent on the seventh crop as they were on the preceding three crops.

TABLE 13

YIELD AND BORON CONTENT OF THE ALFALFA GROWN ON THE OSHTEMO SOIL AT TWO pH LEVELS AND THREE LEVELS OF BORAX

	Initial[1/] pH	Borax Treatment Lbs./Acre	Dry Matter Yields gms.	B[2/] p.p.m.	
1st crop	6.5	0	2.08	43.2	
		20	2.30	126.0	
		40	2.18	206.0	
	7.5	0	2.54	29.2	
		20	3.07	134.0	
		40	2.95	176.0	
L. S. D. 5%			0.63	Comp.[4/]	
L. S. D. 1%			N.S.[3/]		
2nd crop	6.5	0	2.40	23.5	
		20	2.80	82.7	
		40	2.03	140.3	
	7.5	0	2.65	20.2	
		20	3.51	89.3	
		40	3.25	128.2	
L. S. D. 5%			0.26	Comp.	
L. S. D. 1%			N.S.		
				Tops	Bottoms
3rd crop	6.5	0	3.60	8.1	10.7
		20	4.00	39.0	62.7
		40	3.86	65.5	82.2
	7.5	0	4.45	5.5	8.3
		20	4.53	43.7	51.0
		40	4.36	60.7	68.0
L. S. D. 5%			0.64	Comp.	Comp.
L. S. D. 1%			N.S.		

1/ pH at beginning of the experiment.
2/ The alfalfa from the third and fifth crops were divided into tops and bottoms in same manner as was stated previously.
3/ N.S. -- Not significant at the 5% or 1% level.
4/ Comp. -- The four replications were composited for boron determinations.

TABLE 13--Continued

	Initial pH	Borax Treatment Lbs./Acre	Dry Matter Yields gms.	B p.p.m.	
4th crop	6.5	0	4.90	6.0	
		20	7.57	19.1	
		40	7.72	35.3	
	7.5	0	6.99	6.7	
		20	9.61	21.7	
		40	9.44	39.7	
L. S. D. 5%			2.29	7.2	
L. S. D. 1%			3.17	9.9	
				Tops	Bottoms
5th crop	6.5	0	5.16	3.5	7.0
		20	5.73	19.5	18.5
		40	6.30	32.1	27.5
	7.5	0	5.41	4.5	6.2
		20	6.77	17.0	18.3
		40	6.71	40.3	33.0
L. S. D. 5%			1.27	Comp.	Comp.
L. S. D. 1%			1.75		
7th crop	6.5	0	4.98	7.6	
		20	5.08	35.4	
		40	5.04	57.5	
	7.5	0	5.15	11.5	
		20	6.45	36.1	
		40	6.58	56.7	
L. S. D. 5%			0.93	7.1	
L. S. D. 1%			1.29	9.8	

TABLE 14

YIELD AND BORON CONTENT OF THE ALFALFA GROWN ON THE OSHTEMO SOIL AT THREE LEVELS OF BORAX

	Borax Treatment Lbs./Acre	Dry Matter Yields gms.	B[1/] p.p.m.	
1st crop	0	2.31	36.2	
	20	2.69	130.0	
	40	2.56	191.0	
ck. vs. B		N.S.[2/]	Comp.[3/]	
rates of B		N.S.		
2nd crop	0	2.53	21.9	
	20	3.15	86.0	
	40	2.64	134.3	
ck. vs. B		N.S.	Comp.	
rates of B		N.S.		
			Tops	Bottoms
3rd crop	0	4.03	9.5	6.8
	20	4.27	56.9	41.3
	40	4.11	75.1	63.1
ck. vs. B		N.S.	Comp.	Comp.
rates of B		N.S.		
4th crop	0	5.94	6.4	
	20	8.59	20.4	
	40	8.58	37.5	
ck. vs. B.		1%	1%	
rates of B		N.S.	1%	

1/ The alfalfa from the third and fifth crops were divided into tops and bottoms in the same manner as was stated previously.

2/ N. S. -- Not significant at the 5% level.

3/ Comp. -- The four replications were composited for boron determinations.

TABLE 14--Continued

	Borax Treatment Lbs./Acre	Dry Matter Yields gms.	B p.p.m.	
			Tops	Bottoms
5th crop	0	5.29	4.0	6.6
	20	6.25	18.3	18.4
	40	6.51	36.2	30.3
ck. vs. B		1%	Comp.	Comp.
rates of B		N.S.		
7th crop	0	5.06	9.5	
	20	5.76	35.8	
	40	5.81	57.1	
ck. vs. B		5%	1%	
rates of B		N.S.	1%	

It should be mentioned that no yield response was obtained on the Oshtemo soil by the addition of more than enough borax than was required to prevent boron deficiency symptoms from occurring (Table 13) and that no deficiency occurred on any of the treatments on which borax was applied. Furthermore there was no significant yield response on the third crop because the deficiency was not severe enough (Tables 13 and 14).

When the boron contents of the top and bottom portions of the alfalfa are compared in the third crop it can be seen that the apical portions of nondeficient plants were much lower in boron than were the more mature parts. This, of course, is contrary to what was noted previously. The only explanation that can be offered for this anomaly is that the availability of the soil boron was decreasing very rapidly and for this reason there was more available boron at the beginning of the growth cycle than at the end. This explanation appears plausible when the boron contents of the first, second and fourth crops of alfalfa (Table 13) are compared. It can be seen, however, that the boron contents of the top and bottom portions of boron deficient and nondeficient alfalfa from the fifth crop follows the same general pattern as previously outlined (Table 13). Apparently the rate at which boron was supplied to the plants was more stable by the time the fifth crop was harvested.

Since the composition of the plants and the soils changed with each succeeding crop it would be ambiguous to attempt to predict a critical value for the boron content of apical portions of alfalfa grown on this soil. As was pointed out in the literature review and elsewhere in this paper, the boron requirements of plants vary considerably according to environmental conditions. However, a general discussion of the critical level may be of value if these ambiguities are kept in mind. Because the top sample from the third crop contained both deficient and nondeficient plants the critical level must have been less than 8 p.p.m. on the soil of initial pH 6.5 and less than 5.5 p.p.m. on the soil of initial pH 7.5 (Table 13). On the fifth crop, however, the top sample consisted entirely of deficient plants so that the critical level for the conditions that existed at this time must have been 3.5 p.p.m. or higher on the soil of initial pH 6.5 and 4.5 p.p.m. or higher on the soil of initial pH 7.5 (Table 13). Apparently the critical level of the apical portions of alfalfa grown on this soil was lower than that for alfalfa grown on the Wisner soil. This value was found to fall between 9 and 10 p.p.m. in the eighteenth crop of alfalfa grown on the Wisner clay loam (Table 9). These results are in agreement with the statement by Rogers (56) that alfalfa may require more boron when grown on soils with high base exchange capacities than when grown on coarse textured soils.

TABLE 15

YIELD AND BORON CONTENT OF THE ALFALFA GROWN ON THE OSHTEMO SOIL AT TWO pH LEVELS

	Initial[1/] pH	Dry Matter Yields gms.	B[2/] p.p.m.	
1st crop	6.5	2.19	125.1	
	7.5	2.85	113.1	
6.5 vs. 7.5[3/]		1%	Comp.[4/]	
2nd crop	6.5	2.41	82.2	
	7.5	3.14	79.2	
6.5 vs. 7.5		1%	Comp.	
			Tops	Bottoms
3rd crop	6.5	3.82	37.9	51.9
	7.5	4.45	33.3	42.4
6.5 vs. 7.5		1%	Comp.	Comp.
4th crop	6.5	6.73	20.1	
	7.5	8.68	22.7	
6.5 vs. 7.5		1%	N.S.	
			Tops	Bottoms
5th crop	6.5	5.73	18.4	17.7
	7.5	6.30	20.6	19.2
6.5 vs. 7.5		5%	Comp.	Comp.
7th crop	6.5	5.03	33.5	
	7.5	6.06	34.8	
6.5 vs. 7.5		1%	N.S.	

1/ pH at beginning of the experiment.

2/ The alfalfa from the third and fifth crops were divided into tops and bottoms in the same manner as was stated previously.

3/ The average of all the treatments grown on the Oshtemo soil with an initial pH of 6.5 compared to the average of all the treatments grown on the Oshtemo soil with an initial pH of 7.5.

4/ Comp. -- The four replications were composited for boron determinations.

An attempt was made to investigate the effect of different boron levels in soils and plants on the content of other nutrient elements in alfalfa (Tables 16A and 16B). Apparently none of these elements were affected by the boron level above the limit of deficiency. Toxic levels of boron were not attained. When the data from boron deficient plants are compared with those from nondeficient plants the picture changes. Boron deficiency did not appear to alter the calcium level in plants where this element was in ample supply (4th crop, Tables 16A and 16B) but did cause a reduction in plant calcium when that element was in short supply (7th crop, Tables 16A and 16B). The percentages of potassium, magnesium, phosphorus, and protein were all higher in the boron deficient plants than in the nondeficient ones. These differences may well have been due to a dilution effect since the yields of deficient plants were lower except for the seventh crop grown on the soil of initial pH 6.5 (Table 13). It can be seen that the percentages of potassium, magnesium and phosphorus were not significantly higher in deficient plants on this seventh crop (Table 16A) although it does appear that the phosphorus showed a tendency to be higher in the zero borax treatment. The fact that the composition was not altered by boron deficiency when the yields were the same, tends to support the proposition that the higher content of nutrient elements in boron deficient plants than in nondeficient plants, when yield responses were obtained; was due to a dilution effect. Because calcium was not affected by this dilution effect and was even lower in boron deficient plants when calcium was in short supply, there must have been a smaller amount of this element removed by boron deficient plants than by healthy ones. This conclusion is in agreement with a statement made by Purvis (52), but only in so far as comparisons are made between boron deficient and nondeficient plants. He contends that there is a functional relationship between calcium and boron and that as the boron level rises in the plant there is a tendency for a greater uptake of calcium.

It is quite difficult to compromise the results obtained on potassium, magnesium, and protein in this experiment with those obtained on the plant samples collected during the survey trip. However, it must be remembered that in this experiment the analytical data were obtained by analysing the whole plant while the main differences found on the survey samples were between top portions of boron deficient and nondeficient plants. Perhaps if the apical portions of the plants from this experiment had been analysed there would have been more agreement. Also, there is a wide difference between the conditions in the greenhouse and those in the field. One major difference is in the amount of soil that plants have for a rooting zone. In the small rooting zone that the plants have in the greenhouse, nutrients are more likely to become limiting and the probability

TABLE 16

THE COMPOSITION OF THE FOURTH AND SEVENTH CROP OF ALFALFA GROWN ON THE OSHTEMO LOAMY SAND AT TWO pH LEVELS AND THREE LEVELS OF BORAX; AND THE pH AND EXTRACTABLE BORON VALUES OBTAINED ON THE SOILS SAMPLED WHEN THE SEVENTH CROP WAS HARVESTED

A. Comparisons at pH 6.5 and 7.5 and Three Levels of Borox

	Initial[1] pH	Borax Treatment Lbs./Acre	Plant Material Ca %	K %	Mg %	P %	Protein %	Soils pH	B p.p.m.
4th crop	6.5	0	0.95	2.63	0.24	0.329	22.57		
		20	0.94	2.14	0.19	0.326	18.91		
		40	1.07	2.30	0.21	0.321	20.29		
	7.5	0	1.55	2.49	0.24	0.330	22.40		
		20	1.64	2.07	0.20	0.309	20.49		
		40	1.49	2.15	0.19	0.317	20.67		
L.S.D. 5%			0.35	0.27	N.S.	N.S.	1.82		
L.S.D. 1%			0.48	0.33			2.52		
7th crop	6.5	0	0.26	2.53	0.08	0.460		5.7	0.36
		20	0.37	2.29	0.08	0.391		5.5	0.43
		40	0.35	2.47	0.09	0.402		5.6	0.75
	7.5	0	0.48	2.17	0.16	0.468		6.3	0.41
		20	0.54	2.37	0.11	0.454		6.1	0.58
		40	0.52	2.31	0.10	0.404		6.1	0.52
L.S.D. 5%			0.08	.27	0.03	N.S.			0.22
L.S.D. 1%			0.12	N.S.	0.04				N.S.

1/ The pH at the beginning of the experiment.

TABLE 16--Continued

	Initial pH	Borax Treatment Lbs./Acre	Plant Material Ca %	K %	Mg %	P %	Protein %	Soils pH	B p.p.m.
B. Comparisons at Three Levels of Borax 2/									
4th crop		0	1.25	2.56	0.24	0.330	22.48		
		20	1.29	2.10	0.19	0.318	19.70		
		40	1.28	2.22	0.20	0.319	20.48		
ck. vs. B			N.S.	1%	1%	N.S.	1%		
rates of B			N.S.	N.S.	N.S.	N.S.	N.S.		
7th crop		0	0.37	2.62	0.12	0.464			0.39
		20	0.45	2.33	0.10	0.422			0.50
		40	0.43	2.39	0.09	0.403			0.64
ck.vs. B			1%	1%	5%	5%			5%
rates of B			N.S.	N.S.	N.S.	N.S.			N.S.
C. Comparisons at pH 6.5 and 7.5 3/									
4th crop	6.5		0.99	2.35	0.21	0.325	20.59		
	7.5		1.56	2.24	0.21	0.319	21.19		
6.5 vs.7.5			1%	N.S.	N.S.	N.S.	N.S.		
7th crop	6.5		0.33	2.43	0.09	0.417		5.6	0.51
	7.5		0.51	2.46	0.12	0.441		6.2	0.50
6.5 vs.7.5			1%	N.S.	1%	N.S.			N.S.

2/ The data obtained at the two pH levels are averaged.

3/ The data obtained at the three levels of borax are averaged.

of dilution effects occurring is increased. Another point that should be mentioned is that boron deficiency is usually quite spotty in the field so that deficient plants are often in direct competition with healthy ones. This too is likely to cut down on dilution effects.

The data in Table 15 illustrate the consistent increase in yield due to the higher pH level, but it is more important for the purpose of this work to examine the effect of reaction on the boron level in the plant. It can be seen that in the first three crops, the boron level was higher in the plant material at the lower pH level but that there was little difference in the fourth, fifth and seventh crops (Table 15). It should be noted that if the basis for comparison were the total amount of boron removed from the soil by the plants, the plants grown on the soil of highest initial pH consistently removed more boron. It is a point for disagreement as to whether the concentration of an element in the plant or the total amount removed by the plant is the best measure of the availability of that element. For this reason it must be stated that the evidence in this experiment is inconclusive as to the effect of pH or calcium level on the availability of soil boron. As mentioned previously, the extractable boron determinations on the soils were found to be an inferior measure of either the actual or relative amounts of plant available boron but that when single soils were compared there was some correlation between these factors (Table 12). With this in mind it should be noted that there was no difference between the extractable boron values obtained on the soils at the two pH levels after seven cuttings were removed (Table 16C).

As would be expected the calcium content of the plants was significantly higher when grown on the soil of initial pH 7.5 than on the soil of initial pH 6.5 (Table 16C). The only other element significantly affected by soil reaction in this experiment was magnesium in the seventh crop (Table 16C). Perhaps the reason that this element was more concentrated in the plants grown at the higher initial pH level was because the "foraging" ability of the alfalfa roots was improved due to the fact that calcium was not as limiting on this treatment.

As mentioned at the beginning of this section the data from the soil of initial pH 5.5 were not included with the data from the limed soils. The condition of the plants grown on the unlimed soil appeared to improve after an application of NH_4NO_3 and boron deficiency symptoms appeared on the check treatments. This crop was harvested, oven dryed, weighed, and analysed for boron. The soils were also sampled at this time and were tested for reaction and extractable boron content. These data are presented in Table 17. The yields were too variable to be significantly different at the 5% level. It can be seen that the boron contents of the plants tend to be

TABLE 17

THE YIELD AND BORON CONTENT OF THE LAST CROP OF ALFALFA GROWN ON THE UNLIMED OSHTEMO LOAMY SAND AT THREE LEVELS OF BORAX AND THE EXTRACTABLE BORON AND pH OF THE SOIL SAMPLED AT THE TIME THIS CROP WAS HARVESTED

Borax Treatment	Plant Material			Soils	
	Dry Matter Yields gms.	B 1/ p.p.m. Tops	B 1/ p.p.m. Bottoms	pH	B p.p.m.
0	3.95	8.0	6.6	5.5	0.48
20	3.31	41.4	33.0	5.5	0.56
40	6.18	54.4	38.6	5.3	0.57
L.S.D. 5%	N.S. 2/	Comp. 3/	Comp.		N.S.

1/ The alfalfa was divided into top and bottom portions in the same manner as was stated previously.

2/ N.S. -- Not significant.

3/ Comp. -- The four replications were composited for boron determinations.

directly related to the amount of borax applied. Perhaps the main point to be emphasized from this portion of the experiment is that boron deficiency can occur, even under acid conditions if the plants can be made to grow thriftily. This was also noted on the soil of initial pH 6.5. By the time the seventh crop was removed, the soil pH was reduced to about 5.6 and boron deficient plants were still observed.

It is quite difficult to find a consistent meaning from the extractable boron values obtained on the soils sampled at the time the final crops were harvested (Tables 16A and 17). However, the check treatments tended to be uniformly low, due probably to crop removal (Table 18) and fixation due to liming. The high L. S. D. value required for significance at the five percent level indicates that the individual figures obtained for extractable boron were quite variable (Table 16A). Actually, the 0.75 p.p.m. figure obtained for extractable boron on the soil of initial pH 6.5, which was treated with borax at the rate of forty pounds per acre, was the only one significantly higher than that obtained for the check soil. These results are in direct contrast to the highly significant differences in the results obtained from the plant tests (seventh crop, Table 13). Although no statistical analysis could be performed on the boron determinations made on the plant material harvested from the unlimed soils, it appears that they were quite well correlated with the borax treatments

(Table 17). On the other hand there were no significant differences between extractable boron averages obtained on this soil. Clearly then, the amount of boron extracted by the plants was a better indication of the amount of borax added to the limed and unlimed Oshtemo soils than was the amount of boron extracted in the five minute boiling procedure. Obviously, the boron in this soil must have been in a form that was available to the plant but was not readily extractable from the soil with hot water.

It is apparent from an examination of Table 18 that most of the boron applied to the soils was still present after the seventh crop of alfalfa was removed. The reader may recall that there was no means of drainage. Since the remaining boron was not extracted by the five minute boiling technique (Table 18) it obviously was in a much less soluble form than the borax applied. Just what form this boron was in is open to speculation, but because of the low colloid content of this soil, it is improbable that it was fixed by adsorption. One possible means of fixation is by precipitation in some relatively insoluble chemical form; perhaps as a borosilicate or calcium borosilicate. As was mentioned in the literature review, many investigators have shown that boron can be fixed in this manner. Wear (68) found that a borosilicate (Howlite) could be used as a source of boron for plants. More important is the fact that the five minute boiling procedure for extracting boron from this compound gave a very poor indication of its actual boron supplying power.

TABLE 18

AN ACCOUNTING OF THE EXCHANGE OF BORON BETWEEN THE OSHTEMO LOAMY SAND AND THE ALFALFA HARVESTED

Initial[1] pH	Final[2] pH	B Added per Pot Micrograms	B Removed[3] By Plants Micrograms	Added B Remaining in each Pot Micrograms	Remaining Boron p.p.m.	Extractable[4] Boron p.p.m.
5.5						0.48[5]
6.5	5.7	0	313	-313	-0.070	0.36
6.5	5.5	4,550	1,338	3,212	0.714	0.43
6.5	5.6	9,100	2,008	7,092	1.574	0.75
7.5	6.3	0	338	-338	-0.075	0.41
7.5	6.1	4,550	1,703	2,847	0.633	0.58
7.5	6.1	9,100	2,477	6,623	1.472	0.52

1/ The pH at the beginning of the experiment.

2/ The pH at the time the seventh crop was removed.

3/ An estimate had to be made on the amount of boron removed by the sixth crop.

4/ Extractable boron values were made on the soils after the seventh crop was removed.

Conclusions

The data presented in this section indicate that the yield and nutrient composition of alfalfa are not affected by variations in boron levels in soils or plants above the limits of boron deficiency. Moreover, no significant yield response to borax applications could be demonstrated when there were only weak symptoms of boron deficiency on the alfalfa grown on the check treatments of the Wisner clay loam and Oshtemo loamy sand. Considerable yield increments due to borax applications were measured on the limed treatments of the Oshtemo loamy sand when severe boron deficiency occurred. Potassium, magnesium, phosphorus, and protein levels were higher in boron deficient plants than in healthy ones. This relationship was interpreted as being due to a dilution effect. The calcium content of alfalfa was not altered by boron deficiency when calcium was in ample supply but it was lower in boron deficient plants when calcium was limiting. The total amount of calcium taken up by boron deficient plants was lower in both cases than that removed by nondeficient plants. This fact is considered as evidence in support of the work by Purvis (52) who found there was a functional relationship between boron and calcium, and that when one ion was absorbed in large amounts by the plant the other also tended to be taken up in larger amounts.

The critical level of boron in the apical portions of alfalfa tended to be higher in alfalfa grown on the Wisner clay loam than in that grown on the Oshtemo loamy sand. The reason for this can be only speculative but it can be noted that the calcium level in the alfalfa grown on the Wisner soil was much higher than it was in that grown on the Oshtemo soil.

The results of this experiment were found to be inconclusive as to what effect exchangeable calcium level or the pH of the soil had on the plant availability of boron.

The apical portions of boron deficient alfalfa were lower in boron than were the more mature portions and the reverse was true of nondeficient plants except in one case. In this instance it is believed that the boron added to the soil was being fixed so rapidly that there was more boron taken up by the plants at the beginning of the growth cycle than at the end.

It was shown that after very soluble borax was added to the soil it was altered in some way so that it became much less soluble. The less soluble form or forms were still available to the plants but the relative amount present could not be measured with any degree of certainty by the five minute boiling technique of extraction on Wisner clay loam or the Oshtemo loamy sand. The extractable boron values obtained on the Thomas sandy loam, however, correlated very well with the amount of borax added and the amount of boron removed by the plants. Since the Thomas soil contains eight percent organic matter and is calcareous at the surface, these data indicate that the boron fixed by organic matter under alkaline conditions is in a different form than that fixed by mineral soils and can be evaluated by the five minute boiling procedure of extraction. The data in this experiment also indicate that boron is fixed much more rapidly by alkaline organic soils than by mineral soils. The impracticality of using extractable boron tests as a measure of the boron supplying power of soils was once again illustrated in this experiment.

SOXHLET EXTRACTIONS OF SOILS

Introduction

Because of the promising success that Baird (3) and McClung (36) had with the soxhlet extraction procedure for determining available soil boron, it was decided that perhaps this method would prove more fruitful in accounting for the boron content of alfalfa than Berger's (7) five minute boiling procedure.

Methods

Standard soxhlet extracting equipment was used, thus blank determinations of boron had to be run in order to correct for the boron contamination from the glassware and thimbles. Determinations were made using twenty grams of the Oshtemo, Kalkaska, and Fox soils and ten grams of the Wisner and Thomas soils. Two milliliters of five normal NaOH was added to approximately 100 ml. of distilled water in the boiling flask in order to prevent volatilization of the boron during the extraction and drying period. The extractions were made over a six hour period during which time about 750 ml. of hot distilled water leached the soil. The solution in the boiling flask was then placed in a 500 ml. boron free beaker and evaporated almost to dryness and then transferred to a

50 ml. volumetric flask with 0.1 N HCl. The boron determination was made on this solution by the curcumin procedure.

Results and Discussion

Soils were purposely chosen which showed distorted results when a comparison was made between the boron content of the plants and boron extracted from the soil by the five minute boiling procedure. The results of these tests are compiled in Table 19. It is obvious that the determinations made by the soxhlet extracting procedure correspond more closely to the boron content of the plant material and to soil treatments than do the determinations performed by the five minute boiling technique. The one exception to this relationship was a Fox sandy loam which was one of the soils used in Field Experiment II. There is a good possibility that the borax was leached out of the surface and the plants were obtaining their boron from a lower horizon.

A comparison of the data on the Oshtemo loamy sand of initial pH 7.5 in Tables 18 and 19 indicates that only a small portion of the applied boron that remained in the soil after cropping was removed by the soxhlet extracting procedure. Evidently much more work is required; first, to discover in what forms soluble boron compounds are "fixed" in the soil; second, to discover how available the boron in these "fixed"

TABLE 19

A COMPARISON OF THE AMOUNTS OF BORON EXTRACTED FROM SOILS BY THE FIVE MINUTE BOILING AND BY THE SOXHLET PROCEDURE WITH THE BORON CONTENT OF ALFALFA GROWN ON THESE SOILS

Soil Type[1]	Replication	Soil Treatment	Soxhlet Extractable B p.p.m.	5 Minute Extractable B p.p.m.	Alfalfa B p.p.m.
Oshtemo L.S.[2]		original untreated sample	0.983	0.48	
Oshtemo L.S.	4	pH 5.5, 0 lbs. of borax	0.968	0.37	
Oshtemo L.S.	4	pH 5.5, 20 lbs. of borax	1.068	0.57	
Oshtemo L.S.	4	pH 5.5, 40 lbs. of borax	1.257	0.46	
Oshtemo L.S.	3	pH 7.5, 0 lbs. of borax	0.563	0.44	9.5
Oshtemo L.S.	3	pH 7.5, 20 lbs. of borax	0.637	0.51	41.2
Oshtemo L.S.	3	pH 7.5, 40 lbs. of borax	0.688	0.38	48.0
Wisner C.L.[3]		original untreated sample	5.125	1.00	
Wisner C.L.	5	0 lbs. of borax	3.175	0.60	19.7
Wisner C.L.	5	20 lbs. of borax	3.878	0.48	22.0
Wisner C.L.	5	40 lbs. of borax	4.750	0.60	46.5
Thomas S.L.[4]		original untreated sample	7.250	1.14	
Thomas S.L.	1	0 lbs. of borax	3.625	0.76	28.4
Thomas S.L.	1	20 lbs. of borax	4.200	0.73	38.0
Thomas S.L.	1	40 lbs. of borax	7.500	1.11	51.0
Kalkaska L.S.	4	0 lbs. of borax	1.135	0.58	16.5
Kalkaska L.S.	4	30 lbs. of borax	1.500	0.66	59.0
Fox S.L.	3	0 lbs. of borax	1.193	0.46	10.5
Fox S.L.	3	30 lbs. of borax	1.100	0.46	22.9

1/ The Kalkaska and Fox soils were used in Field Experiment II and the remainder were used in the Greenhouse Experiments.

2/ Loamy sand

3/ Clay loam

4/ Sandy loam

compounds is to the plant; and third, to find a method of extraction that removes a quantity of boron similar or in relative proportions to that absorbed by plants.

SUMMARY AND GENERAL CONCLUSIONS

Because of the inherent defects in soil testing procedures for boron, it is the opinion of the author that at the present time, the best procedure for estimating the boron supplying power of a soil is a biological test. The top one or two inches of the growing points of the alfalfa plant should be sampled, but only after a two or three week period of hot, dry weather. This period may be shortened for excessively drained soils or soils with a low water holding capacity. If the boron content of this plant material is found to approach 20 p.p.m. or less, it should be taken as a danger sign and recommendations for borax applications ought to be forthcoming. It is believed that the apical portions of the plant reflect the current supply of boron. Therefore the boron content of this portion of the plant is a measure of the boron supplying power of the lower horizons of the soil when the supply from the surface soil layer is reduced due to desiccation. This type of test is possible only because boron is not translocated from the more mature portions of the plant to the apical meristem.

It has been demonstrated that boron deficiency can occur at levels considerably higher than 20 p.p.m. in alfalfa in exceptional cases. If boron deficiency is suspected, the apparently boron deficient plants should be subdivided by removing the top one or two inches of the plant and boron determinations made on both portions. If the top portions are lower in boron than the more mature portions then this can be taken as a positive test for boron deficiency.

It has been demonstrated that yield responses can be obtained from borax applications on deficient soils. However, as with any other essential element, in order to obtain maximum benefits from borax applications, all other conditions should be maintained at an optimum. Not only must pH and fertility levels be satisfactory but insect damage should be controlled. It was noted that damage by insects was most severe on soils that are most likely to be boron deficient. That is, on coarse textured, droughty soils. The most serious offenders appeared to be the immature spittle bug on the first crop and the potato leaf hopper, grasshoppers, and the mature spittle bug on the second and third crops.

It has also been demonstrated that the mineral and protein contents of the boron deficient portions of alfalfa are lower than in the same portions of healthy plants. A quality response to fertilization with borax is therefore indicated.

Borax should be applied at the rate of 20 or 30 pounds per acre as a top dressing in the early spring or late fall in order to allow time for the borax to be leached into the lower soil horizons where it can be utilized when the surface is dry. The higher rate should be applied on the finer textured soils and on soils with a high organic matter content. The lower rate should be applied to the coarse soils but at more frequent intervals. Just how long an application may remain effective in the various soil types is open to speculation. However, because of their higher "fixing" ability, finer textured soils and soils with a high organic matter content are more likely to require less frequent applications of borax than coarse textured soils. This is especially true since it was demonstrated in these experiments that this so-called "fixed" boron is a rich source of boron to the plant, although it is not readily extracted from mineral soils with hot water. Perhaps the use of less soluble boron fertilizers on excessively drained soils would be profitable if they could be procured economically.

Most investigators are of the opinion that borax applications should not be banded with the seed at planting time because of the danger to the germinating seed. Broadcast applications may be made before planting, but even this is dangerous if a companion crop is associated with the seeding because of the low borax tolerance of grains.

LITERATURE CITED

1. Asen, Sam and O. W. Davidson. The boron distribution in greenhouse rose plants. Amer. Soc. Hort. Sci. 56:433-438, 1950.

2. Bailey, L. F. and J. S. Hargue. Effect of boron, copper, manganese and zinc on the enzyme activity of tomato and alfalfa plants grown in the greenhouse. Plant Phys. 19:105-116, 1944.

3. Baird, Guy B. Some studies on the characterization of available soil boron. Ph.D. Thesis, Cornell Univ. 1952.

4. Barber, S. A. Boron deficiency in Indiana soils. Ind. Exp. Sta. Cir. 387, 1953.

5. Beckenback, J. R. Functional relationships between boron and various anions in the nutrition of the tomato. Fla. Agr. Exp. Sta. Tech. Bul. 395. 1944.

6. Berger, K. C. Boron in soils and crops. Advances in Agronomy I, New York Academic Press, Inc., 1949. 321-351 pp.

7. Berger, K. C. and E. Truog. Boron tests and determinations for soils and plants. Soil Sci. 57:25-36, 1944.

8. Berger, K. C. and E. Truog. Boron availability in relation to soil reaction and organic matter content. Soil Sci. Soc. Amer. Proc. 10:113-116, 1945.

9. Brennan, E. G. and J. W. Shive. Effect of calcium and boron nutrition of the tomato on the relation between elements in the tissue. Soil Sci. 66:65-76, 1948.

10. Brown, B. A., R. I. Munsell and A. V. King. Potassium and boron fertilization of alfalfa on a few Connecticut soils. Soil Sci. Soc. Amer. Proc. 10:134-140, 1945.

11. Colwell, W. E. A biological method for determining the relative boron contents of soils. Soil Sci. 56:71-93, 1943.

12. Colwell, W. E. and R. W. Cumming. Chemical and biological studies on aqueous solutions of boric acid and calcium, sodium and potassium metaborates. Soil Sci. 57:37-50, 1944.

13. Colwell, W. E. and C. Lincoln. A comparison of boron deficiency symptoms and potato leaf hopper injury on alfalfa. Jour. Amer. Soc. Agron. 34:495-498, 1942.

14. Cook, R. L. and C. E. Millar. Some soil factors affecting boron availability. Soil Sci. Soc. Amer. Proc. 4:297-301, 1939.

15. Cook, R. L. and C. E. Millar. The effect of borax on yield, appearance and mineral composition of spinach and sugar beets. Soil Sci. Soc. Amer. Proc. 5:227-234, 1940.

16. Dawson, J. E. and A. F. Gustafson. A study of technique for predicting potassium and boron requirements of alfalfa. II Influence of borax on deficiency symptoms and boron content of the plant and the soil. Soil Sci. Soc. Amer. Proc. 10:147-149, 1945.

17. DeTurk, E. E. and L. C. Olson. Determination of boron in some soils of Illinois and Georgia. Soil Sci. 52:351-357, 1941.

18. Diagnostic techniques for soils and crops. Washington, D. C., The American Potash Institute, 1948, 81 and 174 pp.

19. Dible, W. T. and K. C. Berger. Boron content of alfalfa as influenced by boron supply. Soil Sci. Soc. Amer. Proc. 16:60-61, 1952.

20. Drake, M., D. H. Sieling and G. D. Scarseth. Calcium-boron ratio as an important factor in controlling the boron starvation of plants. Jour. Amer. Soc. Agron. 33:454-462, 1941.

21. Dregne, H. E. and W. L. Powers. Boron fertilization of alfalfa and other legumes in Orgeon. Jour. Amer. Soc. Agron. 34:902-912, 1942.

22. Eaton, F. M. and L. V. Wilcox. The behavior of boron in soils. U.S.D.A. Tech. Bul. 696, 1939.

23. Fisher, E. H. and K. C. Berger. Alfalfa seed production as influenced by insecticides and fertilizer application. Jour. Econ. Ent. 44:113-114, 1951.

24. Haas, A. R. C. The turmeric determination of water soluble boron in soils of citrus orchards in California. Soil Sci. 58:123-137, 1944.

25. Hatcher, J. T. and L. V. Wilcox. Colorimetric determination of boron using carmine. Anal. Chem. 22:567-569, 1950.

26. Haynes, J. L. and W. Rei Robbins. Calcium and boron as essential factors in the root environment. Jour. Amer. Soc. Agron. 40:795-803, 1948.

27. Hobbs, J. A. and B. R. Bertramson. Boron uptake by plants as influenced by soil moisture. Soil Sci. Soc. Amer. Proc. 14:257-261, 1949.

28. Hutcheson, T. B. and R. P. Cocke. Effects of boron on yield and duration of alfalfa. Va. Agr. Exp. Sta. Bul. 336, 1941.

29. Jones, H. E. and G. D. Scarseth. The calcium-boron balance in plants as related to boron needs. Soil Sci. 57:15-24, 1944.

30. Jordan, J. V. and G. R. Anderson. Effect of boron on nitrogen fixation by azotobacter. Soil Sci. 69:311, 1950.

31. Jordan, J. V. and W. L. Powers. Status of boron on Oregon soil and plant nutrition. Soil Sci. Soc. Amer. Proc. 11:324-331, 1946.

32. Kitson, R. E. and M. G. Mellon. Colorimetric determinations of phosphorus as molybdivanadophosphoric acid. Ind. Eng. Chem., A.E. 16:379-383, 1944.

33. Kochler, F. E. and W. A. Albrecht. Biosynthesis of amino acids according to soil fertility. III Bioassays of forage and grain fertilized with trace elements. Plant and Soil 4:336-344, 1953.

34. Kubota, J., K. C. Berger and E. Truog. Boron movement in soils. Soil Sci. Amer. Proc. 13:130-134, 1948.

35. Leggatt, C. W. Germination of boron deficient peas. Sci. Agric. (Ottawa) 28:131-139, 1948.

36. McClung, A. C. and J. E. Dawson. Some studies on the behavior of soil boron under cropping. Soil Sci. Soc. Amer. Proc. 15:268-272, 1950.

37. Marsh, R. P. and J. W. Shive. Boron as a factor in the calcium metabolism of the corn plant. Soil Sci. 51:141-151, 1941.

38. Midgley, A. R. and D. E. Dunkee. The effect of lime on fixation of borates in soils. Soil Sci. Soc. Amer. Proc. 4:302-307, 1939.

39. Muhr, G. R. Available boron as effected by soil treatments. Soil Sci. Soc. Amer. Proc. 5:220-226, 1940.

40. Mulder, E. G. Investigations on nitrogen nutrition of pea plants. Plant and Soil 1:179-212, 1948.

41. Mulvehill, J. F. and J. M. MacGregor. The effect of some trace elements on the yield and composition of alfalfa and oats in Minnesota. Soil Sci. Soc. Amer. Proc. 19:204-209, 1955.

42. Naftel, J. A. Soil liming investigations. V The relation of boron deficiency to over-liming injury. Jour. Amer. Soc. Agron. 29:537-547, 1937.

43. Naftel, J. A. Colorimetric microdetermination of boron. Ind. Eng. Chem. A.E. 11:407-409, 1939.

44. Olsen, R. V. and K. C. Berger. Boron fixation as influenced by pH, organic matter content, and other factors. Soil Sci. Soc. Amer. Proc. 11:216-220, 1946.

45. Page, N. R. and W. R. Paden. Differential response of snapbeans, crimson clover and turnips to varying rates of calcium and sodium borate on three soil types. Soil Sci. Soc. Amer. Proc. 14:253-257, 1949.

46. Page, N. R. and W. R. Paden. Boron supplying power of several South Carolina soils. Soil Sci. 77:427-434, 1954.

47. Parks, R. Q. The fixation of added boron by a Dunkirk fine sandy loam. Soil Sci. 57:405-416, 1944.

48. Parks, R. Q., C. B. Lyon and S. L. Hood. Some effects of boron supply on the chemical composition of tomato leaflets. Plant Phys. 19:404-416, 1944.

49. Parks, R. Q. and B. T. Shaw. Possible mechanisms of boron fixation in soil:1. Chemical. Soil Sci. Soc. Amer. Proc. 6:219-223, 1941.

50. Parks, W. L. and J. L. White. Boron retention by clay and humus systems saturated with various cations. Soil Sci. Soc. Amer. Proc. 16:298-300, 1952.

51. Piland, J. R., C. F. Ireland and H. M. Reisenauer. The importance of borax in legume seed production in the South. Soil Sci. 57:75-84, 1944.

52. Purvis. E. R. and O. W. Davidson. Review of the relations of calcium to availability and adsorption of certain trace elements by plants. Soil Sci. 65:111-116, 1948.

53. Purvis, E. R. and W. J. Hanna. Influence of overliming on boron deficiency. Amer. Fert. 91:5-7, 24, 26, 1939.

54. Reeve, E., A. L. Prince and F. E. Bear. Boron needs of New Jersey soils. N. J. Agr. Exp. Sta. Bul. 709, 1944.

55. Reeves, C. and J. W. Shive. Potassium-boron and calcium-boron relationships in plant nutrition. Soil Sci. 57:1-23, 1944.

56. Rogers, H. T. Water soluble boron on coarse textured soils in relation to need of boron fertilization for legumes. Jour. Amer. Soc. Agron. 39:914-928, 1947.

57. Russel, D. A., L. T. Kurtz and S. W. Melsted. The response of alfalfa to borax fertilizer on Illinois soils. Abstracts of the forty-sixth annual meeting of the Amer. Soc. of Agron. 39 pp., 1954.

58. Schaller, F. W. Boron content and requirements of West Virginia soils. Soil Sci. 66:335-346, 1948.

59. Simmons, C. F. Alabama reports on secondary and minor elements in commercial fertilizers. Plant Food Jour. 2:6, 1948.

60. Sheldon, V. L., W. G. Blue and W. A. Albrecht. Biosynthesis of amino acids according to soil fertility. I Tryptophane in forage crops. Plant and Soil 3:33-40, 1951.

61. Smith, H. V. Boron as a factor in Arizona's agriculture. Univ. of Ari. Tech. Bul. 118, 1949.

62. Smith, P. F. and W. Reuther. The response of young valencia orange trees to differential boron supply in sand cultures. Plant Phys. 26:110-114, 1951.

63. Stinson, C. H. Relation of water soluble boron in Illinois soils to the boron content of alfalfa. Soil Sci. 75:31-36, 1953.

64. Struckmeyer, B. E. and R. MacVicar. Relation of photoperiod to the boron requirements of plants. Bot. Gaz. 109:237-249, 1948.

65. Wadleigh, C. H. and J. W. Shive. A microchemical study of the effects of boron deficiency in cotton seedlings. Soil Sci. 47:33-36, 1938.

66. Walker, J. C. Histologic-Pathological effects of boron deficiency. Soil Sci. 57:51-54, 1944.

67. Wallace, A. and F. E. Bear. Influence of potassium and boron on nutrient-element balance and growth of Ranger alfalfa. Plant Phys. 24:664-680, 1949.

68. Wear, J. I. and C. M. Wilson. Boron materials of low solubility and their use for plant growth. Soil Sci. Soc. Amer. Proc. 18:425-428, 1954.

69. Whetstone, R. R., W. O. Robinson and H. O. Byers. Boron distribution in soils and related data. U. S. D. A. Tech. Bul. 797, 1942.

70. Winsor, H. W. Boron sources of moderate solubility as supplements for sandy soils. Soil Sci. 69:321-332, 1950.

71. Winsor, H. W. Boron retention of Rex fine sand as related to particle size of colemanite supplements. Soil Sci. 71:99-103, 1951.

72. Winsor, H. W. Variations in soil boron with cultivation and season. Soil Sci. 74:359-364, 1952.

73. Winsor, H. W. Penetration and loss of heavy applications of borax in Florida mineral soils. Soil Sci. 74:459-466, 1952.

74. Wolf, B. Factors influencing availability of boron in soil and its distribution in plants. Soil Sci. 50:204-218, 1940.

APPENDIX 1

COMPILED DATA ON PLANT MATERIAL AND SOIL SAMPLES COLLECTED ON SURVEY TRIP THROUGH THE LOWER PENINSULA OF MICHIGAN

				Plant Material					Soils	
Location	Soil Type 1/	Deficiency 2/ Status	Plant 3/ Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	B p.p.m.
1	Hillsdale S.L.	D	T	11.8	1.41	1.78	.29	24.94	5.6	0.88
			B	9.0	1.43	1.65	.29	17.19		
		ND	T	16.7	1.34	2.09	.31	28.38	5.4	0.78
			B	15.2	1.31	1.67	.20	17.00		
2	Hillsdale S.L.	D	T	8.7	1.65	1.21	.29	20.63	6.5	0.58
			B	10.1	1.48	1.25	.28	17.31		
		ND	T	27.5	1.86	1.70	.30	24.75	6.3	0.78
			B	22.5	1.82	1.42	.24	16.00		
3	Brookston L.	ND	T	30.8	1.21	2.76	.30		7.1	1.29
			B	29.5	1.47	2.25	.28			
4	Coloma L.S.	D	T	9.2	.93	2.01	.27	22.75	6.4	0.52
			B	10.6	.82	2.15	.22	18.81		
		ND	T	15.0	1.15	2.16	.29	26.94	6.4	0.58
			B	11.9	1.12	2.02	.22	18.00		
5	Fox S.L.	D	T	8.9	2.05	1.04	.25		6.3	0.62
			B	9.9	2.02	1.05	.36			
6	L.	D	T	9.5	1.86	1.16	.24	16.25	6.8	0.93
			B	13.3	1.82	1.47	.27	14.69		
		ND	T	13.5	1.83	1.35	.31	21.88		
			B	14.9	2.03	1.40	.26	16.94		

1/ S-sand, L-loam, Si-silt, C-clay.
2/ D-boron deficient plants, ND-nondeficient plants.
3/ T-tops, B-bottoms, C-tops and bottoms combined.

APPENDIX 1--Continued

Location	Soil Type	Deficiency Status	Plant Material: Plant Parts	Plant Material: B p.p.m.	Plant Material: Ca %	Plant Material: K %	Plant Material: Mg %	Plant Material: Protein %	Soils: pH	Soils: B p.p.m.
7	Miami L.	D	T	9.6	2.13	1.40	.29	22.19	6.2	0.88
			B	16.2	1.92	1.23	.36	16.94		
		ND	T	29.2	2.08	1.76	.37	23.94	5.0	0.70
			B	20.8	1.98	1.50	.25	17.44		
8	S. L.	ND	C	36.1	1.02	2.40	.24		5.4	0.79
9	Haytville Si. C. L.	ND	T	33.8	.88	3.03	.18		7.0	1.44
			B	28.5	.93	2.78	.11			
10	Wauseon Si. L.	ND	T	43.5	2.38	1.30	.38		6.4	1.04
			B	37.9	2.27	1.51	.35			
11	S. L.	ND	T	34.4	1.83	2.61	.29		6.1	0.94
			B	30.1	2.12	2.41	.28			
12	S. L.	D	T	30.8	2.07	1.03	.27	16.00	7.2	0.82
			B	33.5	2.02	1.35	.37	17.31		
		ND	T	41.6	2.34	1.46	.40	24.31	7.1	0.72
			B	32.3	2.00	1.08	.33	18.06		
13	S. L.	D	T	7.5	1.73	1.27	.24	22.00	5.6	0.46
			B	8.9	1.86	1.43	.33	18.63		
		ND	T	23.1	2.23	1.35	.37	22.44	5.6	0.58
			B	20.2	1.79	1.49	.27	17.63		
14	L. S.	D	T	6.8	1.41	1.08	.25	24.94	6.2	0.72
			B	8.5	1.43	.90	.35	18.81		
		ND	T	20.4	1.68	1.27	.42	25.63	6.1	0.72
			B	15.0	1.58	.82	.34	18.25		

APPENDIX 1--Continued

			Plant Material						Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	B p.p.m.
15	L. S.	ND	T	25.2	2.02	1.15	.43		6.1	0.72
			B	19.4	2.00	.61	.48			
16	Si. L.	ND	T	40.8	1.85	2.18	.50		6.5	1.45
			B	31.4						
17	L. S.	D	T	5.0	1.93	.98	.30	23.81	6.8	0.64
			B	9.8	1.73	.77	.37	17.81		
		ND	T	18.3	1.72	1.69	.41	26.69	6.4	0.80
			B	17.2	1.77	1.24	.41	20.25		
18	Nappanee Si. L.	ND	T	31.2	1.17	2.11	.43		6.5	0.78
			B	26.7	1.33	2.02	.46			
19	S. L.	ND	T	33.4	1.91	1.78	.45		6.6	0.96
			B	18.5	1.61	1.28	.35			
20	Mancelona L. S.	D	T	7.5	2.31	1.48	.33	23.94	6.9	0.82
			B	11.0	2.27	1.51	.44	19.31		
		ND	T	25.6	1.56	2.85	.31	32.94	6.8	1.34
			B	19.0	1.54	2.56	.24	23.38		
21	Marlette L.	ND	T	38.0	1.39	3.08	.30		5.7	0.82
			B	25.2	1.28	2.78	.22			
22	Nester Si. C. L.	ND	T	29.2	1.49	2.18	.37		6.8	0.90
			B	21.8	1.35	2.41	.39			
23	Kawkawlin Si. L.	ND	T	28.9	1.15	2.49	.37		6.8	1.09
			B	20.2	1.31	1.91	.33			

APPENDIX 1--Continued

Location	Soil Type	Deficiency Status	Plant Material: Plant Parts	Plant Material: B p.p.m.	Plant Material: Ca %	Plant Material: K %	Plant Material: Mg %	Plant Material: Protein %	Soils: pH	Soils: B p.p.m.
24	Si. L.	ND	T	25.2	1.20	1.83	.33		6.7	1.08
			B	20.5	1.58	1.43	.40			
25	Mancelona L. S.	D	T	8.3	1.46	1.37	.25	22.63	7.1	0.60
			B	11.1	1.52	1.61	.27	19.50		
		ND	T	36.3	1.53	2.02	.34	27.13	6.3	0.94
			B	25.8	1.70	1.62	.31	18.44		
26	L. S.	D	T	10.6	1.65	1.28	.26		6.6	0.60
			B	11.9	1.65	1.44	.27			
27	Bevart L. S.	ND	T	27.5	1.64	2.18	.37		6.4	0.52
			B	21.0	1.78	2.15	.26			
28	Belding Fine S. L.	D	T	3.9	1.59	1.36	.29	23.94	6.4	0.98
			B	8.8	1.78	1.78	.42	19.69		
		ND	T	34.4	2.22	2.24	.45	31.25	6.2	0.82
			B	25.6				18.94		
29	Gera S. L.	D	T	5.0	1.88	1.18	.26	22.00	6.8	0.94
			B	10.1	2.22	1.54	.48	18.50		
		ND	T	13.9	2.14	1.90	.43	29.56	6.9	0.90
			B	11.7				19.75		
30	Marlette L.	D	T	6.4	1.81	1.18	.28	21.94	7.1	0.70
			B	12.5	1.99	1.22	.38	20.56		
		ND	T	26.3	2.01	1.53	.37	28.63	6.5	0.76
			B	20.4	2.30	1.08	.42	20.69		
31	Montcalm L. S.	D	T	5.4	1.88	.92	.29		7.1	0.54
			B	10.9	1.90	1.02	.41			

APPENDIX 1--Continued

			Plant Material						Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	B p.p.m.
32	Montcalm L. S.	D	T	4.4	1.76	1.20	.31		6.7	0.60
			B	10.3	1.89	1.25	.42			
33	London L.	D	T	10.7	1.98	1.16	.31	20.56	6.9	0.72
			B	15.8	2.02	1.56	.40	20.44		
		ND	T	28.4	2.19	1.58	.34	30.19	7.0	0.70
			B	22.4	2.42	1.22	.38	19.81		
34	McBride S. L.	D	T	11.7	2.06	1.07	.26	17.75	6.1	0.72
			B	19.4	2.23	1.27	.36	18.94		
		ND	T	28.4	2.66	1.38	.33	27.56	5.5	0.56
			B	21.3	2.56	1.14	.33	20.13		
35	Fox S. L.	D	T	9.8				20.56	5.8	0.64
			B	14.8	2.20	1.51	.25	19.56		
		ND	T	26.1	2.02	1.76	.46	24.81	5.5	0.72
			B	23.4	2.14	1.65	.37	18.44		
36	Fox S. L.	D	T	11.7	1.76	.82	.28		6.2	0.70
			B	20.2	2.12	1.50	.43			
37	Coloma L. S.	D	T	8.3	2.34	.79	.35		6.7	0.54
			B	9.3	2.38	.78	.53			
38	Coloma L. S.	D	T	6.3	1.80	1.05	.34		5.8	0.80
			B	11.8	2.04	1.33	.45			
39	Oshtemo L. S.	D	T	6.1	1.69	1.18	.37		5.6	0.72
			B	12.7	1.61	1.14	.35			

APPENDIX 1--Continued

			Plant Material						Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	B p.p.m.
40	Coloma	D	T	11.0	2.76	.94	.31	20.63	6.9	0.86
	L. fine S.		B	14.8	2.42	.96	.29	18.19		
		ND	T	27.5	2.35	1.20	.37	22.63	6.2	0.76
			B	26.9	2.40	1.03	.37	19.06		
41	Kalamazoo	D	T	17.3	1.56	.98	.31		6.0	0.49
	L.		B	17.3	1.45	1.15	.30			
42	S. L.	ND	T	40.0	2.21	2.09	.31		6.7	0.80
			B	25.6	1.67	2.18	.27			
43	Coloma	ND	T	25.6	2.19	1.37	.40		6.6	0.36
	L. S.		B	22.4	1.83	1.24	.38			
44	Coloma	D	T	28.4	1.16	1.88	.19	17.75	6.9	0.66
	L. S.		B	30.3	1.60	2.32	.23	18.06		
		ND	T	31.2	1.30	2.43	.26	27.81	6.5	0.64
			B	27.1	1.25	2.39	.18	20.63		
45	Coloma	ND	T	29.9	1.71	1.30	.43		6.0	0.48
	L. S.		B	26.2	1.37	1.30	.35			
46	Isabella	ND	T	29.6	2.01	2.14	.36		6.8	0.87
	S. L.		B	22.1	1.74	1.92	.26			
47	Isabella	ND	T	33.0	1.57	2.10	.32		6.5	0.51
	S. L.		B	26.7	1.52	1.79	.28			
48	Melita	ND	T	26.6	2.08	1.84	.37		6.7	0.92
	S.		B	28.7	1.67	1.75	.33			

APPENDIX 1--Continued

Location	Soil Type	Deficiency Status	Plant Material: Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	Soils: pH	B p.p.m.
49	Melita	ND	T	33.1	1.44	2.14	.24		6.4	0.66
	L. S.		B	26.5	1.23	2.24	.20			
50	Iosco	ND	T	42.5	1.61	1.91	.45		6.2	0.82
	L. S.		B	31.1	1.42	1.83	.39			
51	Kent	D	T	16.9	1.66	1.34	.27	18.31	6.9	0.86
	L.		B	20.8	1.78	1.66	.33	18.44		
		ND	T	31.3	1.92	1.72	.31	28.81	6.9	0.88
			B	26.0	1.75	1.55	.27	19.56		
52	Melita	ND	T	34.3	1.77	1.78	.33		6.2	0.44
	S.		B	31.3	1.86	1.14	.26			
53	Menominee	ND	T	36.2	2.14	1.75	.27		6.9	0.81
	L. S.		B	27.0						
54	Montcalm	ND	T	28.2	2.34	1.71	.48		6.2	0.58
	L. S.		B	30.0	2.11	1.32	.44			
55	McBride	ND	T	22.8	1.69	2.10	.27		7.0	0.55
	S. L.		B	21.7	1.70	2.28	.25			
56	McBride	ND	T	32.2	1.89	2.90	.37		6.4	0.92
	S. L.		B	22.5	1.62	2.84	.28			
57	Mancelona	D	T	8.8	2.84	1.06	.33		7.0	0.73
	L. S.		B	12.9	2.79	1.09	.50			

APPENDIX 1--Continued

			Plant Material						Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	p.p.m.
58	Bronson S. L.	D	T	7.0	2.08	1.25	.30	23.81	6.7	0.72
			B	8.6	2.36	1.40	.51	21.69		
		ND	T	14.0	2.82	1.32	.46	31.00	6.7	1.00
			B	18.2	2.44	.83	.34	19.38		
59	Hillsdale S. L.	D	T	8.3	1.46	1.63	.30	21.00	7.2	0.50
			B	12.7	1.49	2.23	.34	21.50		
		ND	T	34.2	2.02	2.24	.39	30.25	6.7	0.54
			B	26.0	1.78	1.83	.34	21.69		
60	Hillsdale Fine S. L.	D	T	3.6	1.52	1.09	.29		6.5	0.66
			B	12.1	1.71	1.07	.37			
61	Brookston Si. L.	ND	T	34.4	1.40	2.27	.32		6.3	1.30
			B	38.2	1.57	2.11	.36			
62	Wisner Si. L.	ND	T	32.2	1.45	2.31	.37		6.8	0.70
			B	24.1	1.36	1.72	.35			
63	Nester L.	ND	T	30.9	1.24	2.13	.27		6.2	0.72
			B	26.4	1.28	1.90	.27			
64	Nester S. L.	ND	T	21.7	1.44	1.90	.34		6.9	0.60
			B	19.6	1.53	1.34	.41			
65	McBride Fine S. L.	D	T	5.7	1.92	1.33	.44	20.81	6.7	0.78
			B	12.1	2.30	1.63	.49	19.69		
		ND	T	29.8	2.34	1.62	.47	25.69	6.8	0.74
			B	23.9	2.51	1.41	.55	19.81		

APPENDIX 1--Continued

			Plant Material							Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %		pH	p.p.m.
66	Allendale S.	ND	T	17.1	1.21	2.36	.40			6.6	0.54
			B	18.3	1.03	2.26	.29				
67	Emmet S. L.	ND	T	30.8	1.45	1.81	.23			6.3	0.48
			B	25.2	1.28	1.95	.17				
68	Selkirk Si. L.	ND	T	24.0	1.71	1.72	.30			6.8	0.80
			B	20.3	1.69	1.70	.28				
69	Montcalm L. S.	ND	T	29.3	2.14	1.53	.30			6.7	0.64
			B	25.0	2.03	1.57	.31				
70	Emmet L. S.	ND	T	28.4	1.89	1.71	.34			6.0	0.54
			B	24.2	1.53	1.77	.28				
71	Mancelona S. L.	ND	T	33.4	2.33	1.59	.33			6.4	0.62
			B	28.2	1.97	1.25	.28				
72	Kalkaska S.	D	T	11.0	1.44	1.28	.21			6.5	0.40
			B	19.0	1.84	1.69	.31				
73	Emmet L. S.	ND	T	27.5	3.00	1.60	.39			7.0	0.94
			B	22.1	3.00	1.57	.55				
74	Emmet S. L.	ND	T	39.2	2.18	2.18	.33			6.0	0.62
			B	32.8	1.94	1.94	.40				
75	Karlin Fine S. L.	ND	T	25.8	2.24	1.69	.36			6.1	0.60
			B	19.6	2.18	1.45	.34				

APPENDIX 1--Continued

			Plant Material						Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %	pH	B p.p.m.
76	Montcalm L. S.	ND	T	28.6	1.93	1.90	.37		6.6	0.40
			B	23.1	1.78	1.92	.41			
77	Montcalm L. S.	ND	T	19.2	1.81	1.46	.19		6.7	0.42
			B	20.1	1.78	1.34	.19			
78	Allendale S. L.	D	T	7.0	2.11	1.21	.37	20.81	6.6	0.78
			B	10.6	2.22	1.28	.53	18.69		
		ND	T	24.8	1.56	2.31	.29	26.06	6.4	0.72
			B	23.8	1.78	1.87	.29	17.63		
79	Nester C. L.	ND	T	23.9	1.52	2.48	.46		5.4	0.78
			B	22.5	1.33	2.02	.43			
80	Nester L.	ND	T	23.6	1.91	2.02	.35		6.7	0.96
			B	20.5						
81	Nester L.	ND	T	32.9	1.38	3.35	.30		7.0	1.28
			B	27.8	1.18	3.40	.24			
82	L.	ND	T	30.6	1.94	1.88	.41		6.8	1.00
			B	26.4	1.66	1.68	.37			
83	Miami L.	D	T	8.7	1.71	.83	.26		6.7	1.24
			B	10.7	2.12	.83	.49			
84	Hillsdale S. L.	D	T	4.4	1.93	1.15	.28	20.88	6.4	0.60
			B	9.4	2.03	1.18	.37	19.13		
		ND	T	25.7	2.44	1.15	.42	28.06	6.4	0.64
			B	11.9	2.28	.96	.44	20.38		

APPENDIX 1--Continued

			Plant Material							Soils	
Location	Soil Type	Deficiency Status	Plant Parts	B p.p.m.	Ca %	K %	Mg %	Protein %		pH	B p.p.m.
85	Hillsdale S. L.	D	T	6.8	1.52	1.21	.17			6.5	0.80
			B	9.0	1.85	1.55	.27				
86	Oshtemo L. S.	D	T	17.7	2.10	.78	.27			6.8	0.38
			B	18.8	2.16	.93	.37				
87	Coloma L. S.	D	T	7.1	1.67	1.02	.16			6.4	0.36
			B	10.0	1.87	1.08	.19				
88	Fox S. L.	D	T	11.3	1.46	.69	.17			6.2	0.55
			B	13.8	1.62	.86	.25				
89	Hillsdale S. L.	D	T	7.3	2.14	1.28	.25			6.0	0.74
			B	10.2	2.45	1.73	.29				

APPENDIX II

LEGAL DESCRIPTION OF LOCATIONS SAMPLED DURING THE SURVEY TRIP AND PLOT LOCATIONS USED IN FIELD EXPERIMENT II

Location1/	County	Town and Range	Section
1	Ingham	T 4 N, R 1 W	21
2	Ingham	T 4 N, R 1 E	36
3	Ingham	T 2 N, R 2 E	10
4	Livingston	T 1 N, R 3 E	23
5	Livingston	T 1 N, R 4 E	18
6	Washtenaw	T 2 S, R 4 E	17
7	Washtenaw	T 3 S, R 4 E	22
8	Lenawee	T 7 S, R 5 E	8
9	Lenawee	T 8 S, R 5 E	28
10	Lenawee	T 9 S, R 5 E	1
11	Monroe	T 9 S, R 6 E	6
12	Monroe	T 6 S, R 9 E	20
13	Oakland	T 2 N, R 10 E	9
14	Oakland	T 3 N, R 11 E	36
15	Macomb	T 3 N, R 12 E	10
16	Macomb	T 4 N, R 13 E	32
17	Macomb	T 4 N, R 13 E	36
18	Macomb	T 4 N, R 14 E	23
19	Sanilac	T 10 N, R 14 E	22
20	Sanilac	T 13 N, R 14 E	32
21	Sanilac	T 13 N, R 14 E	29
22	Huron	T 15 N, R 13 E	15
23	Huron	T 18 N, R 12 E	34
24	Huron	T 18 N, R 11 E	4
25	Tuscola	T 13 N, R 10 E	30
26	Tuscola	T 13 N, R 10 E	34
27	Tuscola	T 12 N, R 11 E	4
28	Tuscola	T 12 N, R 11 E	16
29	Tuscola	T 11 N, R 11 E	5
30	Lapeer	T 9 N, R 10 E	1
31	Lapeer	T 9 N, R 10 E	30
32	Genesee	T 9 N, R 8 E	20
33	Saginaw	T 9 N, R 2 E	15
34	Shiawassee	T 8 N, R 1 E	14
35	Shiawassee	T 8 N, R 1 E	27
36	Shiawassee	T 7 N, R 1 E	28
37	Ingham	T 4 N, R 1 W	12
38	Ingham	T 4 N, R 1 W	16
39	Ingham	T 4 N, R 1 W	29

1/ Arabic numerals denote locations sampled during survey trip. Roman numerals denote locations used in Field Experiment II.

APPENDIX II--Continued

Location	County	Town and Range	Section
40	Eaton	T 3 N, R 3 W	18
41	Kalamazoo	T 2 S, R 12 W	35
42	Van Buren	T 4 S, R 15 W	11
43	Cass	T 5 S, R 15 W	13
44	Van Buren	T 4 S, R 13 W	4
45	Ottawa	T 6 N, R 14 W	6
46	Ottawa	T 7 N, R 14 W	2
47	Muskegon	T 9 N, R 14 W	24
48	Muskegon	T 10 N, R 16 W	26
49	Muskegon	T 12 N, R 17 W	19
50	Oceana	T 13 N, R 16 W	9
51	Oceana	T 14 N, R 16 W	29
52	Mecosta	T 13 N, R 9 W	2
53	Mecosta	T 13 N, R 8 W	2
54	Montcalm	T 12 N, R 7 W	4
55	Montcalm	T 11 N, R 6 W	28
56	Montcalm	T 10 N, R 5 W	21
57	Montcalm	T 9 N, R 5 W	36
58	Clinton	T 5 N, R 4 W	9
59	Eaton	T 1 N, R 5 W	21
60	Shiawassee	T 5 N, R 2 E	10
61	Genesee	T 7 N, R 5 E	18
62	Bay	T 17 N, R 4 E	2
63	Arenac	T 19 N, R 3 E	13
64	Arenac	T 20 N, R 4 E	27
65	Ogemaw	T 24 N, R 3 E	18
66	Oscoda	T 27 N, R 3 E	31
67	Alpena	T 30 N, R 6 E	30
68	Alpena	T 31 N, R 7 E	30
69	Montmorency	T 30 N, R 4 E	11
70	Otsego	T 30 N, R 1 W	20
71	Antrim	T 31 N, R 5 W	23
72	Emmet	T 38 N, R 5 W	23
73	Charlevoix	T 33 N, R 8 W	7
74	Grand Traverse	T 25 N, R 12 W	1
75	Grand Traverse	T 25 N, R 12 W	27
76	Manistee	T 23 N, R 15 W	26
77	Manistee	T 22 N, R 15 W	7
78	Mason	T 18 N, R 16 W	16
79	Osceola	T 17 N, R 10 W	7
80	Isabella	T 16 N, R 4 W	2
81	Isabella	T 13 N, R 4 W	11
82	Clinton	T 8 N, R 2 W	9
83	Ingham	T 2 N, R 1 W	28

APPENDIX II--Continued

Location	County	Town and Range	Section
84	Ingham	T 1 N, R 1 W	21
85	Jackson	T 4 S, R 1 W	23
86	Branch	T 6 S, R 7 W	34
87	St. Joseph	T 8 S, R 9 W	3
88	Calhoun	T 4 S, R 6 W	24
89	Jackson	T 1 S, R 3 W	16
I	Tuscola	T 12 N, R 10 E	16
II	Tuscola	T 13 N, R 11 E	30
III	Oscoda	T 27 N, R 2 E	25
IV	Clare	T 18 N, R 4 W	22
V	Montcalm	T 12 N, R 6 W	25
VI	Wexford	T 22 N, R 11 W	26
VII	Newago	T 13 N, R 14 W	33
VIII	Allegan	T 2 N, R 14 W	12
IX	Van Buren	T 4 S, R 15 W	21
X	Ingham	T 4 N, R 1 W	13
XI	Washtenaw	T 1 S, R 3 E	2
XII	Branch	T 6 S, R 7 W	23
XIII	Calhoun	T 2 S, R 6 W	12
XIV	Jackson	T 4 S, R 2 W	26