

EFFECT OF NUTRITION AND REACTION ON THE GROWTH  
OF ALFALFA ON ACID SOILS

by

CLARENCE EDWARD CREWS

B. S., Kansas State Agricultural College, 1928

---

A THESIS

submitted in partial fulfillment of the

requirements for the degree of

MASTER OF SCIENCE

KANSAS STATE AGRICULTURAL COLLEGE

1930

## TABLE OF CONTENTS

	Page
INTRODUCTION.....	3
REVIEW OF LITERATURE.....	4
Nutrition.....	4
Calcium Carbonate.....	4
Phosphates.....	7
Nitrates.....	7
Potassium.....	8
Sulphur.....	9
Reaction.....	9
MATERIALS AND METHODS.....	12
Series A (Muck Soil).....	17
Series B (Sandy Silt Soil).....	21
Series C (Sandy Soil).....	25
EXPERIMENTAL RESULTS.....	25
Series A.....	25
Series B.....	26
Series C.....	28
CONCLUSION AND DISCUSSION.....	29
SUMMARY.....	30
ACKNOWLEDGMENT.....	30
LITERATURE CITED.....	31

## INTRODUCTION

Alfalfa acreage in Kansas has decreased fifty percent in the last 15 years. This has been brought about by two factors to a considerable extent. 1. Large acreages were plowed up during the war. 2. There has been a large percentage of failures to obtain stands in recent years. Many of these poor stands are due to unfavorable soil conditions.

An increase in acreage of alfalfa is needed not only because of the value of the crop for hay, but because of its soil improvement qualities, when used in rotation with grain crops.

In eastern Kansas where alfalfa is more generally adapted because of higher rainfall there are considerable areas of acid soils. Alfalfa is not ordinarily regarded as an acid tolerant plant, however there is some evidence that it will grow under comparatively acid conditions when nutrition requirements are fulfilled. This experiment was designed to investigate the combined effect of very acid soils (pH4-5) and certain fertilizers on growth and nodulation of alfalfa.

## REVIEW OF LITERATURE

### Nutrition

The literature does not agree very well as to the effect of nutrition upon nodulation, which in turn affects the growth of the legume. Hiltner (13) claimed that a plant well supplied with nutrients resists the entrance of bacteria into the root hairs. Later investigations by Perkins (25) found that the elements essential for plant growth do not directly affect the nodulation of legumes. McTaggart (22) finds: "In general it may be said that when any application of fertilizer, with the exception of Gypsum, increased the yield of the legumes grown, there was also an increase in the percentage of nitrogen in the plants."

Calcium Carbonate. Hopkins advises the use of lime and inoculated soil to secure a good growth of alfalfa in Illinois. In 1904 he showed the importance of inoculation and states that the organism found on sweet clover roots will inoculate alfalfa. Moore (23) noted the variable effect of calcium and magnesium under both alkaline and acid conditions, stating that the effect depends upon the soil reaction. Kellerman and Robinson (17) grew alfalfa on a poor sandy soil which had grown alfalfa previously. They found that the addition of lime and artificial inoculation

either together or singly resulted in about the same degree of nodule formation. Plants that were grown in soil that was neither limed or inoculated did not produce nodules. They explained this by saying that probably alfalfa bacteria of low vitality were present in the soil, but under the existing unfavorable soil conditions they were unable to infect the plants. However when this soil was limed the native bacteria were able to produce nodules as effectively as the pure cultures. Morse (24) found that liming caused an increase in assimilation of nitrogen by clover plants apparently by its action on the properties of the soil and not by its action within the plant. Wilson (33) observed that calcium compounds were in most cases effective in stimulating nodulation with the carbonate radical showing no effect whatever. Fellers (8) from both field and pot experiments which he conducted in acid silt loam soil concluded that enough calcium carbonate to neutralize one-half the acidity resulted in the production of as many nodules as did higher applications. Lipman and Blair (20) working with soybeans, and Karraker working with red clover observed that nodules were more numerous and better distributed on soil where limestone had been used. Lipman and Blair (20) found an increase in the percentage of nitrogen in the beans as well as large increases in yield from liming soybeans. Graul and

Fred (12) found that alfalfa and related plants high in calcium can be grown on very acid soils provided a fraction of the acidity is neutralized; and that it is not a question of how much acidity there is in the soil, but how much lime carbonate there is available for the metabolism of the plant. Scanlan (29) found that limestone increased nodulation greatly in acid soils in all cases where used. Albrecht and Davis (2) found that the effect of calcium in stimulating inoculation failed to show a significant correlation with the hydrogen ion concentration or to electrodialyzable calcium in the soil in the cases they studied. Prucha (27) found that calcium chloride inhibited nodule formation on Canada field peas when grown in water culture. Scanlan (28) found that in pot cultures of three different types of acid soils, the addition of a small amount of a neutral calcium salt, either calcium chloride or calcium acetate, sprinkled on the seeds gave fully as good nodule production on soybean as did the addition of calcium carbonate; thus the effect of calcium is not due to an alteration in the reaction but may be due to some physiological effect upon the bacteria or the host plant. Scanlan (29) found that calcium chloride applied at the rate of 1:1500 greatly benefited the viability and the ability to infect by *B. radicicola* when stored in tap water or soil extract.

Phosphates. Increase in nodulation by use of phosphates has been noted by many workers and is generally accepted. Wilson (33) reported that phosphates and calcium salts stimulate nodulation of soybeans. Fellers (8) concluded from field experiments that acid phosphate stimulated nodule formation and materially increased yields of soybeans especially on well limed soils. McTaggart (22) found that phosphorus showed the most marked beneficial effect of any fertilizer used, increasing the dry matter, total nitrogen, and the per cent of nitrogen. Perkins (25) found that phosphates were not necessary for nodulation.

Nitrates. Moore (23) stated that alkaline nitrates in a concentration of 1:100,000 prevent the formation of nodules, and that fertile soils have less nodules than infertile soils. Prucha (27) observed that potassium nitrate inhibited nodulation. Wilson (33) found that nitrates and ammonia-containing salts depress nodule formation of soybeans. He found that 16 out of 18 different nitrates added to the soil in appreciably below the amount necessary to cause injury to the plant reduced or entirely prevented nodule production. Fellers (8) found that nitrate of soda increased the yield but inhibited the nodulation of soybeans. Albrecht (1) found that nodules were produced in the presence of large amounts of either nitrate nitrogen or organic nitro-

gen. McTaggart (22) found that sodium nitrate depressed and in quantities more than five hundred pounds per acre entirely prevented nodulation. Giobel (11) found that some combined nitrogen was desirable and highly beneficial for maximum infection and nodule development, as well as for maximum yields of legumes. This was found to be a necessity with alfalfa until established, then the plants fixed large amounts of nitrogen. His results show that alfalfa fixes very little nitrogen during the first crop before the flowering stage but after this fixation takes place rapidly. Larger yields were always obtained when combined nitrogen was added but there never was a higher per cent of nitrogen to dry matter. Sometimes the plants which received no combined nitrogen had the highest percentage of nitrogen to dry weight. He found that in general the amount of nitrogen fixed was in close correlation with the volume of the nodules but not with the number of nodules. The amount of nitrogen fixed was inversely proportional to the amount of soluble combined nitrogen at the disposal of the plant. Nodule development was adversely affected by the presence of large amounts of  $\text{NO}_3$  in soil or sand cultures.

Potassium. Prucha (27) found potassium hydroxide increased nodulation of Canada field pea. Fellers (8) found that muriate of potash slightly stimulated nodule production



on the limed plots but not on the unlimed ones. McTaggart (22) reported that potassium salts increased the dry matter and nitrogen content for both alfalfa and Canada field peas. Perkins (25) found that fertilizers containing potassium were almost without effect on nodulation of Virginia soybeans.

Sulphur. Duley (7) growing red clover in soil in pots showed that nodule production was greatly increased by application of elemental sulphur alone or with other fertilizers. Prucha (27) found that calcium sulphate or magnesium sulphate exerted a beneficial influence on nodule formation on the Canada field pea grown in water cultures. Pitz (26) grew red clover in potted Miami silt loam soil to which applications of sulphur were made and found no effect on nodule production. With the application of calcium sulphate nodule production and root development were both inhibited. Wilson (33) used ten sulphur containing compounds and found that all but manganese sulphate and potassium sulphate inhibited nodule production. Fellers (8) noted that sulphur and gypsum ( $\text{CaSO}_4$ ) greatly stimulated nodule production on soybeans. McTaggart (22) found that sulphur in the form of gypsum increased somewhat the growth and nitrogen content of alfalfa.

#### Reaction

Effect of reaction on the growth and nodulation of leg-

umes is a problem that has been responsible for much research work and the results have been variable. Fred and Loomis (10) have shown that when the acidity of the media reaches a certain degree the multiplication of alfalfa bacteria is greatly lessened. Fred and Davenport (9) found from experiments which they conducted that a correlation exists between the acid resistance of legume bacteria and the acid resistance of the host plant. Fellers (8) in a summary of his work on the factors affecting nodulation states: The bacterial infection of roots does not take place readily on acid soils even when the root infecting organisms are plentiful in the soil. Truog (32) says, "that under an acid soil condition available soil phosphorus eventually appears to become less available." Stoddard (31) expressed the opinion that this decrease in availability is due to the gradual transformation of the calcium phosphate into iron and aluminum phosphates; in which condition the phosphorus becomes less and less soluble. Truog (32) is of the opinion that acid soils may be as detrimental to the legume plant as to the organism and offers as proof the effect of acid soils on some non-legume crops. Jaffe (16) found poor germination of alfalfa in acid soils having a pH below 4.5, but he found that after the plants once became established they showed normal green color, high vigor, and made excellent growth in soil having a pH as low as 3.8. He obtained considerable

nodulation at a pH 4, (using Colormetric methods to determine the acidity from a water extract of the soil.) Bryan (5) was unable to get alfalfa seedlings to establish themselves at a pH 4. Bryan (6) reports that alfalfa bacteria stored for 75 days in different types of acid soils were killed at a pH of 5, regardless of the type of soil and that the critical H-ion concentration was the same for pure cultures. Alway and Nesom (4) found that when a lime deficient soil has not been limed the soil transfer method of inoculation was more effective than artificial culture for the first seeding. It is possible that soil transfer was more effective because it carried a strain of bacteria more tolerant to acid soils. This seems to indicate an inability of the organism to establish themselves quickly on a lime deficient soil. Karraker (19) concludes that the effect of acid soil is on the nodule bacteria directly and not on the host plant. Scanlan (29) reports that soil reaction was not the controlling factor on nodulation of soybeans. He did not give the pH encountered. Sewell and Gainey (30) found that alfalfa grew well at a pH of 4.5 provided it was supplied with nutrients including calcium. Five plants grown in water cultures maintained at a pH 4 had but one small cluster of nodules. At pH 6 there were few points of inoculation but they were large grape like clusters at pH

7.5, there were many small clusters of nodules well distributed. When alfalfa was grown in pots in south eastern Kansas soil, which is lime deficient and highly acid, no nodules were produced in the inoculated unlimed checks. Lime alone produced a few nodules. Phosphorus alone produced more nodules than lime. Phosphorus and lime produced good nodulation and growth.

#### MATERIALS AND METHODS

Pot experiments were conducted in the greenhouse on three types of very acid North Carolina soils.

Type A is a muck soil pH 4.1 with 5000 pounds total N in surface 7 inches.

Type B is a sandy silt soil pH 4.9 with 1800 pounds total N in surface 7 inches.

Type C is a sandy soil pH 4.6 with 700 pounds total N in surface 7 inches.

These soils are all low in fertility and not very productive. None of them had ever grown alfalfa or sweet clover. There were twelve treatments on type A, and thirteen treatments on each of type B and C. Each treatment was replicated ten times, making a total of 380 pots. Six inch unglazed, clay pots were used and a definite amount of soil was placed in each. Owing to the variation in the volume weight of these soils the quantities necessary to fill the

six inch pots with air dry soil were:

Type A 1800 grams

Type B 2100 grams

Type C 2500 grams

The soil was sifted and mixed uniformly before being placed in the pots.

The reactions of the soils were determined with the Quinhydrone electrode and are expressed as pH. The results of the Quinhydrone pH determinations in Tables I, II, and III are averages of the readings for the ten replications of each treatment. The samples of soil for determining reaction were well mixed and pH readings made immediately.

Fertilizer treatments were applied in the following manner: Calcium carbonate, calcium chloride, and calcium acid phosphate were mixed in the upper two and one-half inches of soil with a spatula while potassium sulphate and sodium nitrate were dissolved and the solution poured on the soil before watering.

Distilled water was supplied to the plants in sufficient quantities for growth but not enough for any leaching of the soil to occur. The pots were weighed occasionally to check the moisture content.

Kansas Common alfalfa seed was used, the seed for all treatments was inoculated except treatment twelve in each series.

Table I. Summarized data. Series A

Soil and treatment	pH (Quin- hydrone) Jan. 30, 1930. Av. 10 deter- minations	pH (Quin- hydrone) Mar. 5, 1930. Av. 10 deter- minations	Wt. of dry plants Av. of 10 pots	Nodulation	
				Periphery	Center
AT1 Check	4.1			none	none
AT2 N	4.2			none	none
AT3 PO <sub>4</sub>	4.1	4.4	3.6	few	none
AT4 K <sup>4</sup>	4.1			none	few
AT5 PO <sub>4</sub> 90 gms.	4.6	4.5	11.3	moderate	none
AT6 K and PO <sub>4</sub>	4.2	4.1	3.3	many	none
AT7 K and PO <sub>4</sub> 2.6 gms.	4.1	4.2	4.5	few	none
AT8 K and PO <sub>4</sub> 18.8 gms.	4.3	4.2	15.4	many	moderate
AT9 K and PO <sub>4</sub> 36.8 gms.	4.2	4.1	11.7	moderate	none
AT10 N, K and PO <sub>4</sub>	4.4	4.1	3.	moderate	none
AT11 CaCl <sub>2</sub> 2. gms.	4.2	4.1	1.	none	none
*AT12 N, K, and PO <sub>4</sub>	4.1	4.1	3.7	moderate	few
AT14 PO <sub>4</sub> 0.8 gm. <sup>4</sup>		4.3		few	none
AT15 KH <sub>2</sub> PO <sub>4</sub> 0.91 gm.		4.6		few	none
AT16 KC <sub>2</sub> H <sub>3</sub> O <sub>2</sub> 0.66 gm.		4.5		few	none
AT17 Ca(C <sub>2</sub> H <sub>3</sub> O <sub>2</sub> ) <sub>2</sub> 19.3 gms.		5.8		many	many
AT18 Ca(C <sub>2</sub> H <sub>3</sub> O <sub>2</sub> ) <sub>2</sub> 32.2 gms.		6.5		many	many
AT19 Ca(C <sub>2</sub> H <sub>3</sub> O <sub>2</sub> ) <sub>2</sub> 51.5 gms.		6.8		many	many

\* Seed not inoculated



Plate I. Alfalfa plants at the age of two months with various fertilizer treatments on an acid muck soil



Plate II. Alfalfa plants at the age of two months with various fertilizer treatments on an acid muck soil



### Series A (Muck Soil)

The variations in treatment and observations for soil A are recorded in Table I. Former investigations made by Dr. P. L. Gainey, have shown that 0.17 per cent  $\text{CaCO}_3$  mixed with this soil will not affect the reaction while 0.1 per cent  $\text{CaCO}_3$  will change the reaction only a very small amount and 5 per cent  $\text{CaCO}_3$  will not neutralize it. It is a very highly buffered soil. Sodium nitrate was applied at the rate of 0.8 gram per pot which is equivalent to 100 pounds of nitrogen per acre. The superphosphate application is equivalent to 600 pounds superphosphate per acre and the potassium sulphate equals a 300 pound rate per acre of  $\text{K}_2\text{SO}_4$ .

All the plants in treatments one and two were stunted and gradually died until there were none left at the end of two months. They were then discarded. Treatment four was discarded at the end of two months as there were only five small stunted plants living. Two of these five plants were found to have numerous small nodules on the roots.

The soil from the discarded treatments one, two and three was mixed into a composite mass and placed into pots; each pot contained 1800 grams as before. The treatments were numbered 14 to 19 inclusive and each was replicated five times.

Table II. Summarized data. Series B

Soil and treatment		pH (Quin- hydron) Jan. 30, 1930. Av. 10 deter- minations	pH (Quin- hydron) Mar. 5, 1930. Av. 10 deter- minations	Wt. of dry plants Av. of 10 pots	Nodulation	
					Periphery	Center
BT1	Check	5.	4.5	2.9	few	none
BT2	N	4.9	4.7	2.8	very few	none
BT3	PO <sub>4</sub>	4.9	4.7	8.2	many	none
BT4	K	4.3	4.4	2.4	many	none
BT5	Ca 2.1 gms.	7.4	7.1	10.9	many	many
BT6	K and PO <sub>4</sub>	4.2	4.7	9.8	many	few
BT7	K and PO <sub>4</sub> 2.1 gms.	4.5	4.6	11.9	many	none
BT8	K and PO <sub>4</sub> 12.6 gms.	4.5	4.3	13.	many	none
BT9	K and PO <sub>4</sub> 21. gms.	4.7	4.4	14.	few	few
BT10	N, K, PO <sub>4</sub>	4.5	4.4	9.3	many	none
BT11	CaCl <sub>2</sub> 3.0 gms.	4.6	4.2		Plants died	
*BT12	N, K, and PO <sub>4</sub>	4.6	4.6	9.8	few	none
BT13	PO <sub>4</sub> and Ca 21. gms.	6.7	6.5	9.9	many	many

\* Seed not inoculated



Plate III. Alfalfa plants at the age of two months with various fertilizer treatments on a sandy silt soil



Plate IV. Alfalfa plants at the age of two months with various fertilizer treatments on a sandy silt soil

Treatment 14 was 0.8 gram of superphosphate or the equivalent of 600 pounds per acre. Treatment 15 was 0.91 gram  $\text{KH}_2\text{PO}_4$  which is equivalent phosphorus to that in treatment 14. Treatment 16 was 0.66 gram potassium acetate which is equivalent to the potassium in treatment 2. Treatment 17 was 19.3 grams calcium acetate. This is calcium equivalent to 3 tons  $\text{CaCO}_3$  per acre. Treatment 18 was 32.25 grams calcium acetate. This is calcium equivalent to 5 tons  $\text{CaCO}_3$  per acre. Treatment 19 was 51.5 grams calcium acetate per pot which is  $\text{CaCO}_3$  equivalent to 8 tons per acre.

The photographs of the plants in series A (Plate I and II) were made when the plants were two months old. There were some variations in the replications of each treatment, but the pots selected for photographing were as near respective as possible.

#### Series B (Sandy Silt Soil)

The variations in treatments and observations for soil type B are recorded in Table II.

The amount of calcium carbonate used in each treatment was calculated according to former investigations by Dr. Gainey as to lime requirements of this particular soil.

The calcium chloride was used to get a higher amount of available calcium with no change in acidity. The remaining treatments were the same as series A. The calcium carbonate

Table III. Summarized data. Series C.

Soil and treatment	pH (Quin- hydrone) Jan. 30, 1930. Av. of 10 de- terminations	pH (Quin- hydrone) Mar. 5, 1930. Av. 10 deter- minations	Wt. of dry plants Av. of 10 pots	Nodulation	
				Periphery	Center
CT1 Check	5.1	5.2	2.4	many	none
CT2 N	5.	5.1	3.2	many	none
CT3 PO <sub>4</sub>	4.6	4.8	7.2	many	none
CT4 K	4.6	4.7	3.1	many	none
CT5 Ca 12.5 gms.	7.6	7.1	7.9	many	many
CT6 K and PO <sub>4</sub>	4.8	5.2	9.4	many	few
CT7 K and PO <sub>4</sub> 1.25 gms.	5.2	4.9	8.3	many	none
CT8 K and PO <sub>4</sub> 2.5 gms.	5.9	5.2	10.0	many	few
CT9 K and PO <sub>4</sub> 12.5 gms.	5.2	4.9	7.2	many	few
CT10 N, K, and PO <sub>4</sub>	5.4	5.2	9.7	many	few
CT11 CaCl <sub>2</sub> 1.5 gms.	5.1	4.7	2.5	many	none
*CT12 N, K, and PO <sub>4</sub>	5.2	5.2	8.	many	few
CT13 PO <sub>4</sub> and Ca 12.5 gms.	7.	7.2	8.	many	many

\* Seed not inoculated



Plate V. Alfalfa plants at the age of two months with various fertilizer treatments on a sandy soil



Plate VI. Alfalfa plants at the age of two months with various fertilizer treatments on a sandy soil



was mixed in the lower half of the pots on treatment 13 and the inoculated seeds were planted in the untreated soil above.

The photographs of the plants in series B (Plate III and IV) were made when the plants were two months old. There were some variations in the pots of each treatment but the ones selected for photographing were as near representative as possible.

#### Series C (Sandy Soil)

The variations in treatments and observations for soil type C are recorded in Table III.

All the fertilizer treatments are the same as for soil A and B except the amount of calcium was reduced in accordance with the small amount of buffers in this soil.

The calcium carbonate in treatment 13 was mixed with the soil in the lower half of the pots and the inoculated seeds were placed in the untreated soil above.

### EXPERIMENTAL RESULTS

#### Series A

There were significant increases in both growth and nodulation by the use of large applications of superphosphate.

The plants getting phosphorus alone made the best

growth and had the best color of any in the series for the first month; then they gradually decreased in vigor and showed signs of chlorosis. About one-third of the plants died by the end of three months.

The plants getting phosphorus and potassium started growth a little slower but were about the same as the ones treated with phosphorus alone. The plants getting mono-calcium phosphate, and potassium sulphate were healthy throughout and showed the most size and vigor at the end of three months of any in this series.

Calcium, phosphorus, and potassium each seemed to stimulate nodule production. The plants that received phosphates, potassium sulphate, and sodium nitrate were healthy and made a fair growth but did not seem to have the vigor of the ones getting some calcium. Apparently nodulation was not repressed by the use of nitrates.

#### Series B

The plants that received no treatment were stunted and chlorotic, many of them died. There were no nodules formed.

The plants that were treated with sodium nitrate grew about the same as those with no treatment. The top roots went straight down, there was very little branching, few fibrous roots, and no nodules.

The phosphorus treatment grew healthy, fairly vigorous plants. There were numerous nodules around the edge of the pot but none in from the edge. The plants in treatment 4, ( $K_2SO_4$ ) were slightly better than those in treatment 1 and treatment 2, but not as good as those receiving phosphorus alone. The plants getting calcium alone were healthy and of good color but did not have as large an amount of growth as the ones getting phosphorus and potassium in addition. The calcium treated plants had numerous nodules on the roots throughout the pots and the fibrous roots were well distributed throughout the soil.

The phosphorus and potassium combination grew the best plants in this series. Plants were all of good color, thrifty, and vigorous. The ones in the pots treated with a medium application of superphosphate were the most vigorous. All the plants were well nodulated around the edges of the pots but very few nodules in the center.

The plants in the nitrate, phosphate and potassium treatment grew nearly as well at first but were soon surpassed by the phosphate and potassium treated plants. These plants were all healthy and vigorous and the roots were nodulated around the edges of the pots but not in away from the edge.

### Series C

Significant increases in growth was obtained by the use of both lime and phosphorus on this soil.

The plants in this soil were all well nodulated in the bottoms of the pots and around the edges of the pots but no nodules were found in the soil away from the edges except those receiving calcium carbonate; these were nodulated throughout.

There was very little or no increase in growth of those plants receiving nitrates or potassium alone. There was some increase, however, when they were used in combination with phosphates.

Calcium chloride had very little or no effect on the growth of alfalfa on this soil. The plants treated with calcium chloride alone were slow in germinating, very stunted and a large percent of them soon died.

With treatment 13 where the calcium carbonate and phosphate were mixed in the lower half of the pot, the plants started slowly but grew with more vigor later. These plants were well nodulated from top to bottom, but there were many more fibrous roots in the lower half of the pot than in the upper half.

## CONCLUSION AND DISCUSSION

The roots in series A, and to a less extent in series B, had quite a pronounced tendency to grow laterally to the edge of the pot, then follow down the side of the pot. The roots in the more acid or unfavorably treated pots contained no nodules until they came in contact with the pot wall, and from there down the side of the pot there were large grape-like clusters of nodules. There are several factors that may contribute to this condition.

1. The pots may have an influence on the reaction of the soil immediately in contact with the porous clay pot. With regard to this view, the particles of soil from several of the more acid pots were carefully scraped loose from the pot and the reaction of this sample of soil was found to vary from 0.3 to 1.0 pH toward neutrality.

2. These pots were not sterilized before starting this experiment, although they had been stored dry for a year and were washed previous to potting this soil. They may have been contaminated with *B. radiculicola* which were possibly more adapted to these surroundings than was the artificial culture used. There is evidence of this in that the uninoculated soil, which had never grown alfalfa or sweet clover, grew nodulated plants.

3. The excess moisture and aeration available between the soil and the pot wall are both known to be favorable to nodulation and may be largely responsible for these large grape-like clusters of nodules.

#### SUMMARY

There were significant increases in both growth and nodulation by the use of calcium or phosphorus alone on any of these types of acid soils. Growth and nodulation were best when lime and phosphorus were used in combination.

There seemed to be little or no effect on growth by the use of nitrates, potassium, or sulphates. Potassium sulphate stimulated nodulation slightly. Nitrates had no inhibiting effect on nodulation. Calcium chloride depressed growth on series A and B but had no effect on growth or nodulation on series C. Nodulation was more pronounced around the edges and the bottom of the pots than in the center in all except the high calcium applications.

#### ACKNOWLEDGMENT

The writer wishes to make acknowledgment of the helpful criticisms and suggestions made by his major instructor, Dr. M. C. Sewell, and to express appreciation of the interest and attention given by him throughout the progress of this work.

## LITERATURE CITED

1. Albrecht, W. A.  
1920. Symbiotic Nitrogen Fixation as Influenced by Nitrogen in the Soil. Soil Sci. 9:275.
2. Albrecht, W. A. and Davis, F. L.  
1929. Relation of Calcium to the Nodulation of Soybeans on Acid and Neutral Soils. Soil Sci. 28: No. 4.
3. Albrecht, W. A. and Davis, F. L.  
1929. Physiological Importance of Calcium in Legume Inoculation. Bot. Gazette LXXXVIII No. 3, Nov.
4. Alway, F. I. and Nesom, G. H.  
1927. Inoculation of Alfalfa on Lime-deficient Sandy Soils; Soil Transfer versus Use of Culture. Minnesota Agricultural Experiment Station Tech. Bulletin No. 46.
5. Bryan, O. C.  
1923. Effect of Reaction on Growth, Nodule Formation, and Calcium Content of Alfalfa, Alsike Clover and Red Clover. Soil Sci. 15:23-35.
6. Bryan, O. C.  
1923. Effect of Acid Soils on Nodule-forming Bacteria. Soil Sci. 15:37-40.
7. Duley, F. L.  
1916. Relation of Sulphur to Soil Productivity. Jour. Amer. Soc. Agron. Vol. 8, p. 154-160.
8. Fellers, C. R.  
1918. The Effect of Inoculation, Fertilizer Treatments, and Certain Minerals on Yield, Composition, and Nodule Formation of Soybeans. Soil Sci. 6:81-130.
9. Fred, E. B. and Davenport, A.  
1918. Effect of Reaction on Nitrogen Assimilating Bacteria. Jour. Agr. Res. 14:317.
10. Fred, E. B. and Loomis, N. E.  
1917. Influence of Hydrogen-ion Concentration. Jour. Bact. Vol. 2, p. 629-633.

11. Giobel, Gunnar.  
1926. The Relation of Soil Nitrogen to Nodule Development and Fixation of Nitrogen by Certain Legumes. New Jersey Exp. Sta. Bul. 436.
12. Gravel, E. J. and Fred, E. B.  
1922. The Value of Lime in Inoculation for Alfalfa and Clover on Acid Soils. Wisconsin Agr. Exp. Sta. Res. Bul. 54.
13. Hiltner, L.  
1900. Uber die Vrsachen Welche die Grosse Zahl Stellung and Wirking der Wurzelknollchen bedingen. Arb. K. Gsndhtsamt Bio. Abt. 28:1-47.
14. Hopkins, C. G.  
1902. Alfalfa on Illinois Soil. Illinois Exp. Sta. Bul. 76.
15. Hopkins, C. G.  
1904. Nitrogen Bacteria and Legumes. Illinois Exp. Sta. Bul. 94.
16. Joffe, J. S.  
1920. The Effects of Soil Reaction on Growth of Alfalfa. Soil Sci. 10:301-307.
17. Kellerman, K. F. and Robinson, T. R.  
1906. Conditions Affecting Legume Inoculation. U.S. D.A. Bur. of Plant Ind. Bul. 100:79.
18. Karraker, P. E.  
1914. The Effect of Liming on Acid Soil on Growth of Certain Legumes. M. A. Thesis U. of Mo. p. 56.
19. Karraker, P. E.  
1927. Production of Nodules on Different Parts of the Root System of Alfalfa Grown in Soils of Different Reaction. Soil Sci. 24:103-109.
20. Lipman, J. G. and Blair, A. W.  
1920. The Lime Factor in Permanent Soil Improvement. Soil Sci. 9:91-114.
21. Lipman, J. G. and Blair, A. W.  
1922. Influence of Lime on Yield and Nitrogen Content of Soybeans. New Jersey Exp. Annual Report 355.



22. McTaggart, A.  
1921. The Influence of Certain Fertilizer Salts on Growth and Nitrogen Content of Some Legumes. Soil Sci. 11:435.
23. Moore, Geo. T.  
1905. Soil Inoculation for Legumes. U.S. Bur. of Plant Industry Bul. 71:1-72.
24. Morse, F. W.  
1915. The Effect on a Crop of Clover of Liming the Soil. Mass. Agr. Exp. Sta. Bul. 161.
25. Perkins, A. T.  
1924. Effect of Several Mineral Fertilizers Upon Nodulation of Virginia Soybeans.
26. Pitz, W.  
1916. Effect of Elemental Sulphur and of Calcium Sulphate on Certain of the Higher and Lower Forms of Plant Life. Jour. Agr. Res. 5:771-780.
27. Prucha, M. J.  
1916. Physiological Studies of B. Radicicola of Canada Field Pea. N. Y. (Cornell) Agr. Exp. Sta. No. 5
28. Scanlan, R. W.  
1926. Physiological Studies of B. Radicicola of Soybean. M. A. Thesis, U. of Mo.
29. Scanlan, R. W.  
1928. Calcium as a Factor in Soybean Inoculation. Soil Sci. 25:313-327.
30. Sewell, M. C. and Gainey, P. L.  
1930. Unpublished Data, Kansas Exp. Sta.
31. Stoddard, C. W.  
1909. Soil Acidity in its Relation to Lack of Available Phosphates. Jour. Ind. Eng. Chem. Vol. 1 No. 2, p. 69.
32. Truog, E.  
1918. Soil Acidity Its Relation to the Growth of Plants. Soil Sci. 5:169-195.
33. Wilson, J. K.  
1917. Physiological Studies of B Radicicola of Soybean and Factors Influencing Nodule Formation. N. Y. (Cornell) Agr. Exp. Sta. Bul. 386.