ALFALFA GROWTH ON ACID SOIL
AS INFLUENCED BY
AL, CA, pH AND MO

by

John Edward Rechcigl

Dissertation submitted to the Faculty of the
Virginia Polytechnic Institute and State University
in partial fulfillment of the requirements for the
degree of

DOCTOR OF PHILOSOPHY
in
Agronomy

Approved:

R. B. Reneau, Jr. Co-chairman	W. Kroontje Co-chairman
D. C. Martens	J. L. Neal, Jr.
S. W. Van Scoyoc	D. D. Wolf

October, 1986
Blacksburg, Virginia

ALFALFA GROWTH ON ACID SOILS AS INFLUENCED BY Al, Ca, pH AND Mo

by

John Edward Rechcigl

Agronomy

(ABSTRACT)

Soil acidity is a major cause of low yields of alfalfa (Medicago sativa L.) in the southeastern United States. Two field experiments were conducted on an Ernest silt loam soil (fine-loamy, mixed, mesic Aquic Fragiudult) to determine what conditions are necessary for optimal alfalfa growth in an acid soil environment. In the first experiment alfalfa was no-till planted to evaluate the influence of surface applied dolomitic limestone at 0 and 6.7 Mg/ha either 8 or 20 months before planting and at planting on alfalfa performance under acidic conditions. Yield increases, greater than two fold, resulted from surface application of limestone regardless of time of application. In the second field experiment treatments included surface and incorporated dolomitic limestone at either 6.5 or 13.0 Mg/ha, gypsum at 13.0 Mg/ha, foliar Mo at 560 g/ha and N as a split. Yields increased in response to surface and incorporated

lime, Ca, Mo and N application, possibly as a result of Al activities in soil solution being as low as 0 and 0.05 uM in the surface and 1 m depth samples, respectively, for the surface limed soils. Low activity of Al in soil solution may explain why subsurface acidity was not toxic to alfalfa grown on these soils. This study showed that surface limestone at half the recommended rate is adequate for the growth of alfalfa under acidic conditions.

The implanted soil mass technique was used to evaluate the influence of subsurface amendments (Ca, N, Ca and N, KOH and $Ca(OH)_2$) on alfalfa root growth and N fixation on soils that received either 0 or 6.7 Mg/ha surface applied limestone. Of the various subsurface amendments the $Ca(OH)_2$ treatment produced the most root growth and N fixation. This study showed that both the reduction in acidity and the availability of Ca are necessary for optimal N fixation and root growth.

In vitro studies conducted in growth pouches showed that pH 4.5 nutrient solution was not detrimental to alfalfa growth unless Al was present. Addition of 3.0 mM Ca to a nutrient solution containing 0.08 mM Al alleviated toxic effects of Al on alfalfa growth. No-till alfalfa can thus be grown on acid soils with surface lime by providing Ca and reducing acidity.

ACKNOWLEDGEMENTS

The author wishes to express his sincerest appreciation to his major professors Dr. R. B. Reneau, Jr. and W. Kroontje who guided and assisted him throughout the program of study. Their wisdom and attention to the author in discussing research was extremely helpful. The author is also grateful to Drs. D. C. Martens, J. L. Neal, Jr. and S. W. Van Scoyoc for serving on his graduate committee. Special thanks is extended to Dr. D. D. Wolf who was particularly helpful and encouraging with his sound advice and extensive help with field and laboratory work.

Gratitude is extended to Lenoor Oosterhuis for dedicated laboratory assistance and the helpful advice she gave the author throughout his research program. Appreciation also goes to Kathy Molton for her help in soil classification. Thanks are also expressed to Allan Wolf for his help in the field and the laboratory. Acknowledgments are also extended to the Statistics Department for their valuable assistance in analyzing the data.

The author wishes to express his thanks to his fellow graduate students, i.e., Keith Edmisten, Sherri Buttrey, Phil Jardine, Jay Roberts, Bruce Rappaport, Dave

Little, Dave Parker, Dennis Dove, Stacy Gettier, Jim McKenna, Jim Fredericks and Garry Payne for their assistance, advice and friendship.

The author is greatly indebted to Diane Beckwith for her many hours of typing annual research reports, tables and graphs.

Much appreciation is extended to the author's family and his sister, for their continued support and constant encouragement during the course of this study.

The author extends special appreciation to his wife for her patience, understanding and moral support throughout the graduate program.

Last, but not least, special acknowledgement is due to T.V.A. for financial support.

LIST OF TABLES

TABLE PAGE

1 Alfalfa shoot and root growth as affected by pH and Al in nutrient solutions....................65

2 Alfalfa shoot and root growth after 1 and 2 weeks as affected by Ca and Al in acidic (pH 4.5) nutrient solutions....................................67

3 Alfalfa shoot and root chemical composition as affected by Ca and Al in acidic (pH 4.5) nutrient solutions...72

4 Influence of application date of surface-applied agricultural limestone on soil pH, exchangeable Al and Ca in an Ernest silt loam soil.................82

5 Influence of application date of surface-applied agricultural limestone on alfalfa yield and purity of stand...87

6 Influence of application date of surface-applied agricultural limestone on nutrient concentration and calculated N uptake of alfalfa herbage. Concentrations are averages of three hay harvests in 1984..89

7 Chemical composition of the implanted soil mass (Tatum clay loam + amendment) after 1 y of equilibrium in the field...............................98

8 Influence of surface applied limestone on soil pH, exchangeable Al, Ca and P in an Ernest silt loam soil 3 y after application.........................99

9 Influence of dolomitic lime, gypsum and foliar applied Mo on pH, exchangeable Al, Ca and Mn in an Ernest silt loam soil..........................112

10 Influence of dolomitic lime, gypsum, foliar applied Mo and N on alfalfa yields in 1985 and 1986 grown on an Ernest silt loam soil.............114

11 Influence of dolomitic lime, gypsum, foliar applied Mo and N on nutrient concentration of first cutting alfalfa in May 1986...................118

12 Influence of dolomitic lime, gypsum, foliar applied Mo and N on nutrient concentration of second cutting of alfalfa in June 1986..............119

13 Influence of surface applied limestone and gypsum at 13 Mg/ha on soil solution pH and ionic strength at different depths..................................127

14 Influence of surface applied limestone and gypsum at 13 Mg/ha on Al and Ca activity in soil solution at different depths..................................135

15 Influence of surface applied limestone and gypsum at 13 Mg/ha on Al speciation in soil solution at different depths as predicted by Geochem............137

LIST OF FIGURES

FIGURE PAGE

1 Alfalfa grown in growth pouches suspended from file rack....48

2 Macronutrient impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis....52

3 Micronutrient impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis....53

4 Anion impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis....54

5 Alfalfa grown with control nutrient solution (lacking Ca, Al) in modified growth pouch containing white filter paper wick (left) and original brown paper wick (right)....56

6 Calcium and Al effects on alfalfa growth two weeks after germination. Visual scorings ranged from 1 (poorest growth) to 10 (best growth)....68

7 Calcium and Al effects on alfalfa root penetration two weeks after germination....69

8 Calcium and Al effects on Al concentration in alfalfa roots two weeks after germination....71

9 Influence of surface limestone (applied 8 months before planting) on alfalfa root abundance at various soil depths for experiment 2. Numbers above each bar indicate pH values at different depths....84

10 Influence of surface limestone on alfalfa as compared to control (center) 86

11 Root mass in ISM as influenced by surface lime and various amendments....................................101

12 Nitrogen fixation in ISM as influenced by surface lime and various amendments........................104

13 Influence of lime and gypsum on soil solution K, Ca and Mg in the surface 5 cm of an Ernest silt loam soil...129

14 Influence of lime and gypsum on soil solution K, Ca and Mg at the 1 m depth in an Ernest silt loam soil...130

15 Influence of lime and gypsum on soil solution micronutrients in the surface 5 cm of an Ernest silt loam soil..132

16 Influence of lime and gypsum on soil solution micronutrients at the 1 m depth in an Ernest silt loam soil...133

CONTENTS

ACKNOWLEDGEMENTS iv

LIST OF TABLES vi

LIST OF FIGURES viii

Chapter

page

I. INTRODUCTION 1

II. CHEMICAL AND PHYSIOLOGICAL ASPECTS OF SOIL ACIDITY WITH FOCUS ON ALFALFA: A REVIEW 6

- ABSTRACT 6
- Growth Limiting Factors in Acid Soils 6
  - Soil pH 7
  - Base Saturation 8
  - Manganese Toxicity 9
  - Aluminum Toxicity 12
  - Calcium Deficiency 15
  - Molybdenum Deficiency 17
  - Phosphorus Deficiency 18
- Effects of Soil Acidity on Plants 21
  - Root Growth 21
  - Nutrient Uptake 24
  - Nodulation and N-Fixation 26
  - Crop Yield 28
  - Plant Tissue Composition 29
  - Plant Anatomy and Morphology 33
- CONCLUSIONS 34
- LITERATURE CITED 34

III. GROWTH POUCH METHOD FOR STUDYING THE INFLUENCE OF CA, AL AND PH ON CROP GROWTH IN NUTRIENT SOLUTIONS 45

- ABSTRACT 45
- INTRODUCTION 46
- MATERIALS AND METHODS 47
- RESULTS AND DISCUSSION 50
  - Experiment 1 50
  - Experiment 2 51
  - Experiment 3 55
- CONCLUSION 55
- LITERATURE CITED 57

IV. ALFALFA SEEDLING GROWTH IN NUTRIENT SOLUTIONS AS INFLUENCED BY ALUMINUM, CALCIUM AND PH . . . 59

ABSTRACT 59
INTRODUCTION 60
MATERIALS AND METHODS 62
  Pouch Preparation 62
RESULTS AND DISCUSSION 64
  Al Concentration and pH 64
  Calcium Influence on Al Toxicity 66
CONCLUSIONS 73
LITERATURE CITED 74

V. NO-TILL ALFALFA YIELD AND NUTRIENT CONCENTRATION AS INFLUENCED BY SURFACE LIME ON AN ACID ERNEST SILT LOAM SOIL 77

ABSTRACT 77
INTRODUCTION 78
MATERIALS AND METHODS 79
RESULTS AND DISCUSSION 81
  Soil Acidity 81
  Root Growth 83
  Crop Yield 85
  Tissue Composition 88
CONCLUSION 90
LITERATURE CITED 90

VI. THE INFLUENCE OF CA, N AND PH ON ALFALFA ROOT GROWTH AND NITROGEN FIXATION USING THE IMPLANTED SOIL MASS TECHNIQUE 92

ABSTRACT 92
INTRODUCTION 93
MATERIALS AND METHODS 95
RESULTS AND DISCUSSION 100
  Root Weight 100
  N Fixation 103
LITERATURE CITED 106

VII. RESPONSE OF ALFALFA GROWN ON ACID SOIL TO DIFFERENT CHEMICAL AMENDMENTS 108

ABSTRACT 108
INTRODUCTION 109
MATERIALS AND METHODS 110
  Sample Collection and Analysis 110
  Statistics 111
RESULTS AND DISCUSSION 111
  Soil Factors 111

Crop Yields 113
Tissue Analyses 117
LITERATURE CITED 120

VIII. SOIL SOLUTION AL AS A MEASURE OF AL TOXICITY IN ALFALFA ON ACID SOILS 122

ABSTRACT 122
INTRODUCTION 123
MATERIALS AND METHODS 126
RESULTS AND DISCUSSION 126
LITERATURE CITED 138

IX. SUMMARY AND CONCLUSIONS 140

VITA 142

Chapter I

INTRODUCTION

I The Problem

Soil acidity is a major problem faced by farmers growing alfalfa, particularly in the southeastern United States. Crops grown on acid soils often exhibit physiological disorders resulting in reduced crop yields. Soil acidity causes stunting of roots, which may also bring about drought susceptibility and poor use of subsoil nutrients. The no-till practice, which is normally effective in controlling erosion, has not, however, been advocated for alfalfa establishment and production on acid soils. This is based on the general belief that surface liming will not adequately correct soil acidity for efficent plant growth. The results of recent experiments at Virginia Tech have raised questions regarding the validity of this assumption. According to these studies, alfalfa roots could penetrate into acidic soils with high levels of Al under some conditions. These observations have left a number of unresolved questions:

1) Is the Ca status of the plant rather than the Al/soil pH level in the

subsoil more important in determining optimal performance of alfalfa?

2) Is the presence of available Mo near the surface vital to establish alfalfa (N fixed in roots near the soil surface in a high pH environment) while other parts of the root system are less effected by acidity and are capable of extracting water and possibly nutrients from acid subsoils?
3) Is application of lime at any location within the top 30 cm of the soil adequate for plant growth?

These basic questions need to be addressed before conservation tillage of legumes can be used at its full potential.

No-till alfalfa is becoming increasingly popular in southwest Virginia and other parts of the country. In addition to conserving valuable topsoil, this practice minimizes soil compaction and saves the producer valuable time. Extension agents at Virginia Tech currently recommend growing no-till alfalfa only if the soil pH is above 6.5. A survey conducted at Virginia Tech has shown that over 58% of farmers wanting to plant no-till alfalfa have soils with pH values less than 6.4 and 48% of farmers with established alfalfa stands have soils with pH levels less than 6.4. Conservation tillage practices would be much more effective if they could be extended to soils with pH levels below 6.4.

II Objectives

The proposed study will investigate various limiting and/or toxic factors in an attempt to establish conditions which would permit growing no-till alfalfa on acidic soils. Specifically, the following hypotheses will be tested:

1) No-till alfalfa can be grown under acidic conditions if only the soil surface is sufficiently limed;
2) molybdenum near the soil surface is critical in establishing alfalfa nodulating system for N assimilation;
3) if alfalfa receives sufficient Ca it can grow into any soil regardless of pH or Al content;
4) calcium content influences root penetration depth more than pH;

5) nitrogen fixation can be enhanced on acid soils by the addition of Ca, KOH or $Ca(OH)_2$;
6) exchangeable soil Al may not reflect accurately the suitability and adequacy of acid soils for growing alfalfa.

An extensive review of the physiological and chemical aspects of soil acidity on alfalfa is presented in Chapter II. Chapter III describes a new method for studying the influence of Ca, Al and pH on crop growth in nutrient solutions. Chapter IV deals with the influence of Al, Ca and pH on alfalfa growth in nutrient solutions.

Field studies involving the influence of various chemical amendments on alfalfa growth under acidic soil conditions are presented in Chapters V-VIII. Chapter V specifically deals with the influence of surface lime on the growth of no-till alfalfa under acidic soil conditions. Chapter VI examines the influence of Ca, N and pH on root growth and N fixation using the implanted soil mass technique. The results from a field study designed to show the response of alfalfa grown on acid soils to various chemical amendments is described in Chapter VII. Chapter VIII describes the use of soil solution Al as a measure of

Al toxicity in alfalfa on acid soils. The overall summary and conclusions are presented in Chapter IX.

Chapter II

CHEMICAL AND PHYSIOLOGICAL ASPECTS OF SOIL ACIDITY WITH FOCUS ON ALFALFA: A REVIEW

2.1 ABSTRACT

Soil acidity is a major growth-limiting factor for alfalfa (Medicago sativa L.) production. Subsurface soil acidity may limit the development of alfalfa root systems, which may result in reduced plant uptake of moisture and nutrients. Soil acidity may also contribute to either Al or Mn toxicity, or both. This often results in plant P deficiency. Unlike other micronutrients, Mo is less available in strongly acid soils. Acidity may inhibit N fixation by rhizobia, resulting in N deficiency and reduced yields. This chapter presents an overview of current concepts on soil acidity and its influence on alfalfa.

2.2 GROWTH LIMITING FACTORS IN ACID SOILS

The reasons for unsatisfactory growth of plants on acid soils are complex and in many instances, unpredictable. At a given soil pH value, the cause of poor growth may vary with soil type and also with plant species or variety (Foy, 1964).

2.2.1 Soil pH

Early investigators attributed the beneficial effect of liming to neutralization of soil acidity. This theory, however, received considerable criticism (Albrecht, 1941; Moser, 1943). Soil acidity or H activity itself is apparently not injurious to plants at pH values usually observed in soils, since plants showing adverse effects when growing in acid soils grow normally in nutrient solutions at the same pH values (Couto, 1982). Jacobson et al. (1950) and Nielsen and Overstreet (1955) have shown that H may decrease the absorption of metal cations by competition for carrier sites, so that a higher solution concentration of a given ion is necessary to support a given rate of absorption at lower pH values.

Aside from possible direct toxicity, high concentrations of H ions (Arnon and Johnson, 1942; Aslander, 1929; Cormack, 1945; Vlamis, 1953), have been shown to reduce differentially the uptake of certain metal ions. Phosphorus and Mo availability are greater at pH values near neutrality, while B, Fe, Zn and Mn availability increase with decreased pH. Arnon et al. (1942) observed that the increased H concentrations in nutrient solution decreased the uptake of Ca in tomato (*Lycopersicon esculentum*) and lettuce (*Latuca sativa*).

2.2.2 Base Saturation

The existence for any one soil of a relationship between pH value and base saturation has been shown by Pierre and Scarseth (1931), Mehlich (1941), Mehlich and Colwell (1944) and Peech (1941). According to Mehlich (1941), 2:1 type clay minerals have strongly acidic properties, and H is 70% neutralized at pH 5.4 and 100% at pH 7.4. Allaway (1945) and Mehlich and Colwell (1944, 1946) showed that plants could use exchangeable Ca more readily from kaolinitic than from montmorillonitic clays, and that Ca availability increased with the increasing degree of Ca saturation of clays.

The influence of base saturation in reference to yield and mineral composition of alfalfa was studied by Dawson (1958), using four Oregon soils differing in morphological and chemical properties. Soil pH values at the same base saturation level varied considerably for the different soils. The pattern of alfalfa response to lime also varied. On an Oxisol, optimum growth was obtained at 40% base saturation, while the other three soils (Oxisol, calcic Brown and Old Alluvial soils) showed optimum growth at either 80 or 100% base saturation. The yield response in these studies was attributed to either direct or indirect influence of lime. Concurrently, a split-root experiment was conducted to ascertain the effects of additional Ca supplied to an isolated section of alfalfa roots at each base saturation level. Additional Ca supplied to isolated root sections, independent of the soil, increased alfalfa yield significantly. The addition of Ca to the roots also decreased the Mn content of the foliage.

2.2.3 Manganese Toxicity

Excess soluble Mn has long been suspected as a major factor of acid soil infertility (Funchess, 1919). Fried and Peech (1946) observed that an increased concentration of Mn ions in soil solution reduced yield of alfalfa and increased the Mn content of the plant.

Sherman and Fujimoto (1947) obtained increased yields of carrots (*Daucus carota*) and lettuce with either paper mulch or applications of $Ca(OH)_2$; both treatments decreased the soil exchangeable Mn. The paper mulch lowered the soil temperature and conserved the soil moisture which produced a condition favorable to the hydration of the Mn in the soil, and thus reduced the exchangeable soil Mn level. According to Kipps (1947), alfalfa growth was significantly reduced and the plants became chlorotic when the Ca/Mn ratio in the tissues was less than 66.

The significance of Mn toxicity as a factor contributing to poor growth of plants on English soils was investigated by Hewitt (1945) and Wallace et al. (1945). They demonstrated that typical foliar toxicity symptoms in vegetables grown on acid soils could be reproduced by growing plants in sand cultures in the presence of 12 to 25 mg Mn L^{-1} in the nutrient solution. The detrimental effects of Mn were alleviated by increasing the concentration of Ca in the culture medium. As a consequence of these studies, the investigators concluded that lime serves two principal functions when added to acid soils: first, it decreases the soluble Mn and second, it provides a better Ca/Mn balance.

Foy (1964) investigated toxic factors in acid soils of the southeastern United States as related to the response of alfalfa to lime. Manganese toxicity was shown to be the primary yield limiting factor in nine of the 17 soils under study.

Liming of soil from a pH of 5.1 to 5.3 reduced the soluble soil Mn content (MacLean et al., 1972) and decreased the Mn content of plant tissues from 175-293 mg kg^{-1} in the controls to 68-148 mg kg^{-1}. Alfalfa grown in sand cultures showed symptoms of Mn toxicity when the concentration of Mn in the plants exceeded 175 mg kg^{-1} (Oullette and Dessureaux, 1958).

Plant availability of soil Mn is controlled by the oxidation-reduction potential of the soil environment, through physical, chemical, biochemical, biological and mineralogical properties of the soil as well as pH. The availability of Mn in soils is closely related to the activities of microorganisms which can oxidize the soluble and toxic divalent Mn to the tetravalent, non-toxic form (Bromfield, 1978).

Manganese toxicity, can be aggravated in alfalfa, and other crops by poor drainage . This phenomenon was noted as early as 1928 by Godden and Grimmett (1928) who observed decreased Mn solubility following waterlogging of the soil. Piper (1931) further showed that the content of soluble Mn in the soil was increased by flooding.

In more recent studies by Graven et al. (1965), flooding, regardless of soil pH, increased the content of exchangeable Mn and Mn content of alfalfa grown in a Kellner loamy sand. In the absence of a source of easily decomposable organic matter, Mn mobilization by flooding was considerably slower at a soil pH of 4.7 than at 7.3. The slower mobilization of Mn may have been related to the adverse effect of low pH on microbial activity. Liming promoted immobilization of Mn on the resumption of normal soil moisture relations after flooding. Seventy-two h of flooding increased the Mn content of the alfalfa on the unlimed soil from 426 mg kg^{-1} to more than 6,000 mg kg^{-1}. Excess Mn tended to accumulate in the leaves and growing points of the plants. The results suggest that the well-known susceptibility of alfalfa to Mn toxicity may also account for its sensitivity to poorly aerated soils.

In well-drained soils, Mn toxicity generally occurs only at soil pH values of 5.5 or below, but in flooded or compacted soils excess Mn can limit plant growth at soil pH values of 6.0 or above provided the soil parent materials contain sufficient total Mn (Siman et al., 1974).

2.2.4 Aluminum Toxicity

Because Al is a major constituent of soil clay minerals, Al toxicity is theoretically possible in most, if not all mineral soils (McLean, 1976). Aluminum toxicity can occur when the soil pH decreases below pH 5.5 as a result of acid rain or other means (Rechcigl and Sparks, 1985). These acidic conditions result in the decomposition of clay mineral structures, generally below pH 5.5, but particularly below 5.0 (Foy, 1976). When this point is reached, some of the Al, formerly a part of the clay particles, occupies additional cation exchange sites on clay surfaces and subsequently increases Al activity in soil solution.

Aluminum has become recognized as the dominant cation which gives rise to soil acidity and restricts crop growth on these soils (Kamprath, 1970; Hoyt and Nyborg, 1971; Foy, 1984).

Vlamis (1953) provided evidence that Al toxicity is the primary factor responsible for poor plant growth on an acid California soil. He displaced the soil solution from an unlimed soil and from a limed subsample and grew barley (Hordeum vulgare L.), plants on each. The plants grown in the solution displaced from the unlimed soil grew as well as those in the solution displaced from the limed soil only when $Ca(OH)_2$, NaOH or KOH was added. The addition of H_2SO_4 in sufficient quantities to reduce solution pH from the limed soil to that of the solution from the unlimed soil did not seriously reduce shoot growth of barley nor did the addition of both H_2SO_4 and $MnSO_4$. However, when $Al_2(SO_4)_3$ was added along with H_2SO_4 and $MnSO_4$, shoot growth was reduced to approximately 70% of maximum. It was noted that the solution level of Al in the unlimed soil was 1.8 mg kg^{-1} suggesting that Al is responsible for reduced barley growth. Adverse effects of Al on mature alfalfa growth in sand culture was shown by Ouellette and Dessureaux (1958).

A close correlation between the decrease of exchangeable Al in the soil and increased yield and longevity of alfalfa was also demonstrated by Moschler et al. (1960). In a study by Foy (1964) of toxic factors in acid soils of the southeastern United States, Al toxicity appeared to be a primary growth-limiting factor on Bladen soil (pH 4.8). Alfalfa yield responses to applications of lime on this soil were closely related to decreases in extractable soil Al.

Critical levels of toxic Al appear to be different for each soil. Inhibition of plant growth in any particular acid soil is apparently related to the relative amount of exchangeable Al, the level of Al saturation or water soluble Al. Levels of these factors in one soil may, however, be well below toxic levels in another soil (Adams and Lund, 1965). Soils having equal concentrations of soluble Al are differentially toxic to a given cotton (*Gossypium hirsutum*) variety (Adams and Hathcock, 1984).

Also, crops responded differently to different saturation levels. Corn was not affected when the Al saturation was less than 44%; however, Al saturation of 20% or less was required for the optimum soybean (*Glycine max*) and cotton growth (Wallace et al., 1945). These results are in general agreement with the relative tolerances of these crops to Al (Foy, 1984).

The mechanism of Al action is not well understood. Wright (1943) believed that Al interfered with the uptake and translocation of P. Schmehl et al. (1952) showed that Al also interfered with the absorption of Ca. Similar relationships have been shown by Simpson et al. (1977) and Edwards and Horton (1977).

2.2.5 Calcium Deficiency

Since the application of lime to acid soils may increase the Ca content of the crop, it has been suggested that the beneficial effect of liming results from the increased supply of Ca as a nutrient (Albrecht, 1941; Allaway, 1945; Mehlich and Colwell, 1946; Probert, 1980; Juo and Uzo, 1977). The soil pH in many of these studies was not controlled, thus it is possible that the uptake of Ca by the plant may have been influenced by the H activity and the amount of readily soluble Al and Fe.

If the principal beneficial effect of liming resulted from the supply of Ca, the soluble sources of Ca should be as satisfactory as limestone. Greenhouse studies of Fried and Peech (1946) and Schmehl et al. (1950) have shown that gypsum, in contrast to lime, fails to improve plant growth on acid soils. Moreover, the use of soluble Ca salts on some soils may actually result in a decrease in yield, because of increased Al concentration from displacement of Al from the exchange complex. If this excess Al is leached out, Ca could be beneficial by increasing Ca saturation and decreasing Al saturation in subsoil zones (Adams and Hathcock, 1984).

Nevertheless, the beneficial effects of Ca have been observed under certain conditions. In the early studies of Moser (1943), Ca was observed to be a more important growth factor than pH at low pH values in sand cultures. In other greenhouse investigations, Dawson (1958) reported increased alfalfa yield with soils by inclusion of Ca in the nutrient solution. The response was evident in a Melbourne (fine, mixed, mesic, Ultic Haploxeralf) and a Olympic (clayey, mixed, mesic, Xeric Haplohumult) soil, both of which contained considerably higher exchangeable and readily reducible Mn contents than did a Willamette (fine-silty, mixed, mesic, Ultic Argixeroll) soil. Although the investigations do not indicate clearly the nature of the response to Ca _per se_, they do suggest a Mn-Ca relationship. Juo and Uzo (1977) concluded that inadequate Ca was a more important growth limiting factor than Al concentration in two acid, coarse textured soils of Nigeria (pH 4.6 and 5.3). This is in agreement with work conducted by Probert (1980). In more recent studies it has been shown that surface application of gypsum increased alfalfa yields by 27% on an acid Cecil sandy clay (Sumner et al., 1985). They attributed the beneficial effects of gypsum to reduced soluble Al and increased Ca through the soil profile.

However, the overall evidence suggests that many, if not all, of the Ca deficiencies reported on acid soils are due to Al-Ca antagonism, and not low Ca *per se* (Foy et al., 1974 and Foy, 1984).

2.2.6 Molybdenum Deficiency

Molybdenum is necessary for legumes to carry out N_2 fixation; liming soils will generally increase the availability of soil Mo sufficiently to correct any observed Mo deficiency (Robinson et al. 1951). Molybdenum deficiencies reported on the Eastern Seaboard, the Great Lakes States and the Pacific Coast of the United States are generally associated with acid soils. Liming these soils will usually prevent Mo deficiency (Kubota, 1978). Similar interactions between lime and Mo in the growth and nutrition of alfalfa have been reported by other investigators (Ahlrichs et al., 1963; Evans et al. 1951; Giddens and Perkins, 1960; Kamprath and Foy, 1972). It is now recognized that one of the benefits of adding lime to acid soils is the increase in availability of Mo to alfalfa and other crops.

In greenhouse studies designed to investigate the response of legumes to Mo and lime on a Mardin silt loam (Coarse-loamy, mixed, mesic, Typic Fragiochrept) soil, application of Mo also corrected N deficiency by stimulating symbiotic N fixation (Kliewer and Kennedy, 1960). Of the four legumes studied, birdsfoot trefoil (_Lotus_ _corniculatus_) showed the greatest response to Mo. This was especially true in the absence of lime. Molybdenum added to the virgin soil increased the yield of the legume to nearly the yield obtained with 2.2 Mg ha^{-1} of lime.

2.2.7 Phosphorus Deficiency

Phosphorus requirements for plants are higher on strongly acid soils than on those limed to neutralize excess exchangeable Al (Foy, 1984). Jones and Fox (1978) reported that tomato required more P at pH values 5.7 than at pH values from 6.3 to 7.2. The accumulation of P in the roots of plants in acid soils and the development of P deficiency symptoms in the shoots have long been associated with Al and P interactions (Hartwell and Pember, 1918). Early research established that application of large amounts of superphosphate to very acid soils will reduce, at least comparatively, the detrimental effects of these soils on the growth of plants (Pierre and Stuart, 1933). Santana and

Braga (1977) observed P concentration in rice (Oryza sativa) tops to decrease with increasing Al saturation of soil.

Liming also increased availability of P as evidenced by increased P uptake by plants (Dunn, 1943). Quiros and Gonazalez (1979) concluded that best crop yields were obtained when acid soils were limed at rates equivalent to 1.5 to 3.0 times the exchangeable Al level, and P added at 3 to 3.75 times the P fixing capacity of the soil. The fact that P can act as a liming material by precipitating Al has been known for many years (Kamprath and Foy, 1972; Foy, 1984). Beradze (1977) reported that P fertilization greatly reduced Al toxicity in corn (Zea mays). Similar results were reported with alfalfa (Sumner, 1979). Burgess and Pensker (1923) suggested that P reduced the toxicity of Al by precipitation within the plant tissues. This was based on the fact that application of P fertilizers greatly increased the concentration of P within the plant without appreciably affecting the Al concentration in the tissues. Furthermore, application of lime had much less effect than P on increasing P absorption and yields. Pierre and Stuart (1933) drew similar conclusions from their studies of soils and plants grown in nutrient solutions. However, they did not specify where the internal precipitation was located.

Wright (1943) analyzed barley plants grown in culture solutions with and without Al and showed a greater percentage of P in the Al-toxic plants than in the normal plants. The accumulation of P was particularly apparent in the roots. A low percentage of water-soluble P in these plants indicated inactivation of P. The high total P and low water-soluble P contents of plants grown in the presence of Al was attributed to the precipitation of P by Al within the plants, specifically within the roots (Wright, 1943; Wright and Donahue, 1955; Foy, 1984). Wallihan (1948) was unable to find evidence of such internal precipitation and proposed instead that Al, and perhaps P is held to root surfaces by some other mechanism such as ionic exchange.

In subsequent studies by Randall and Vose (1963) it was concluded that Al-induced P uptake is largely a metabolic process, although precipitation effects on the root could not be ruled out. The authors suggested that subsequent to the uptake process, P is bound by Al within the plant, thus causing the symptoms of P deficiency, characteristic of Al toxicity.

The data of Rorison (1965) and Clarkson (1967) suggest that two types of reactions take place between Al and P. The first occurs at the cell surface and results in fixation of P by an adsorption reaction; the second occurs within the cell. Their evidence did not support the view that Al enhances the uptake of P. Viets (1944), on the other hand, provided evidence that polyvalent cations, including Al, stimulate the uptake of P. Similarly, Ragland and Coleman (1962) reported a seven fold increase in the uptake of P by excised roots of snap beans (Phaseolus vulgar L.) following pretreatment with Al.

Jones and Fox (1978) reported that excess Al in acid soils fixed P in less plant available forms and that Al was toxic to plant roots and reduced their ability to exploit the soil for P. In similar studies, Helyar (1978) concluded that Al toxicity effects were largely associated with Al interference with P metabolism and with Al binding to root cell wall pectins, stopping root elongation. Juo and Uzo (1977) showed optimum P availability occured at pH values between 5.0 and 6.0 in two Nigerian Ultisols.

2.3 EFFECTS OF SOIL ACIDITY ON PLANTS

2.3.1 Root Growth

Failure of plants to develop deep, vigorous root systems is often due to subsoil acidity (Foy et al., 1974). Fox and Lipps (1955) studied the development of alfalfa root systems growing on limed and unlimed acid soils for 3 years. During the first season alfalfa roots penetrated more than

152 cm into the limed soil while those in unlimed reached a maximum of only 91 cm. Wattenpaugh (1936) reported little or no alfalfa root growth below pH 4.7 in a virgin DeKalb silt loam. When the pH was raised to 5.0 or above by liming, roots were observed to proliferate in that layer. There also is some evidence that alfalfa roots may penetrate into acidic soils when lime is applied to the soil surface (Rechcigl and Reneau, 1984). In this study alfalfa roots penetrated to 84 cm, where the pH was 4.1 and the exchangeable Al was 225 mg L^{-1}. There also is evidence that no-till alfalfa can be grown on some acid soils containing high levels of exchangeable Al provided surface liming is employed at planting (Rechcigl et al., 1985c).

Soil acidity may result in Ca deficiency in the root. The symptoms are straight small-diameter primary roots with brown tips which may die in severe cases (Howard and Adams, 1965). Studies by Adams and Moore (1983) showed Ca deficiency to be a major problem in cotton roots grown on Coastal Plain soils with less than 17% Ca saturation. Research by Ragland and Coleman (1959) and Howard and Adams (1965) suggest that except for sandy soils, poor root growth in acid subsoils is not generally the result of Ca deficiency.

Other studies have shown the factor most responsible for reduced root growth is excess soluble Al (Adams, 1981; Munns, 1965). Ragland and Coleman (1959) reported that the growth of roots into unlimed subsoils was inversely related to exchangeable Al. The detrimental effects of small amounts of Al on root growth have been well documented (McLean and Gilbert, 1928; Ragland and Coleman, 1959; Rios and Pearson, 1964). As little as 1.0 mg L^{-1} Al in solution can inhibit root growth in alfalfa (Simpson et al., 1977; MacLeod and Jackson, 1965). Aluminum toxicity is manifested by swollen, stubby and gnarled, primary roots (Rios and Pearson, 1964). Under conditions of high Al saturation, soybean roots were larger in diameter and less branched as compared to roots grown under low Al saturation (Sartain and Kamprath, 1975).

In solution culture studies with Al, root weight was generally increased with the reduction of cationic Al concentration (Foy and Brown, 1964). However, in a soil study a decrease in the dry weight of alfalfa roots has been observed as Al levels were reduced (John et al., 1972). With barley (Hordeum vulgare L.) the root length decreased with increasing levels of Al in solution (Reid et al., 1971). In soil studies sorghum (Sorghum bicolor) roots decreased as percentage Al saturation increased, but roots grew well in each subsoil when lime was applied (Ragland and Coleman, 1959). Even H toxicity is not a major factor involved in the stunting of roots, it has been shown that cotton roots were reduced as a result of H toxicity when soil pH was below 4.25 (Howard and Adams, 1965). Exchangeable Mn has limited affect on root growth and thus will not be discussed in detail (Adams and Moore, 1983).

2.3.2 Nutrient Uptake

Two general factors are largely responsible for controlling the availability of a given ion to plants. One is the effective concentration of the ion in the soil solution (intensity) and the other is the rate at which solution concentration is renewed (quantity) as ions are removed by plant roots. Once mineral elements are made available in soils, plant availability depends on transport to and into the root. Factors affecting ion transport from the soil to the root surface and into the root have been discussed in reviews (Wilkinson, 1972; Olsen and Kemper, 1968; Barber, 1984).

The profound effect of pH on the nutrient uptake by plants has long been recognized (Moore, 1972). Cation absorption is sharply reduced at pH values below 5; the optimum conditions are in the range of pH 5 to 7. Anions are considerably less affected by low pH; at pH values above 6, however, the rate of absorption is reduced by increasing OH concentration.

Marked competition with respect to plant uptake of nutrients usually occurs only between chemically similar ions, such as K-Rb and Ca-Sr. Competitive effects between dissimilar ions apparently occur more readily in soils than in nutrient solutions because of soil factors which influence the availabilities of the ions (Coleman et al., 1967). Schmehl et al. (1952) showed that the rate of absorption of Ca by alfalfa was markedly reduced by the presence of Al ions and to a lesser degree by Mn and H ions in the nutrient solution. Heavy liming of soils with calcitic limestone application normally reduces Mg uptake by plants. This is believed to result from the chemical similarity of Ca and Mg and the replacement of Mg by Ca. The use of calcic limestone on low-Mg soils may contribute to the development of Mg deficiency in soils.

In greenhouse studies conducted by Thorp and Hobbs (1956), lime application significantly increased total uptake of Ca by alfalfa grown in acid Kansas soils. No differences were observed for total Mg uptake from liming with dolomitic limestone. The ineffectiveness of Ca in lowering the Mg uptake may have been due to the large amounts of Mg in the soils. Although no apparent effect resulted from application of lime on the P concentration in the plant, there was a greater uptake of P from the limed than from the unlimed soils. Potash uptake was similarly increased by lime applications.

2.3.3 Nodulation and N-Fixation

Symbiotic N fixation in legumes is affected by soil acidity (Jo et al., 1981). Most legumes grow best in neutral or nearly neutral soils, in part at least because of the favorable conditions for activities of the nodule bacteria in such soils (Karraker, 1927). Munns (1978) showed that acidity inhibits nodulation of alfalfa in the early stages, but has no effect on nodulation after nodule infection. Acidity prevents root hair infection, thus resulting in no nodule formation.

The limiting pH for rhizobial survival and adequate nodule formation in alfalfa is about pH 6.0 (Date, 1970). In order for nodulation to take place, sufficient rhizobia must be present in the soil. However, in soils below pH 6.0, this condition is not met due to poor survival of acid sensitive rhizobia. A great deal of variability in pH tolerance has been shown, however among some rhizobia. In studies comparing different rhizobia species to acidity, Vincent (1965) concluded that *R. meliloti* is acid sensitive while *R. trifolii* is less so and *R. japonicum* is acid tolerant. It was also shown that different strains within the same *Rhizobium* sp. vary considerably in response to soil pH. Holding and King (1963) showed that the addition of lime to acidic soil rendered *R. trifolii* strains more effective, presumably as a result of increased pH.

Studies have shown that other side effects beside the direct effects of acidity on rhizobia activity may be responsible for inhibition. In acid soils there is an increase in uptake of heavy metals. Also, nutrients like Ca and Mo which are essential for symbiosis are less available in acid soils (Skerman, 1977).

The sensitivity of many legumes to acid soils may be explained by Mn toxicity on the N-fixing process (Foy, 1984). Dobereiner (1966) showed the N content of bean plants (*Phaseolus vulgaris*) was reduced by 30 to 60% when plants were grown in sand culture-solutions containing 25 mg L^{-1} Mn. However, when the plants were supplied with mineral N, the corresponding reduction was only 13%. In plants that were totally dependent upon symbiotic N-fixation the N contents of the bean plants decreased linearly with the logarithm of the Mn concentrations in plant tops, however this was not true in plants supplied with mineral N.

Keyser and Munns (1979) showed evidence that with soybean rhizobia in acid soils, Al toxicity and soil acidity are probably more important than Mn toxicity in restricting N-fixation. Rhizobia of some legume plants appear more sensitive to Al than do their host plants (Foy, 1984). Carvalho et al. (1981) concluded tht Al toxicity decreased the growth of *Stylosanthes* species more severely when plants were dependent on symbiotic N fixation than when combined N was applied as fertilizer. In earlier studies, Pieri (1974) demonstrated that the nodulation of groundnut (*Arachis hypogaea*) was reduced when the Al saturation of the soil CEC reached 30% in sandy soils of Senegal, while higher levels of Al saturation were required for toxicity of the host plant.

2.3.4 Crop Yield

Soil acidity is a major problem in the humid regions of the United States. Many investigators have observed soil acidity to have detrimental effects on crop yield (Adams and Pearson, 1967; Sumner, 1979; Lathwell and Peech, 1964; Adams, 1981; Coleman et al., 1967; Foy, 1984; Rechcigl et al., 1985a, 1985c; Rechcigl et al. 1986a, 1986b).

2.3.5 Plant Tissue Composition

The detrimental effect of acid soils on plant tissue composition and its reversal by liming has been demonstrated by a number of investigators. Application of lime to acid soils usually increases the Ca content of the crops (Albrecht, 1932; Naftel, 1937; Dunn, 1943; Schmehl et al., 1950). Such evidence was used by some investigators as a basis for hypothesizing that Ca deficiency was responsible for reduced crop yields in acid soils. As discussed elsewhere, gypsum normally does not affect growth favorably and frequently fails to increase the Ca content of the plants grown under acid soil conditions. Other factors, such as antagonistic effect of Al or Mn on the Ca uptake may thus be involved (Schmehl et al., 1950; 1952).

Liming also reduces the Mn content of plants. For example, in the studies of Fried and Peech (1946), the Mn content of the alfalfa leaves was reduced to one eighth that present in the control in response to lime application. Marked decreases in the Mn content of alfalfa following liming were also noted by Foy (1964) and MacLean et al. (1972).

Thorp and Hobbs (1956) conducted a greenhouse study with acid Kansas soils to evaluate the effect of lime application on the composition of alfalfa. Lime application significantly increased the Ca content in the plants. No significant effects in response to treatment were present for the percentage of plant Mg, P and K. The increased Ca content in these studies was attributed to the higher Ca content of the soils. Calcium content of the soils apparently did not depress the Mg and K content of the plants since both of these elements were above the critical levels, as established by Bear and Wallace (1950).

Another greenhouse investigation on alfalfa was conducted by Dawson (1958), using pedologically contrasting soils limed to different base saturation levels. In these studies Mn content of the foliage was decreased by increasing the base status from 60 to 80% in 2 of the 3 soils studied. The omission of Ca from the nutrient solution increased the Mn content in the tops of plants, particularly at the lower base saturation levels. The Ca content of the foliage generally increased with the increased base saturation levels.

The problem of acid soils in Australia with specific reference to the effects of Al on chemical composition of pasture legumes were investigated by Andrew et al. (1973). Several tropical and temperate legume species were grown in culture solution under greenhouse conditions, in the presence of 0.0, 0.5, 1.0 and 2.0 mg L^{-1} of Al. Aluminum concentrations in the plant tops were increased as a result of Al treatments. In the tropical species the increases were relatively small and uniform, but less than those for the temperate Trifolium species. These in turn were less than those for the temperate Medicago species. Aluminum concentrations in the roots were much higher than the concentrations in the corresponding plant tops at all treatments. Ratios of Al to P concentrations in the roots to those in the tops of control plants varied from 1 to 6, whereas ratios for plants grown in the 1.0 mg L^{-1} Al treatments varied from 20 to 40.

The effect of treatment on P concentrations in the plant tops varied with species. Species which were sensitive to Al showed reduced P concentrations in their tops when grown in the presence of Al at any concentration. Concentrations of P in the plant roots were generally higher than those in the plant tops and the levels of the former were further increased following Al treatments.

Aluminum treatment also reduced the concentration of Ca in the tops for all species studied. Calcium concentrations in the roots were considerably less than those in the tops but the trends in concentrations resulting from Al treatment were similar to those in the tops.

In most of the sensitive species, treatment with Al reduced plant K. In the majority of the tolerant species, however, K concentrations were increased or not affected by Al treatment. Potassium concentrations in the roots were generally lower than in the tops, but were little affected by Al except for the 2 mg L^{-1} treatment on M. sativa (Andrew et al., 1973) where a substantial reduction occurred. Aluminum treatment also had little effect on Mg levels, particularly in the tolerant species. The effects of Al on plant Na also were not significant.

Species which were tolerant to Al had generally constant total cation concentrations over the four treatments, but the sensitive species had considerably lower values as a result of treatment. In those species in which increased Al reduced the Ca concentration, the reduction in Ca was balanced by increase in K and, to a lesser extent, Mg and Na (Andrew et al., 1973).

A relatively low concentration of Al in the plant tops compared with those in the roots has also been observed (Randall and Vose, 1963; Foy and Brown, 1963 and 1964; Munns, 1965; Clarkson, 1967; MacLeod and Jackson, 1967). The reduction of P concentrations in plant tops with a commensurate increase in the P concentration in plant roots due to Al treatment is well documented (Wright and Donohue, 1953; Randall and Vose, 1963; Foy and Brown, 1964; MacLeod and Jackson, 1965; Munns, 1965; Clarkson, 1967).

2.3.6 Plant Anatomy and Morphology

This subject has been extensively reviewed by Foy (1984). Selected sections of Foy's review will be summarized in this section.

The foliar symptoms of Al toxicity are not always easily identified. They are often similar to P deficiency, consisting of stunting, small dark green leaves and late maturity, purpling of stems, leaves and leaf veins and yellowing and death of leaf tips. In other instances, Al toxicity may appear as an induced Ca deficiency which results in curling or rolling of young leaves and a collapse of growing points of petioles. The most characteristic symptom of Al toxicity is the stunting of the root system. Root tips and lateral roots may become thickened, stubby, brittle and turn brown.

Unlike Al, excess Mn generally affects plant tops to a greater degree than roots. Plant symptoms of Mn toxicity include marginal chlorosis and necrosis of leaves. In severe cases of Mn toxicity, plant roots will turn brown.

2.4 CONCLUSIONS

A review of the chemical and physiological aspects of soil acidity shows that information on many aspects of soil acidity is either lacking or incomplete. In order to fully assess the implications of soil acidity on crop growth, additional knowledge is needed. More attention must be given to the mechanisms responsible for reduced yields due to soil acidity and tie the physiological and biochemical nature of differential tolerances to acid soil stress factors among plant genotype within species. Only then will we fully understand the cause of reduced yields under acidic soil conditions and be able to resolve the problem. With the help of conventional plant selection, breeding, genetic engineering and innovative approaches to research in soil fertility management, it appears that these two goals can be attained.

2.5 LITERATURE CITED

Adams, F. 1981. Alleviating chemical toxicities in liming acid soils. p. 269-301. In G. F. Arkin and H. M. Taylor (ed.) Modifying the root environment to reduce crop stress, ASAE monograph no. 4. Am. Soc. Agric. Eng., St. Josephs, MI.

Adams, F. and P. J. Hathcock. 1984. Aluminum toxicity and calcium deficiency in acid subsoil horizons of two coastal plains soil series. Soil Sci. Soc. Am. J. 48:1305-1309.

Adams, F. and Z. F. Lund. 1965. Effect of chemical activity of solution aluminum on cotton root penetration of acid subsoils. Soil Sci. 101:193-198.

Adams, F. and B. L. Moore. 1983. Chemical factors affecting root growth in subsoil horizons of Coastal Plain soils. Soil Sci. Soc. Am. J. 47:99-102.

Adams, F. and R. W. Pearson. 1967. Crop response to lime in the southern United States and Puerto Rico. p. 161-206. In R. W. Pearson and F. Adams. (ed.) Soil Acidity and Liming. Agronomy 12, Am. Soc. Agron., Madison, WI.

Ahlrichs, L. E., R. G. Hanson, and J. M. MacGregor. 1963. Molybdenum effect on alfalfa grown on thirteen Minnesota soils in the greenhouse. Agron. J. 55:484-486.

Albrecht, W. A. 1932. Calcium and hydrogen ion concentration in the growth and inoculation of soybeans. J. Am. Soc. Agron. 24:792-806.

Albrecht, W. A. 1941. Plants and the exchangeable calcium of the soil. Am. J. Bot. 28:394-402.

Allaway, W. H. 1945. Availability of replaceable calcium from different types of colloids as affected by degree of calcium saturation. Soil Sci. 50:207-217.

Andrew, C. S., A. D. Johnson, and R. L. Sandland. 1973. Effect of aluminum on the growth and chemical composition of some tropical and temperate pasture legumes. Austral. J. Agr. Res. 24:325-339.

Arnon, D. I., W. E. Fratzke, and C. M. Johnson. 1942. Hydrogen-ion concentration in relation to absorption of inorganic nutrients in higher plants. Plant Physiol. 17:515-524.

Arnon, D. I. and C. M. Johnson. 1942. Influence of hydrogen-ion concentration on the growth of higher plants under controlled conditions. Plant Physiol. 17:525-539.

Aslander, A. 1929. Concentration of the nutrient medium versus its hydrogen-ion concentration as manifested by plant growth. Svensk. Bot. Tidskr. 23:96-140.

Barber, S. A. 1984. Soil nutrient bioavailability: a mechanistic approach. Wiley Press, New York, NY.

Bear, F. E. and A. Wallace. 1950. Alfalfa: Its mineral requirements and chemical composition. New Jersey Agr. Exp. Sta. Bull. 74.

Beradze, E. N. V. 1977. Some aspects of water status of plants on subtropical soil in relation to effect of mineral elements. Rudy Pedagogicheskikh Instituton Guzinskoi SS, Estestvoznainza: Ruperativnyi Zhurnal 6:55-153.

Bromfield, S. M. 1978. The effect of manganese-oxidizing bacteria and pH on the availability of manganous ions and manganese oxides to oats in nutrient solution. Plant Soil 49:23-39.

Burgess, P. S. and F. R. Pensker. 1923. "Active" aluminum as a factor detrimental to crop production in many acid soils. Rhode Island Agr. Exp. Sta. Bull. 194.

Carvalho, M. M., D. G. Edwards, C. S. Andrew, and C. J. Asher. 1981. Aluminum toxicity, nodulation and growth of stylosanthes species. Agron. J. 73:261-265.

Clarkson, D. T. 1967. Interactions between aluminum and phosphorus on root surfaces and cell wall material. Plant Soil. 27:347-56.

Coleman, N. T., E. J. Kamprath, and S. B. Weed. 1967. Liming. p. 476-522. In R. W. Pearson and F. Adams (ed.) Soil Acidity and Liming. ASA No. 12. Am. Soc. Agron., Madison, WI.

Cormack, R. G. H. 1945. Cell elongation and development of root hairs in tomato roots. Am. J. Bot. 32:490-496.

Couto, W. 1982. Soil pH and plant productivity. p. 71-83. In M. Rechcigl (ed.) CRC Handbook of Agricultural Productivity. Boca Raton, FL CRC Press. Vol. 1.

Date, R. A. 1970. Microbiological problems in the inoculation and nodulation of legumes. Plant Soil. 32:703-725.

Dawson, M. D. 1958. Influence of base saturation and calcium levels on yield and mineral content of alfalfa. Soil Sci. Soc. Am. J. 22:328-333.

Dobereiner, J. 1966. Manganese toxicity effects on nodulation and N fixation of beans (Phaseolus vulgaris L.) in acid soils. Plant Soil 24:153-166.

Dunn, L. E. 1943. Effect of lime on availability of nutrients on certain Western Washington soils. Soil Sci. 56:297.

Edwards, J. H. and B. D. Horton. 1977. Aluminum induced calcium deficiency in peach seedlings. J. Am. Soc. Hort. Sci. 102:459-461.

Δ Evans, H. J., E. R. Purvis, and F. E. Bear. 1951. Effect of soil reaction on availability of molybdenum. Soil Sci. 71:117-124.

Fox, R. L. and R. C. Lips. 1955. Influence of soil profile characteristics upon the distribution of roots and nodules of alfalfa and sweetclover. Agron. J. 47:361-367.

Foy, C. D. 1964. Toxic factors in acid soils of the southeastern United States as related to the response of alfalfa to lime. USDA, ARS Production Report No. 80. 26 p.

Foy, C. D. 1984. Physiological effects of hydrogen, aluminum, and manganese toxicities in acid soil. p. 57-97. In F. Adams (ed.) Soil Acidity and Liming. Am. Soc. Agron. Madison, WI.

Foy, C. D. 1976. General principles involved in screening plants for and manganese tolerance. p. 255-267. In M. J. Wright and S. A. Ferrari (ed.) Plant Adaptation to Mineral Stress in Problem Soils. Cornell Univ. Agric. Exp. Stn. Spec. Pub., Ithica NY.

Foy, C. D. and J. C. Brown. 1963. Toxic factors in acid soils. I. Characterization of aluminum toxicity in cotton. Soil Sci. Soc. Am. Proc. 27:403-407.

Foy, C. D. and J. C. Brown. 1964. Toxic factors in acid soils: II. Differential aluminum tolerance of plant species. Soil Sci. Soc. Am. Proc. 28:27-32.

Foy, C. D., R. G. Orellana, J. W. Schwartz, and A. L. Fleming. 1974. Response of sunflower genotypes to aluminum in acid soil and nutrient solution. Agron. J. 66:293-96.

Fried, M. and M. Peech. 1946. The Comparative effects of lime and gypsum upon plants grown on acid soils. J. Am.Soc. Agron. 38:614-623.

Funchess, M. J. 1919. Acid soils and toxicity of manganese. Soil Sci. Soc. Am. Proc. 8:69.

Δ Giddens, J. and H. F. Perkins. 1960. Influence of molybdenum on growth and composition of alfalfa and distribution of molybdenum in a Cecil-Lloyd soil. Soil Sci. Soc. Am. 24:496-497.

Godden, W. and R. E. Grimmett. 1928. Factors affecting iron and manganese contents of plants with special reference to herbage causing pinning and bush sickness. J. Agr. Sci. 18:363-372.

Graven, E. H., O. J. Attoe, and D. Smith. 1965. Effect of liming and flooding on manganese toxicity in alfalfa. Soil Sci. Soc. Am. Proc. 29:702-706.

Hartwell, B. L. and F. R. Pember. 1918. The presence of aluminum as a reason for the difference in the effect of so-called acid soil on barley and rye. Soil Sci. 6:259-281.

Helyar, K. R. 1978. Effects of aluminum and manganese toxicity on legume growth. p. 207-221. In C. S. Andrew and E. J. Kamprath (ed.) Mineral Nutrition of legumes in Tropical and Subtropical Soils. Melbourne, Australia.

Hewitt, E. J. 1945. The resolution of the factors in soil acidity. Ann. Rpt. Long Ashton Res. Sta. p. 51-60.

Holding, A. J. and J. King. 1963. The effectiveness of indigenous populations of Rhizobium trifolii in relation of soil factors. Plant Soil 18:191-198.

Howard, D. D. and F. Adams. 1965. Calcium requirements for penetration of subsoils by primary cotton roots. Soil Sci. Soc. Am. Proc. 29:558-562.

Hoyt, P. B. and M. N. Nyborg. 1971. Toxic metals in acid soil: 1. Estimation of plant available aluminum. Soil Sci. Soc. Am. Proc. 35:236-240.

Jacobson, L., R. Overstreet, H. M. King, and R. Handley. 1950. A study of potassium absorption by barley roots. Plant Physiol. 25:639-647.

Jo, J., S. Yoshida, and R. Kayama. 1981. Influences of soil acidity and aplied nitrogen on the growth and chemical composition of ladino clover and alfalfa. J. Japan. Grassl. Sci. 27:79-84.

John, M. K., V. W. Case, and L. Vanlaerhoren. 1972. Liming of alfalfa (Medicago sativa L.): I. Effect of plant growth on soil properties. Plant Soil 37:353-361.

Jones, J. P. and R. L. Fox. 1978. Phosphorus nutrition of plants influenced by manganese and aluminum uptake from an oxisol. Soil Sci. 126:230-236.

Juo, A. S. R. and F. O. Uzo. 1977. Liming and nutrient interactions in two ultisols from southern Nigeria. Plant Soil 47:419-430.

Kamprath, E. J. 1970. Exchangeable aluminum as a criterion for liming leached mineral soils. Soil Sci. Soc. Am. Proc. 34:252-254.

Kamprath, E. J. and C. D. Foy. 1972. Lime fertilizer-plant interactions in acid soils. p. 105-151. In R. A. Olsen, et al. (ed.) Fertilizer Technology and Use Soil Sci. Soc. Am., Madison, WI.

Karraker, P. E. 1927. Production of nodule and different parts of the root systems of alfalfa plants growing in soils of different reaction. Soil Sci. 24:103-107.

Keyser, H. H. and D. N. Munns. 1979. Effects of calcium, manganese and aluminum on growth of rhizobia in acid media. Soil Sci. Soc. Am. J. 43:500-503.

Kipps, E. H. 1947. The calcium of manganese ratio in relation to the growth of lucerne at Camberra. ACT J. Council Sci. Indust. Res. 20:176-189.

Kliewer, W. M. and W. K. Kennedy. 1960. Studies on responses of legumes to molybdenum and lime fertilization on Mardin silt loam soil. Soil Sci. Soc. Am. Proc. 24:377-380.

Kubota, J. 1978. Molybdenum status of United States soil and plants. p. 555-582. In W. R. Chapell and K. Petersen (ed.) Vol 2. Chap G, The Biology of Molybdenum Marcel Dekker, New York, NY.

Lathwell, D. J. and M. Peech. 1964. Interpretations of chemical soil tests. New York (Cornell U.). Ag. Exp. Sta. Bull. 995.

MacLean, A. J., R. I. Halstead, and B. I. Finn. 1972. Effects of lime on extractable aluminum and other soil properties and on barley and alfalfa grown in pot tests. Can. J. Soil Sci. 52:427-438.

MacLeod, L. B. and L. P. Jackson. 1965. Effect of concentration of the aluminum ion on root development and establishment of legume seedlings. Can. J. Soil Sci. 45:221-234.

Mclean, F. T. and B. E. Gilbert. 1928. Aluminum toxicity. Plant Physiol. 3:292-302.

McLean, E. O. 1976. Chemistry of soil aluminum. Commun. Soil Sci. Plant Anal. 7:619-636.

Mehlich, A. 1941. Base unsaturation and pH in relation to soil type. Soil Sci. Soc. Am. Proc. 6:150-156.

Mehlich, A. and W. E. Colwell. 1944. Influence of nature of soil colloids and degree of base saturation on growth and nutrient uptake by cotton and soybeans. Soil Sci. Soc. Am. Proc. 8:179-184.

Mehlich, A. and W. E. Colwell. 1946. Absorption of calcium by peanuts from kaolin and bentonite at varying levels of calcium. Soil Sci. 61:369-374.

Moore, D. P. 1972. Mechanism of micronutrient uptake by plants. p 171-198. In J. J. Mortvedt, P. N. Giordano and W. L. Lindsay (ed.) Micronutrients in Agriculture. Soil Sci. Soc. Am., Madison, WI.

Moschler, W. W., G. D. Jones, and G. W. Thomas. 1960. Lime and soil acidity effects of alfalfa growth in a red yellow Podzolic soil. Soil Sci. Soc. Am. Proc. 24:507-509.

Moser, F. 1943. Calcium nutrition at respective pH levels. Soil Sci. Soc. Am. Proc. 7:339-344.

Munns, D. N. 1965. Soil acidity and growth of a legume II. Reactions of aluminum and phosphate in solution and effects of aluminum phosphate, calcium and pH on Medicago sativa L. and Trifolium subterraneium L. in solution cultures. Aust. J. Agr. Res. 16:743-55.

Munns, D. N. 1978. Soil acidity and nodulation. p. 247-263. In C. S. Andrew and E. J. Kamprath (ed.) Mineral Nutrition of Legumes in Tropical an Subtropical Soils. Commonwealth Scientific and Industrial Research Organization, Australia.

Naftel, J.A. 1937. Soil liming investigations. J. Am. Soc. Agron. 29:537-547.

Nielson, T. R. and R. Overstreet. 1955. A study of the role of the hydrogen ion in the mechanism of potassium absorption by excised barley roots. Plant Physiol. 30:303-309.

Olsen, S. R. and W. D. Kemper. 1968. Movement of nutrients to plant roots. Adv. Agron. 20:91-151.

Ouellette, G. J. and L. Dessureaux. 1958. Chemical composition of alfalfa as related to degree of tolerance to manganese and aluminum. Can. J. Soil Sci. 38:26-214.

Peech, M. 1941. Availability of ions in light sandy soils as affected by soil reaction. Soil Sci. 51:473-485.

Pieri, C. 1974. First experimental results on the susceptibility of groundnut to aluminum toxicity. L' Agronomie Tropicale 29:685.

Pierre, W. H. and G. D. Scarseth. 1931. Determination of the percent base saturation of soil and its value in different soils at definite pH values. Soil Sci. 31:99-114.

Pierre, W. H. and A. D. Stuart. 1933. Soluble aluminum studies: IV. The effects of phosphorus in reducing the detrimental effects of soil acidity on plant growth. Soil Sci. 36:211-226.

Piper, C. S. 1931. The available manganese in the soil. J. Agr. Sci. 21:762-779.

Probert, M. E. 1980. Growth responses to various calcium sources in a yellow earth soil with low calcium status. Aust. J. Expt. Agric. Anim. Husb. 20:240-246.

Quiros, S. and M. A. Gonzalez. 1979. Neutralization of exchangeable aluminum and uptake of phosphorus in three Costa Rica soils. Agronomica Costaniceuse 3:5, 137-149.

Ragland, J. L. and N. T. Coleman. 1959. The effect of solution aluminum and calcium on root growth. Soil Sci. Soc. Am. Proc. 23:355-357

Ragland, J. L. and N. T. Coleman. 1962. The influence of aluminum on phosphorus uptake by snap bean roots. Soil Sci. Soc. Am. Proc. 26:88-90.

Randall, P. J. and P. B. Vose. 1963. Effect of aluminum on phosphorus by snap bean roots. Proc. Soil Sci. Soc. Am. 26:88-90.

Rechcigl, J. E. and R. B. Reneau, Jr. 1984. Effect of subsurface acidity on alfalfa in a Tatum clay loam. Commun. Soil Sci. Plant Anal. 15:811-818.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985a. Effect of subsurface amendments and irrigation on alfalfa growth. Agron. J. 77:72-75.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985b. Movement of fertilizer and limestone in a Tatum clay loam. Commun. Soil Sci. Plant Anal. 16:427-438.

Δ Rechcigl, J. E. and D. L. Sparks. 1985. Effect of acid rain on the soil environment: A review. Commun. Soil Sci. Plant Anal. 16:653-680.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., and W. Kroontje. 1985c. Influence of surface liming on the yield and nutrient concentration of alfalfa established using no-tillage techniques. Agron. J. 77:956-959.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1986a. Movement of subsurface fertilizer and limestone under irrigated and non-irrigated conditions. Soil Sci. 140:442-442.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., S. W. Van Scoyac and W. Kroontje. 1986b. Alfalfa seedling growth as influenced by Al, Ca and pH. Commun. Soil Sci. Plant Anal. 17:24-44.

Reid, D. A., A. L. Fleming, and C. D. Foy. 1971. A method of determining aluminum response of barley in nutrient solution in comparison to response in Al toxic soils. Agron. J. 63:600-603.

Rios, M. A. and R. W. Pearson. 1964. The effect of some chemical environmental factors on cotton behavior. Soil Sci. Soc. Am. Proc. 28:232-235.

Δ Robinson, W. O., G. Edgington, W. H. Armiger, and A. V. Breen. 1951. Availability of molybdenum as influenced by liming. Soil Sci. 72:267-274.

Rorison, I. H. 1965. Effect of aluminum on legume nutrition. p. 43-61. In E. G. Hallsworth (ed) Nutrition of the Legumes. London: Buttersworth Sci. Publ.

Santana, M. B. M. and J. M. Braga. 1977. Interacao aluminio - fosforo em um solo acido do sul da Bahia, Rev. ceres 24(132):200-211.

Sartain, J. B. and E. J. Kamprath. 1975. Effect of liming on a highly Al saturated soil on the top and root growth and soybean nodulation. Agron. J. 67:507-510.

Schmehl, W. R., M. Peech, and R. Bradfield. 1950. Causes of poor growth of plants on acid soils and beneficial effects of liming. Soil Sci. 70:393-410.

Schmehl, W.R., M. Peech, and R. Bradfield. 1952. Influences of soil acidity on absorption of calcium by alfalfa as revealed by radio calcium. Soil Sci. 73:11-21.

Sherman, G. D. and C. K. Fujimoto. 1947. The Effect of the use of lime, soil fumigants, and mulch on the availability of manganese in Hawaian soil. Soil Sci. Soc. Am. Proc. 11:206-210.

Siman, A., F. W. Addock, and W. A. Hudson. 1974. The development of manganese toxicity in pasture legumes under extreme climatic conditions. Plant Soil 41:129-140.

Simpson, J. R., A. Pinkerton, and J.Lazdovskii. 1977. Effects of subsoil calcium on the root growth of some lucerne genotypes (Medicago sativa L.) in acidic soil profiles. Aust. J. 28:629-638.

Skerman, P.J. 1977. Tropical Forage Legumes. Food and Agriculture Organization of the United Nations. Rome. p. 103-120.

Sumner, M. E. 1979. Response of alfalfa and sorghum to lime and P on highly weathered soils. Agron. J. 71:763-766.

Sumner, M. E., J. H. Bouton, J. Hammel, and H. Shahandeh. 1985. Enhanced alfalfa production through profile modification in the southeastern U. S. 1985 Forage and Grassland Conference. Hershey, PA.

Thorp, F. C. and J. A. Hobbs. 1956. Effect of lime aplication on nutrient uptake of alfalfa. Soil Sci. Soc. Am. Proc. 20:544-547.

Wallihan, E. F. 1948. The influence of aluminum on the phosphorus nutrition of plants. Am. J. Bot. 35:106-112.

Viets, F. G. 1944. Calcium and other polyvalent cations as accelerators of ion accumulation by excised barley roots. Plant Physiol. 19:406-480.

Vincent, J. M. 1965. Environmental factors in the fixation of nitrogen by the legume. p. 384-435. In W.V. Bartholomew and F. E. Clark (ed.) Soil Nitrogen. Am. Soc. Agron., Madison, WI.

Vlamis, J. 1953. Acid soil infertility as related to soil solution and solid phase effects. Soil Sci. 75:383-394.

Wallace, T., E. J. Hewitt, and D. J. D. Nicholes. 1945. Determination of factors injurious to plants in acid soils. Nature 156:778-779.

Watenpaugh, H. N. 1936. The influence of the reaction of soil strata upon the root development of alfalfa. Soil Sci. 41:449-468.

Wilkinson, H. F. 1972. Movement of micronutrients to plant roots. p. 139-169. In J.J. Mortvedt, P. M. Giordano, and W. L. Lindsay (ed.) Micronutrients in Agriculture. Soil Sci. Soc. Am., Madison, WI.

Wright, K. E. 1943. Internal precipitation of phosphorus inrelation to aluminum toxicity. Plant Physiol. 18:708-712.

Wright, K. E. and B. A. Donahue. 1953. Aluminum toxicity. Studies with radioactive phosphorus. Plant Physiol. 28:674-680.

Chapter III

GROWTH POUCH METHOD FOR STUDYING THE INFLUENCE OF CA, AL AND PH ON CROP GROWTH IN NUTRIENT SOLUTIONS

3.1 ABSTRACT

Response of plants to soil acidity is difficult to study under field conditions due to the complexity of soil systems. To circumvent this difficulty, it was thought that growth pouches, which have been used for nodulation and plant breeding studies, would be a practical method for studying the effects of acidity on alfalfa (Medicago sativa L.) growth, since single factors could be studied independently. Preliminary observations with commercial pouches showed that when pH 4.5 nutrient solutions remained in the pouches for as little as 5 h the pH increased to 6.5. Chemical analysis of a control nutrient solution (lacking Ca and Al) that remained in the pouches for 5 h contained Ca, Al and other ion contaminants. This indicated that the conventional pouch method is not reliable for Ca, Al and pH studies. Because of the potential for using the pouch method for nutrient studies, modifications of commercial pouches were investigated. Acid washing of the original brown paper wick in the pouch was not adequate for removing ion impurities. The problem was corrected by replacing the

brown paper wick with a white filter paper wick. Chemical analysis of solutions from these modified pouches showed no impurities and pH levels remained constant, making this an adequate method for Ca, Al and pH studies.

3.2 INTRODUCTION

Due to the complexity of soil systems and the difficulty in studying roots in a field situation, solution culture studies have been advocated as a preferred method for studying the influence of growth factors on crop development (Konzak et al., 1976). One of the main advantages of growing plants in solution cultures is that root systems can be observed in situ without the time consuming washing and cleaning procedure. Various types of containers and apparatus with different facilities to control environmental conditions have been described (Kemp, 1972; Rieley and Summerfield, 1972; Kendall and Leath, 1974; Summerfield and Minchin, 1976). These methods have been used for selecting Al tolerant varieties, due to the ease with which root growth may be studied (Foy et al., 1969; Reid et al., 1971; Kerridge et al., 1971). Most of these investigators used large containers for studying the crop response to nutrients. Large containers are adequate but they present a problem because of the very large space requirements. Use of large containers makes it very difficult to simultaneously study several treatments under controlled conditions. It would be advantageous to use a system capable of studying plants in nutrient solutions while utilizing minimal space. This chapter describes a method for studying the influence of Ca, Al and pH on crop growth utilizing plastic growth pouches.

3.3 MATERIALS AND METHODS

Commercial plastic growth pouches originally developed by Porter et al. (1966) for germination studies were used for this study. Pairs of the pouches were stapled together at three points along the top edge (two at the extreme outer edge to reduce evaporation) to facilitate handling and support. Each pair of pouches was placed over a metal bar (hanging folder) with one pouch suspended on each side. The hanging folders (such as those used for office files) were then suspended on file racks (Fig. 1). The racks were used to carry the pouches and occupied minimal space.

Experiment 1 investigated the effect of the brown wicked pouches on ion contamination and pH of the nutrient solution. Nutrient solutions used in this experiment contained 0.5 mM $MgSO_4$, 1 mM KCl, 10 uM FeDTPA, 5 uM NH_4PO_4, 1 mM NH_4NO_3, micronutrients and were adjusted to pH 4.5 as

Fig. 1 Alfalfa grown in growth pouches suspended from file rack.

described by Hohenberg and Munns (1984). Fifty mL of this nutrient solution were added to 4 replicates of pouches and allowed to equilibrate for 5 h.

Experiment 2 was initiated to determine if ion contaminants could be removed from the brown paper wick by acid washing or if a Whatman No. 4 white filter paper wick would eliminate ion contamination of the nutrient solution. Fifty mL of deionized water were placed in pouches that contained either the original intact brown paper wicks, brown paper wicks that had been acid washed with pH 2.8 solution or Whatman chromatography filter paper No. 4 wick equilibrated for 24 h. These treatments were replicated four times. The filter paper was cut to 19 cm by 13 cm and perforated to simulate the original paper wick. The paper was then folded along the holes and again 2 cm below the holes in order to make a V in the paper wick to allow for seed placement and growth. The filter paper was then autoclaved at 121°C for 20 minutes at 15 psi and placed in the pouch.

Experiment 3 was conducted to compare alfalfa (Medicago sativa) grown in brown vs white wicked pouches containing nutrient solution deficient in Ca at pH 4.5. Approximately 25 to 30 sterilized 'Arc' alfalfa seeds were placed in each of the growth pouches. Fifty mL of nutrient solution at pH

4.5 as described in the first experiment were added to each pouch. The rack of pouches was placed in a dark growth chamber at 25-30°C for 48 h, followed by 16/8 h light/dark cycles. Nutrient solutions were changed twice a week to assure sufficient nutrient levels and constant pH.

Solutions removed from pouches were analyzed for cations on an Inductively Coupled Plasma Spectrometer and anions using an Anion Chromatograph (Donohue and Gettier, 1979). In addition pH measurements were made on the solutions using a microprocessor ionanalyzer, equipped with a glass electrode assembly.

3.4 RESULTS AND DISCUSSION

3.4.1 Experiment 1

Preliminary studies indicated that the pH of the nutrient solutions in the growth pouches containing the original brown paper wicks increased from an initial pH of 4.5 to 6.5 after equilibrating in the pouches for 5 h.

Initially it was thought that the pH of the solutions increased due to atmospheric carbon dioxide. This hypothesis was soon discounted after pH studies conducted in glass beakers showed pH levels remained constant following equilibration of the nutrient solutions for 5 h. Analysis of the solutions from the pouches (lacking Ca and Al) revealed contamination of the nutrient solution with Ca and Al. Studies were conducted to ascertain the source of this contamination. Results of these studies showed the contamination was from the paper wick used in the pouches and not the pouch itself. This agrees with the findings of other investigators who reported that most absorbent papers, including those used for seed test laboratory germination purposes may contain high Al and often high dye concentrations which can contaminate the test solution at low pH (Konzak et al., 1976).

3.4.2 Experiment 2

Attempts were made to remove the impurities from the original paper wick by washing the paper with pH 2.8 solution, followed by rinsing with deionized water. Analysis of the solutions following the washings still showed the presence of ion contamination even though the pH remained constant at 4.5, making this method unsuitable for studies involving Ca, and Al (Figs. 2,3,4).

The growth pouches were then modified by replacing the original brown paper wick with a white filter paper wick. Analysis of the solutions from the modified pouches showed no contamination of either Al, Ca or other ions and pH levels of the solutions remained constant (Figs. 2,3,4).

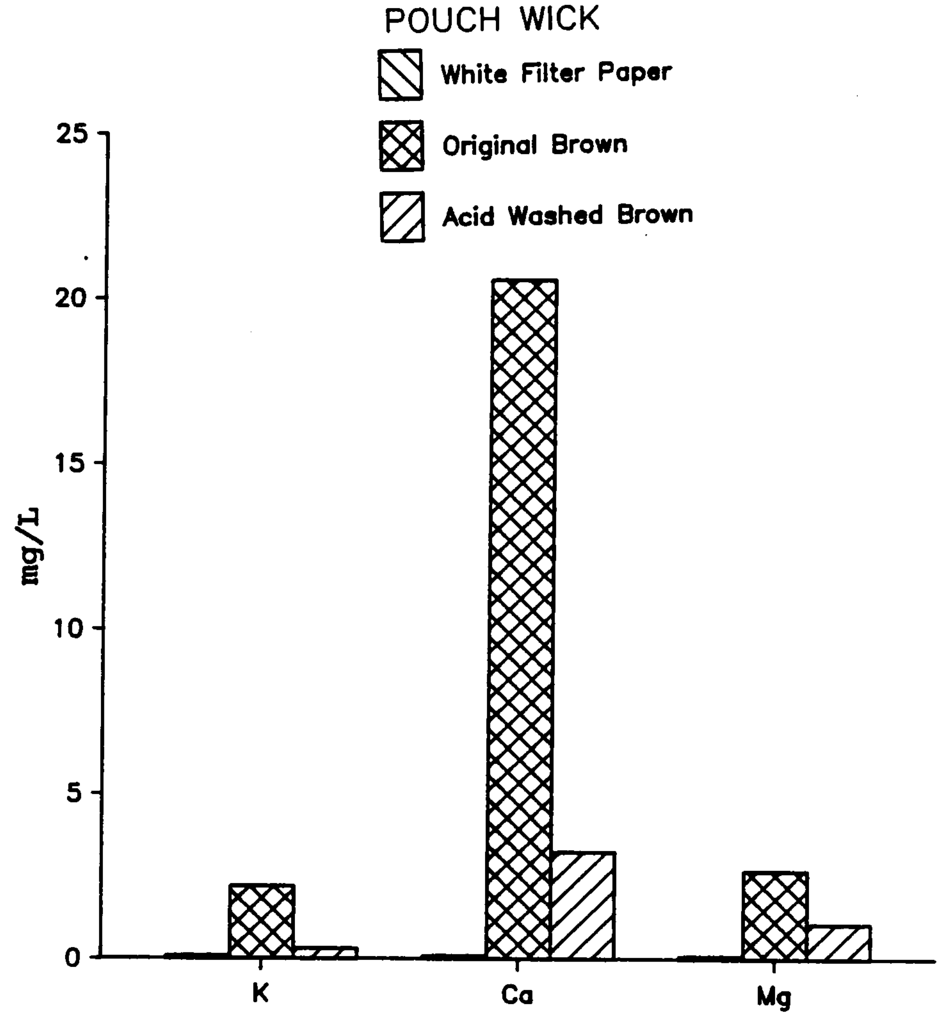


Fig. 2 Macronutrient impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis.

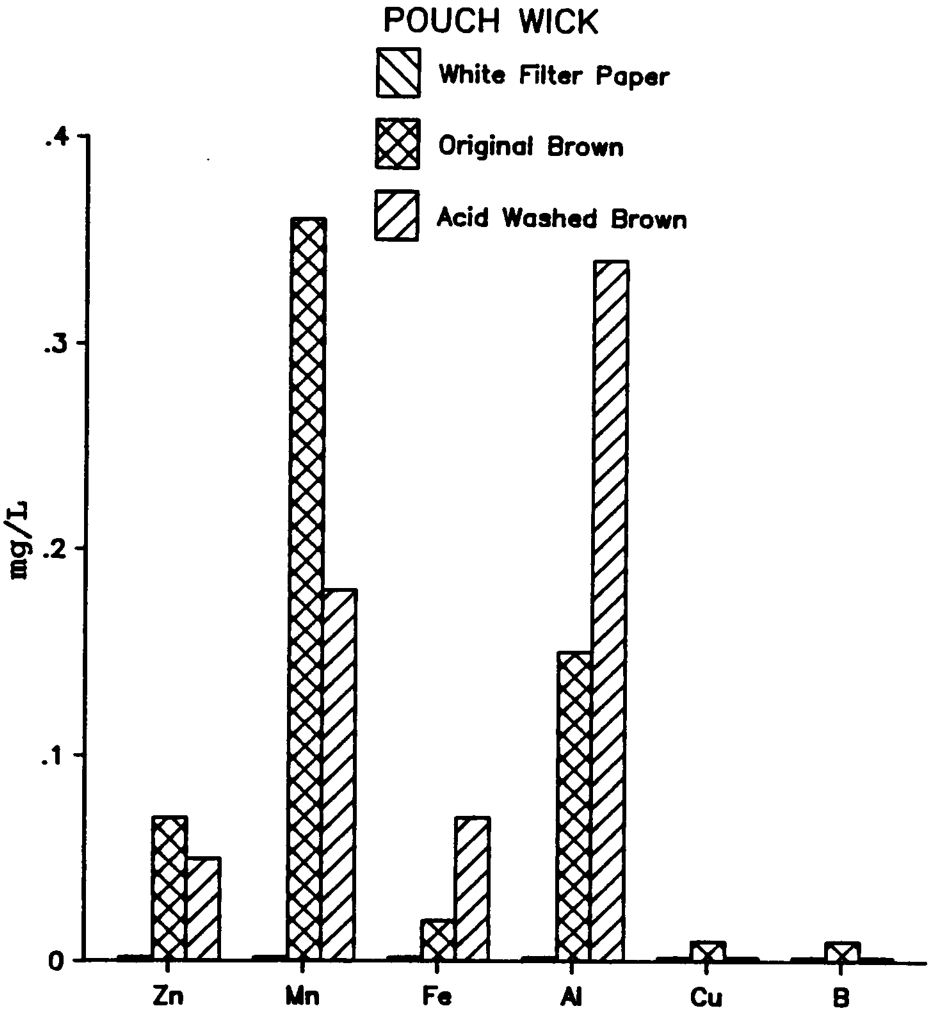


Fig. 3 Micronutrient impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis.

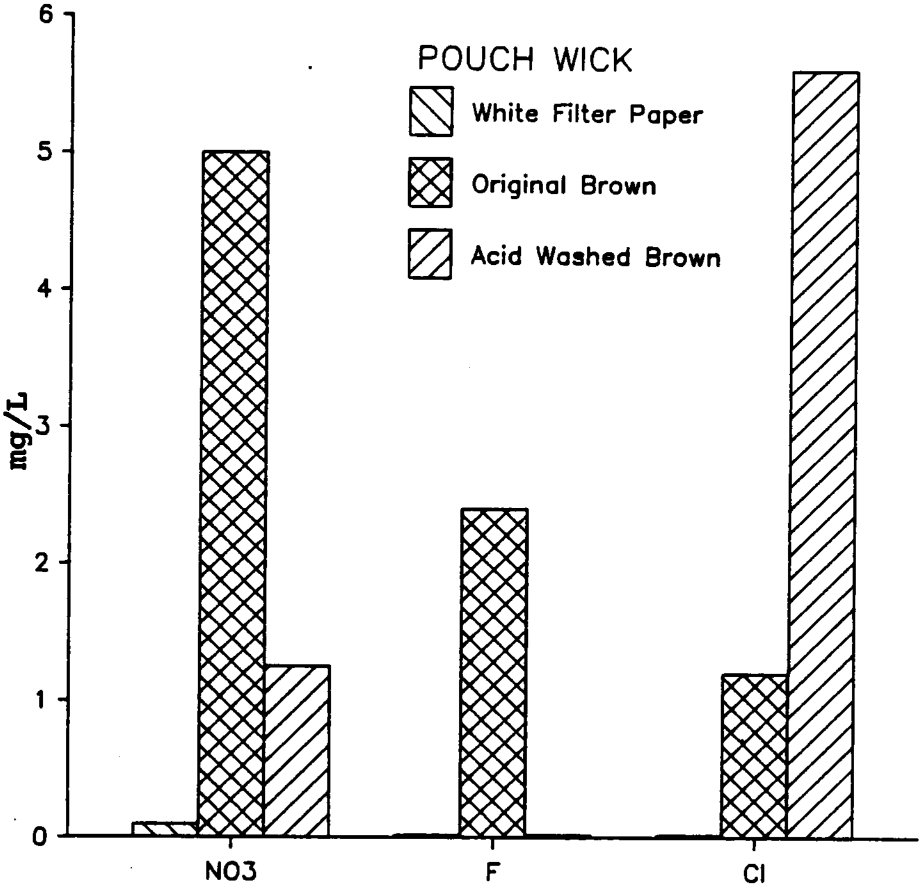


Fig. 4 Anion impurities in growth pouches containing different paper wicks. All pouches contained 50 ml of deionized water for 24 h prior to analysis.

3.4.3 Experiment 3

The comparison between alfalfa grown in brown vs white wicked pouches containing pH 4.5, Ca deficient nutrient solution showed that alfalfa roots and shoots grown in the white wicked pouches were stunted, typical of Ca deficiency (Fig. 5). However, alfalfa grown in brown wicked pouches showed extensive root and shoot growth indicating Ca contamination from the brown wick to be contributing to plant growth. This study clearly demonstrates that brown wick pouches are inadequate for Ca studies.

3.5 CONCLUSION

Use of the pouches containing white filter paper wicks were shown to be an effective method for studying the influence of Ca, Al and pH on alfalfa growth (Rechcigl et al., 1986). This method may also be applicable for studies involving Al tolerance of crops, and other studies involving the influence of other acidity factors on crop growth.

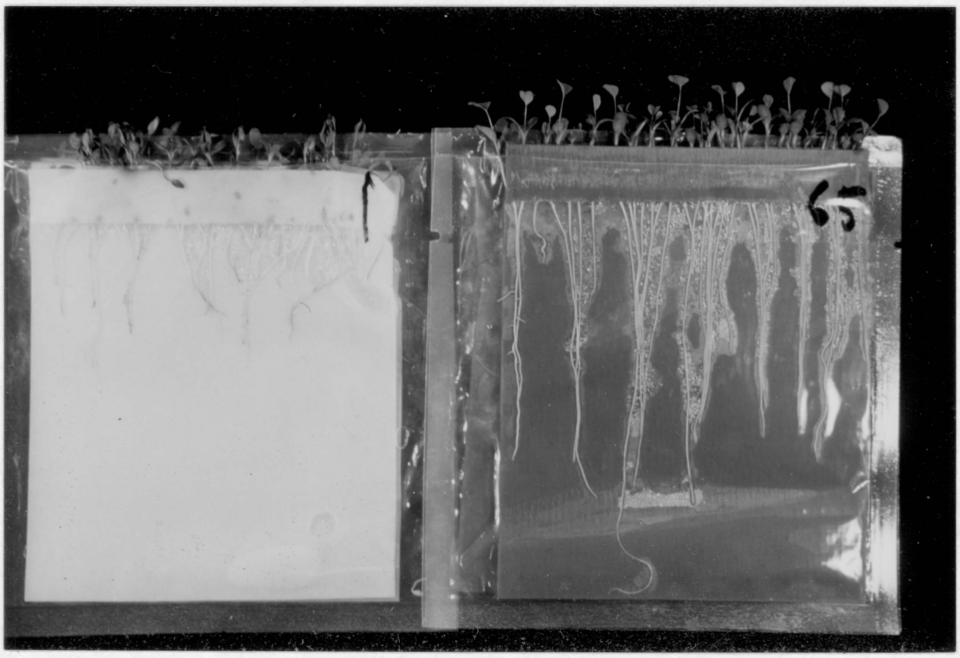


Fig. 5 Alfalfa grown with control nutrient solution (lacking Ca, Al) in modified growth pouch containing white filter paper wick (left) and original brown paper wick (right).

3.6 LITERATURE CITED

Donohue, S. J. and S. W. Gettier. 1979. Laboratory procedures Virginia Polytechnic Institute and State University Soil Testing and Plant Analysis Laboratory. Virginia Polytechnic Institute and State Univ. Pub. 853.

Foy, C. D., A. L. Fleming and W. H. Arminger. 1969. Aluminum tolerance of soybean varieties in relation to calcium nutrition. Agron. J. 61:505-511.

Hohenberg, J. S. and D. N. Munns. 1984. Effects of soil acidity factors on nodulation and growth of *Vigna unguiculata* in solution culture. Agron. J. 76:477-481.

Kemp, G. A. 1972. Waterbath for germination and root development studies under various temperature combinations of soil and ambient air. Can. J. Plant Sci. 52:672-679.

Kendall, W. A. and K. T. Leath. 1974. Slant-board culture methods for root observations of red clover. Crop Sci. 14:317-320.

Kerridge, P. C., M. D. Dawson, and D. P. Moore. 1971. Separation of degrees of aluminum tolerance in wheat. Agron. J. 63:586-591.

Konzak C. F., E. Polk, and J. A. Kttrick. 1976. Screening several crops for Al tolerance. p. 311-328. *In* M. J. Wright (ed.) Plant Adaptation to Mineral Stress in Problem Soils, Washington DC

Porter, F. E., I. S. Nelson, and E. K. Wold. 1966. Plastic pouches. Crops Soils 18:10.

Rechcigl, J. E., R. B. Reneau, Jr., D. D. Wolf, W. Kroontje and S. W. Van Scoyoc. 1986. Alfalfa seedling growth in nutrient solution as influenced by aluminum, calcium, and pH. Commun. Soil Sci. Plant Anal. 17:27-44.

Reid, D. A., A. L. Fleming, and C. D. Foy. 1971. A method for determining Al response of barley in nutrient solution in comparison to response in Al-toxic soil. Agron. J. 63:600-603.

Rieley, J. O. and R. J. Summerfield. 1972. The use of inert polymers in hydroponic studies. Plant Soil 37:183-185.

Summerfield, R. J. and F. R. Minchin. 1976. Root growth in cowpea [*Vigna unguiculata* (*L*.) *Walp*.] seedlings. Trop. Agric. 53:199-209.

Chapter IV

ALFALFA SEEDLING GROWTH IN NUTRIENT SOLUTIONS AS INFLUENCED BY ALUMINUM, CALCIUM AND PH

4.1 ABSTRACT

Soil acidity is a major factor responsible for reduced alfalfa (Medicago sativa L.) yields in humid regions of the United States. The purpose of this study was to investigate the effect of Ca, Al and pH interactions on alfalfa growth. Two experiments were conducted in modified plastic growth pouches. Experiment 1 studied the influence of pH (4.5 and 6.5) at varying levels of Al (0.00, 0.08 and 0.15 mM) with 1 mM Ca. Experiment 2 investigated the interaction between Ca (0.00, 0.30, 1.00, 3.00 and 6.00 mM) and Al (0.00, 0.08, 0.15 and 0.30 mM) at pH 4.5 on alfalfa shoot and root development. In the absence of Al, root and shoot growth were not affected by a pH of 4.5 as compared to a pH of 6.5. Increasing Al from 0.00 to 0.30 mM caused a reduction in both shoot and root growth at pH 4.5 when Ca was less than 3.00 mM. In the presence of 0.08 mM Al, additional Ca at 3.00 and 6.00 mM increased shoot growth equal to that of the 0.00 Al treatment. The optimum level of Ca for root development was at least 1.00 mM in the absence of Al; however, in the presence of 0.08 mM Al, additional Ca (≥3.00

mM) reduced Al toxic effects. Increasing levels of Ca (0.00 to 6.00 mM) decreased Al in roots from 9.4 to 4.7 g kg^{-1} in treatments containing 0.30 mM Al. Additional Ca (≥3.00 mM) in the pH 4.5 nutrient solution was beneficial for shoot and root growth in the presence of 0.08 mM Al and this pH was not detrimental to alfalfa seedling growth unless at least 0.08 mM Al was present.

4.2 INTRODUCTION

Soil acidity is a major growth limiting factor for agronomic crops in many parts of the world (Foy, 1984; Adams, 1981). Numerous studies have demonstrated that Al toxicity is the major factor involved in reduced crop yields on acid soils with a pH of less than 5.5 (Foy, 1984). Studies conducted by Franco and Munns (1982) have shown that as little as 0.02 mM Al reduced shoot and root growth of bean (Phaseolus vulgaris L.) plants. The problems of acidity are especially acute in legumes, particularly with alfalfa (Medicago sativa L.), because of a deep rooting habit and sensitivity of the symbiotic N fixing bacteria to low pH (Munns, 1978). Extreme acidity in subsoils has been shown to be harmful to alfalfa because it promotes shallow rooting, increased drought susceptibility and reduced use of subsoil nutrients (Rechcigl et al., 1985a,b).

Investigators have generally taken a rather simplistic view of the problem of plant growth in acid soils. Reduced crop yields are normally assumed to be due to Al toxicity and thus increasing the pH to alleviate the condition has been advocated. Although this may be true in many cases, one cannot surmise that this would hold true for every situation. Some investigators have clearly shown that reduced plant growth in acid soils may be corrected by the addition of Ca (Colwell and Brady, 1945), while others have corrected the problem by the addition of N (Andrew, 1978). There is also evidence that alfalfa roots may penetrate acidic subsoils to a depth of 84 cm in the absence of subsurface lime if surface limed (Rechcigl and Reneau, 1984). Low pH (5.1) and high exchangeable Al (220 mg kg^{-1}) in the subsoil did not appear to be harmful to alfalfa (Rechcigl et al., 1985b) when the soil was surface limed. In that study Al toxicity may have been averted by Ca supplied by surface applied lime. The purpose of this study was to investigate the effect of Ca, Al and pH interactions on alfalfa performance. Because of the complexity of soil systems and the need for maintaining known Ca, Al and pH levels, the present study was conducted using nutrient solutions in plastic growth pouches.

4.3 MATERIALS AND METHODS

4.3.1 Pouch Preparation

Two growth pouches[1] were stapled together and placed over a hanging folder. Hanging folders such as those used for office files were then suspended on file racks (Fig. 1).

Growth pouches were modified by replacing the original paper wick with a sheet of chromatography filter paper. The filter paper was cut to 19 by 13 cm and perforated to simulate the original paper. The paper was folded along the perforations and again 2 cm below the perforations in order to make a V in the paper wick to allow for seed placement. The filter paper was then autoclaved at 121°C for 20 min at 1.0×10^5 Pa and placed in the pouch. Approximately 25 to 30 'Arc' alfalfa seeds and 50 mL of the nutrient solution for each test were placed in each of the modified growth pouches. Nutrient solutions for both experiments described below contained 0.50 mM $MgSO_4$, 1.00 mM KCl, 10.00 uM FeDTPA, 5.00 mM $NH_4H_2PO_4$, 1.00 mM NH_4NO_3 and micronutrients as described by Hohenberg and Munns (1984). The rack of pouches was placed in a dark growth chamber at 25-30°C for 48 h; after this initial period 16/8 h light and dark periods were alternated.

Two experiments were conducted. Experiment 1 tested the influence of pH (4.5 and 6.5) and varying levels of Al (

0.00, 0.08 and 0.15 mM) with 1 mM Ca on alfalfa shoot and root growth. Experiment 2 tested the influence of several levels of Ca (0.00, 0.30, 1.00, 3.00 and 6.00 mM) and Al (0.00, 0.08, 0.15 and 0.30 mM) at pH 4.5 on alfalfa seedling development. Both experiments were replicated four times in a randomized complete block design. The pH of the nutrient solutions were monitored daily and the solutions were changed after 3.5 days to assure a constant pH and optimal nutrient levels.

Alfalfa growth was evaluated after 2 weeks by visual scoring, root penetration and shoot and root dry weight in both experiments. In addition Al, Ca and P concentration in the tissues were measured in the second experiment. Root samples were rinsed three times in distilled water and then dried at 89 to 93°C for 12 h. Shoot tissue was dried in the same manner as roots. Following tissue digestion with nitric and perchloric acids (Johnson and Ulrich, 1959), analyses for Al, Ca and P were conducted for both root and shoot tissue. Aluminum was analyzed by the aluminon procedure (Jayman and Sivasubramaniam, 1974). Calcium concentration was determined with a Perkin-Elmer atomic absorption spectrophotometer, and P was determined by the sulfuric molybdate colorimetric method (Jackson, 1958). All data were statistically analyzed, utilizing the Statistical Analysis System (Council and Sall, 1982).

4.4 RESULTS AND DISCUSSION

4.4.1 Al Concentration and pH

Shoot growth and root penetration did not differ at pH 4.5 or 6.5 in the absence of Al (Table 1). This is in agreement with the studies of Kamprath and Foy (1985), and Moore (1974) who have concluded that Al toxicity is more important than H toxicity in limiting the growth of crops in acid soils (pH > 4.0). Hydrogen toxicity has been shown; however, to be detrimental to crops in soils with pH levels below 4.0 (Arnon et al., 1942 ; Isam et al., 1970).

Root penetration, shoot and root dry weight and visual scorings were not affected by Al concentration at pH 6.5 (Table 1). This is expected since Al normally precipitates at pH values greater than 5.5, and consequently is not toxic to alfalfa. These results are in agreement with studies of Hohenberg and Munns (1984) who observed no changes in root and shoot growth of *Vigna unguiculata* L. under comparable conditions. Alfalfa grown in pouches containing varying Al concentrations at pH 4.5 had reduced root penetration and visual shoot scorings with increased Al concentration. Even though root penetration decreased with increased Al concentration root dry weights between treatments were not different.

Table 1. Alfalfa shoot and root growth as affected by pH and Al in nutrient solutions.

Al[†] conc.	pH	Visual[‡] scorings (1 week)	Root penetration (1 week)	Visual scorings (2 week)	Root penetration (2 weeks)	Shoot dry weight (2 weeks)	Root dry weight (2 weeks)
(mM)			(cm)		(cm)	------ g/pouch	------
0.00	6.5	6.8	10.0	7.8	12.0	0.10	0.03
0.00	4.5	6.4	9.5	8.2	11.8	0.10	0.03
0.08	6.5	6.2	9.6	7.8	12.0	0.10	0.03
0.08	4.5	6.2	8.1	6.8	9.0	0.10	0.03
0.15	6.5	6.5	9.2	8.0	11.7	0.10	0.04
0.15	4.5	5.0	5.9	6.2	6.8	0.08	0.03
$LSD_{(0.05)}$	=	NS[#]	0.8	1.0	0.8	NS	NS

[†] All treatments contained 0.5 mM $MgSO_4$, 1 mM KCL, 1 mM $CaSO_4$, 10 µM FeDTPA, 5 µM $NH_4H_2PO_4$, 1 mM NH_4NO_3 and micronutrients.

[‡] Visual scorings ranged from 1 (poorest growth) to 10 (best growth).

[#] NS Not significant at the 0.05 level.

4.4.2 Calcium Influence on Al Toxicity

Visual scorings of plants and measured root length after one week were greater for plants that received Ca compared to plants without Ca, while increased Al levels significantly reduced root length (Table 2). Visual scorings of plants taken the second week indicated significant differences for Ca and Al treatments. Visual scorings indicated that plants grew best at high Ca levels in the absence of Al (Fig. 6). In the presence of 0.08 mM Al, 6.00 mM Ca increased growth equal to that of the treatment with no Al. Root penetration was significantly affected by Ca, Al and the Ca and Al interaction. Root penetration was reduced as Al increased (Fig. 7). Root penetration increased in pouches containing increased levels of Ca at all levels of Al. The optimum level of Ca for root development in the absence of Al was 1.00 mM; however, in the presence of 0.08 mM Al, additional Ca (≥3.00 mM) reduced Al toxic effects which normally stunt roots. This is in agreement with the work of Lund (1970) who observed that increasing the Ca level from 0.25 to 1.00 mM decreased soybean root (Glycine max L.) injury caused by Al added at 0.02, 0.04 or 0.08 mM. Similar findings have been made by other investigators (Mathur et al., 1983; Simpson et al., 1978; Wallace et al., 1980). There were also trends for

Table 2. Alfalfa shoot and root growth after 1 and 2 weeks as affected by Ca and Al in acidic (pH 4.5) nutrient solutions.

Treatment[+]		Visual[++] scorings (1 week)	Root penetration (1 week)	Shoot dry weight (2 weeks)	Root dry weight (2 weeks)
Element	Conc.				
Ca	(mM)		cm	g/pouch	
	0.00	2.8	4.4	0.05	0.01
	0.30	4.7	5.7	0.08	0.03
	1.00	5.5	6.1	0.08	0.03
	3.00	5.2	6.1	0.09	0.04
	6.00	5.3	6.2	0.10	0.04
$LSD_{(0.05)}$		0.5	0.6	0.02	0.01
Al	(mM)				
	0.00	4.9	6.1	0.09	0.04
	0.08	4.8	6.4	0.08	0.03
	0.15	4.6	5.5	0.08	0.03
	0.30	4.5	4.8	0.07	0.03
$LSD_{(0.05)}$		NS[+++]	0.5	NS	0.01

[+] All treatments contained 0.5 mM $MgSO_4$, 1 mM KCL, 10 µM FeDTPA, 5 µM $NH_4H_2PO_4$, 1 mM NH_4NO_3 and micronutrients.

[++] Visual scorings ranged from 1 (poorest growth) to 10 (best growth).

[+++] NS Not significant at the 0.05 level.

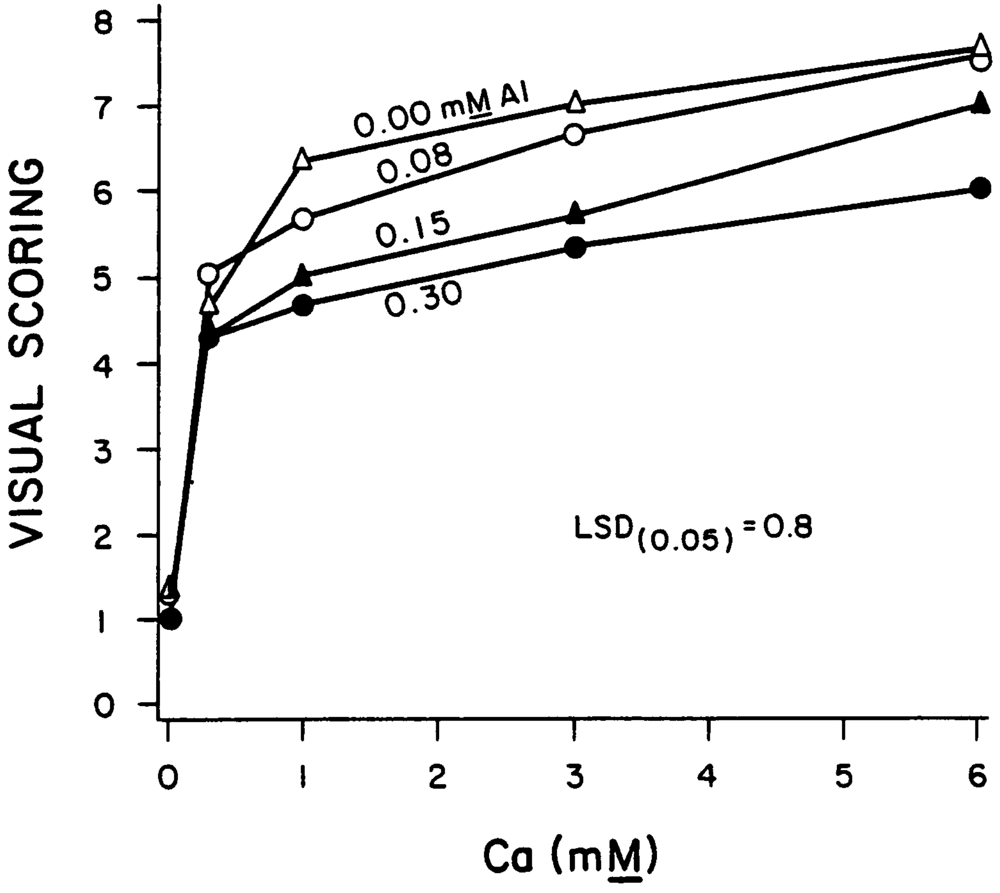


Fig. 6 Calcium and Al effects on alfalfa growth two weeks after germination. Visual scorings ranged from 1 (poorest growth) to 10 (best growth).

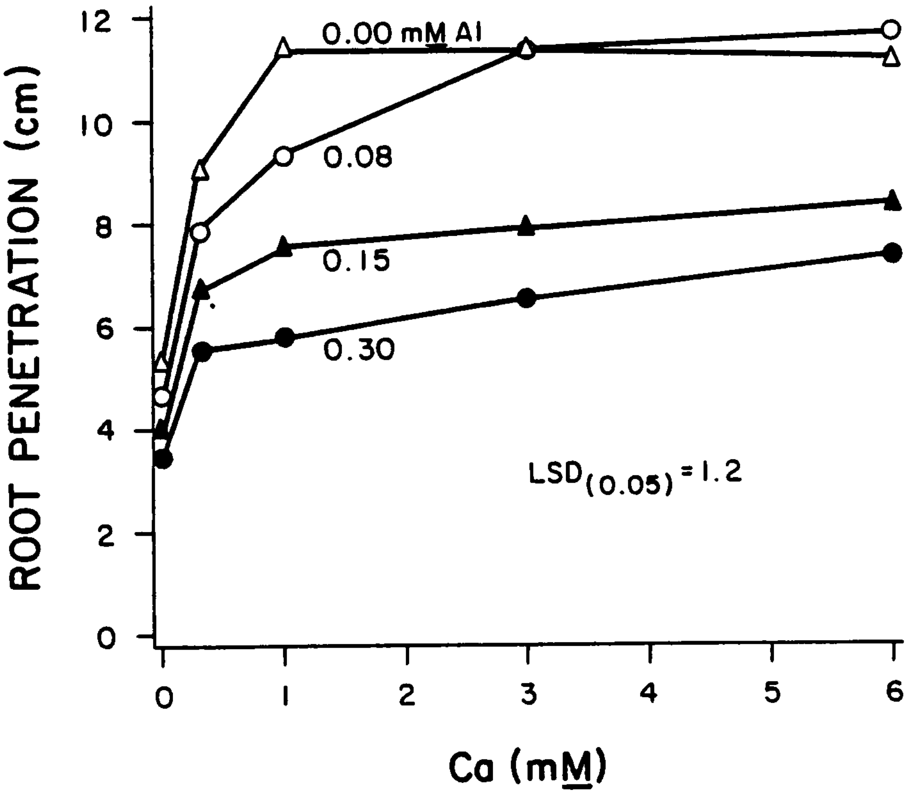


Fig. 7 Calcium and Al effects on alfalfa root penetration two weeks after germination.

shoot and root weights showing the benefits of Ca and the toxicities of Al (Table 2).

Levels of Al in roots varied with both Ca and Al treatments (Fig. 8). Aluminum content of roots increased as the Al in solutions increased. Increasing the concentrations of Ca in nutrient solutions (0.00 to 6.00 mM), decreased the Al content of the root. Aluminum content in roots grown in the 0.30 mM Al nutrient solutions decreased from over 9.4 to 4.7 g Al kg^{-1} in pouches containing increased levels of Ca (from 0.00 to 6.00 mM). This shows that Ca can reduce Al uptake by the plant roots. This may account for the increased root penetration in the presence of high Al levels in pouches containing increased levels of Ca.

Calcium concentration of the roots increased from 1.6 to 28 g kg^{-1} with increased Ca concentrations in the nutrient solution (Table 3). Varying levels of Al in nutrient solution did not affect the Ca concentration of the roots.

Phosphorus in the root was increased with increasing levels of Al. This is expected since Al has been reported to precipitate P as aluminum phosphate on the root surface (Foy, 1984). Increasing levels of Ca in the nutrient solution reduced P concentration in the root. This may be

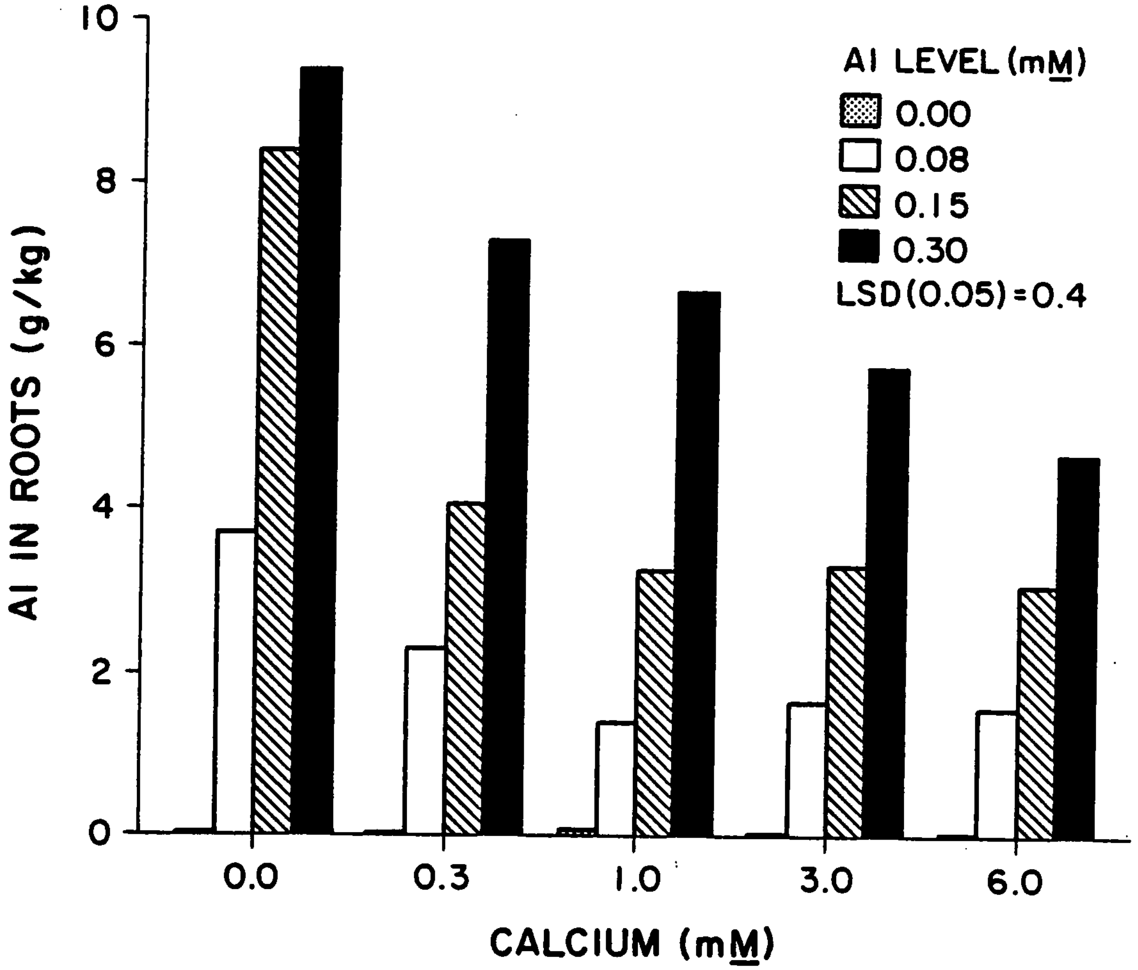


Fig. 8 Calcium and Al effects on Al concentration in alfalfa roots two weeks after germination.

Table 3. Alfalfa shoot and root chemical composition as affected by Ca and Al in acidic (pH 4.5) nutrient solutions.

Al^{+} conc.	Ca conc (mM)				
	0.00	0.30	1.00	3.00	6.00
(mM)	Ca in roots (g kg^{-1})				
0.00	2.0	2.9	4.1	8.7	25.5
0.08	1.5	2.8	4.7	9.2	25.4
0.15	1.5	2.3	4.8	12.4	28.9
0.30	1.3	2.0	4.0	9.6	30.0
LSD$_{(0.05)}$ 10.3					
	P in roots (g kg^{-1})				
0.08	4.1	2.9	2.8	2.7	2.8
0.08	4.3	3.2	3.4	2.9	2.5
0.15	4.0	3.4	3.5	3.3	2.9
0.30	5.4	4.0	4.4	3.6	3.0
LSD$_{(0.05)}$ 1.0					
	Ca in shoots (g kg^{-1})				
0.00	0.08	1.4	2.8	5.7	8.1
0.08	0.07	1.4	2.6	5.6	7.7
0.15	0.8	2.3	3.2	4.6	8.2
0.30	0.6	2.3	2.9	3.8	6.0
LSD$_{(0.05)}$ 1.5					
	P in shoots (g kg^{-1})				
0.00	3.0	2.2	2.0	2.1	2.3
0.08	2.8	2.0	2.4	2.0	1.8
0.15	2.5	2.2	2.4	2.2	2.1
0.30	3.0	2.5	2.6	2.3	2.3
LSD$_{(0.05)}$ 0.4					
	Al in shoots (mg kg^{-1})				
0.00	0	0	0	86	0
0.08	0	41	52	193	5
0.15	0	146	41	27	15
0.30	0	57	141	35	140
LSD$_{(0.05)}$ *NS*$^{++}$					

$^{+}$All treatments contained 0.5 mM $MgSO_4$, 1 mM KCL, 10µM FeDTPA, 5 µM $NH_4H_2PO_4$, 1 mM NH_4NO_3 and micronutrients.

$^{++}$NS Not significant at the 0.05 level.

due to differences in root weights, in that we would expect greater levels of P in a stunted root as opposed to a larger root where P is less concentrated. Also increased Ca would lower Al activity and result in reduced Al phosphate precipitation.

Calcium was increased in the shoots from 0.70 to 8.00 g kg^{-1} in pouches containing higher levels of Ca (0.00 to 6.00 mM). Shoot P was reduced with the addition of Ca while Al did not appear to have any influence on P content of shoots. The reduction in P concentration is probably due to dilution with increased growth.

There was a trend for Al content of shoots to increase with increasing levels of Al in nutrient solution, however, the differences were not significant. There did not appear to be any relationships between the Ca and Al content of shoots.

4.5 CONCLUSIONS

Based on these experiments, the following conclusions can be drawn: 1) pH 4.5 is not detrimental to alfalfa seedling performance unless Al is present; 2) Aluminum was not toxic to alfalfa growth at pH 6.5; 3) Calcium supplied at 1.00 mM optimized root growth and resulted in near maximum top growth in absence of Al. However with the presence of 0.08 mM Al, 3.00 mM Ca was required for root and top growth to equal that produced where 1.00 mM Ca and no Al was used.

FOOTNOTES

[1] Northrup King, 1500 Jackson St., N. E., Minneapolis, MN 55413

4.6 LITERATURE CITED

Adams, F. 1981. Nutritional imbalances and constraints to plant growth in acid soils. J. Plant Nutr. 4:81-87.

Andrew, C. S. 1978. Mineral characterization of tropical forage legume. p. 93-112. In C. S. Andrews and E. J. Kamprath (ed.) Mineral nutrition of legumes in tropical and subtropical soils. Est Melbourne, Australia, CSIRO.

Arnon, D. I., W. E. Fratzke, and C. M. Johnson. 1942. Hydrogen ion concentration in relation to absorption of inorganic nutrients by higher plants. Plant Physiol. 17:515-524.

Colwell, W. E. and N. C. Brady. 1945. The effect of calcium on yield and quality of large seeded type peanuts. Agron. J. 37:413-28.

Council, K. A. and J. P. Sall (ed.). 1982. SAS User's Guide. Statistical Analysis Systems Institute Inc., Cary, N. C.

Foy, C. D. 1984. The physiology of metal toxicity in plants. p.57-86. In F. Adams (ed.) Soil Acidity and Liming. ASA Monograph Agronomy 12, second edition. American Society of Agronomy, Madison, WI.

Franco, A. H. and D. N. Munns. 1982. Acidity and aluminum restraints on nodulation, nitrogen fixation and growth of Phaseolus vulgaris in nutrient solution. Soil Sci. Soc. Am. J. 46:296-301.

Hohenberg, J. S. and D. N. Munns. 1984. Effects of soil acidity factors on nodulation and growth of Vigna unguiculata in solution culture. Agron. J. 76:477-481.

Islam, A. K. M. S., D. G. Edwards, and C. J. Asher. 1980. pH optima for crop growth: Results of a flowing solution culture experiment with six species. Plant Soil 54:339-357.

Jackson, M. L. 1958. Soil Chemical Analysis. Prentice-Hall, Inc. Englewood Cliffs, NJ

Jayman, T. C. Z. and S. Sivasubramaniam. 1974. The use of ascorbic acid to eliminate interference from iron in the aluminon method for determining aluminum in plant and soil extracts. Analyst 99:296-301.

Johnson, C. M. and A. Ulrich. 1959. Plant analysis. Bull. Calif. Agric. Exp. Sta. No. 766.

Kamprath, E. J. and C. D. Foy. 1985. Lime fertilizer-plant interactions in acid soils. In O. P. Engelstad (ed.) Fertilizer technology and use. 3rd edition. Soil Science Society of America, Madison, WI.

Lund, Z. F. 1970. The effect of calcium and its relation to several cations in soybean root growth. Agron. J. 34:456-459.

Mathur, B. S., H. Sinha, N. K. Rana, and S. Lall. 1983. Use of calcium as a nutrient for legume crops in acidic red loam soils of Ranchi. J. Indian Soc. Soil Sci. 31:334-36.

Moore, D. P. 1974. Physiological effects of pH on roots. p. 565-600. In E. W. Carson (ed.) The plant root and its environment. University Press of Virginia, Charlottesville, VA.

Munns, D. N. 1978. Soil acidity and nodulation. p. 247-263. In C. S. Andrew and E.J. Kamprath (ed.). Mineral nutrition of legumes in tropical and subtropical soils. Commonwealth Scientific and Industrial Research Organization, Australia.

Rechcigl, J. E. and R. B. Reneau, Jr. 1984. Effects of subsurface acidity on alfalfa in a Tatum clay loam. Commun. Soil Sci. Plant Anal. 15:811-818.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985a. Effect of subsurface amendments and irrigation on alfalfa growth. Agron J. 77:72-75.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., and W. Kroontje. 1985b. Influence of surface liming on the yield and nutrient concentration of alfalfa established using no-tillage techniques. Agron J. 77:956-959.

Simpson, J. R., A. Pinkerton, and J. Lazdorski. 1978. Effects of subsoil calcium on the root growth of some lucerne genotypes (*Medicago sativa*) in acidic soil profiles. Aust. J. Agric. Res. 28:629-38.

Wallace, A., E. M. Romney, R. T. Mueller and G. V. Alexander. 1980. Calcium-trace metal interactions in soybean plants. J. Plant Nutrition 2:79-86.

Chapter V

NO-TILL ALFALFA YIELD AND NUTRIENT CONCENTRATION AS INFLUENCED BY SURFACE LIME ON AN ACID ERNEST SILT LOAM SOIL

5.1 ABSTRACT

Soil acidity is believed to be a major cause of limited root penetration and low yields for alfalfa (Medicago sativa L.). Alfalfa was seeded in April 1981 and 1982 into an orchardgrass sod (Dactylis glomerata L.) to evaluate the influence of surface applied limestone on no-till alfalfa growth under acidic soil conditions. Dolomitic agricultural limestone was surface applied at 0 or 6.7 Mg ha^{-1} to an Ernest silt loam soil (fine-loamy, mixed, mesic Aquic Fragiudult) either 6 or 18 months before planting (April 1981 or 1982) or at planting. Yield (from 1982 through 1984) increases, greater than 2 fold, resulted from surface application of limestone. Time of lime application (6 months or 18 months prior to planting or at planting) had no influence on yields. Roots were more prevalent at the 1.0 and 1.5 m depths as a result of surface lime application. Soil pH (0 to 10 cm depth) ranged from 6.2 to 6.9 in the surface limed plots as compared to 5.3 to 5.6 in the unlimed plots, regardless of time of lime application. The pH averaged 5.1 below 45 cm irrespective of lime application.

Aluminum levels ranged from 21 at the 0 to 5 cm depth to 186 mg kg^{-1} at the 145 to 150 cm depth on unlimed plots. Lime application reduced Al levels by 2 to 20 fold in the upper 15 cm, but had no influence on Al levels at depths of 95 cm and greater. Calcium levels were elevated in the top 15 cm as a result of lime application and then decreased with increasing depth. This study indicates that no-till alfalfa may be grown on acid soils containing high Al, without lime incorporation prior to seeding, provided surface lime is applied.

5.2 INTRODUCTION

No-till farming is becoming increasingly popular as a conservation practice. In addition to conserving valuable top soil, no-tillage methods save the producer time and machinery cost. No-till practices for corn (Zea mays L.) and soybeans (Glycine max, Merr.) are well established and widely accepted (Brann et al., 1983; Baylor, 1983). However, no-till establishment procedures for small-seeded legumes such as alfalfa (Medicago sativa L.) are only now being developed (Baylor, 1983; Mueller and Chamblee, 1984; Sperow, 1983; White et al., 1982).

No-till alfalfa is recommended only if the soil pH is above 6.5 (White et al., 1983). Extreme acidity in subsoils has been shown to be harmful to alfalfa because of shallow rooting, resulting in drought susceptibility, and poor use of subsoil nutrients (Rechcigl et al., 1985; Kauffman, 1977).

Studies have shown yield increases in alfalfa on acidic soil with plowsole application of lime (Rechcigl et al., 1985), or by mixing lime in the top 15 cm (Kauffman, 1977). There is also some evidence that alfalfa roots may penetrate acidic subsoils, in the absence of plowsole lime when surface limed (Rechcigl and Reneau, 1984). In the latter study alfalfa roots penetrated soil to 84 cm, where the pH was 4.1 and the exchangeable Al was 225 mg kg^{-1}.

Liming studies with alfalfa have used conventional tillage practices where lime if needed is incorporated during preplant field operations. The objective of this paper is to evalute the influence of surface lime application on alfalfa growth when no-till planted in acidic soil conditions.

5.3 MATERIALS AND METHODS

'Arc' alfalfa was no-till planted into orchardgrass (Dactylis glomerata L.) in an Ernest silt loam soil (fine-loamy, mixed, mesic Aquic Fragiudult) at the Agronomy Research Farm near Blacksburg, VA. Plantings were made in the third week of April 1981 and 1982. A contact herbicide was applied the day before planting. A no-till drill was used to place the seed at a 2.5 cm depth in 20 cm rows at a rate of 16.5 kg ha^{-1}. Dolomitic agricultural limestone was surface applied either 6 or 18 months before planting (April 1981 or 1982) or at planting. A randomized complete block design included four replications. Harvests and fertility programs were selected to maintain productive stands.

Composite soil samples were collected from 0 to 5, 5 to 10, 10 to 15, 45 to 50, 95 to 100 and 145 to 150 cm depths. Soil samples were air dried and crushed to pass a 2-mm sieve. Soil pH and Ca measurements were conducted as reported by Donohue and Gettier (1979). For Al determination, 5 g of soil were shaken with 50 ml of $\underline{M}$ KCl for 48 h and the solution analyzed by the aluminon procedure (Jayman and Sivasubramaniam, 1974).

Plant samples from 3 harvests in 1984 were dried at 65°C for 12 h and ground in a stainless steel mill to pass a 20-mesh sieve. Nitrogen, Ca, P and Mn were determined as described by Donohue and Gettier (1979) and Al as described Jayman and Sivasubramaniam (1974).

Trenches were excavated in limed and unlimed plots planted in April 1982 to measure the depth of root penetration. Roots were measured at the 0.1, 0.5, 1.0 and

1.5 m depth utilizing the foil method to count root intersections on a 21 by 28 cm grid (Bohm, 1979). Roots were then sampled and washed 10 times in distilled water to remove soil contaminants and tested qualitatively for Al using the Quinalizarine method (Kalavoulos and Misopolinas, 1977).

Data were analyzed by analysis of variance utilizing the Statistical Analysis System (Council and Sall, 1982) procedures, and least significant differences were calculated for mean separation at the 0.05 level of probability.

5.4 RESULTS AND DISCUSSION

5.4.1 Soil Acidity

Surface limed plots had an average pH of 6.8 in the surface 5 cm, in comparison to 5.5 for unlimed soil (Table 4). The pH decreased with increasing depth regardless of lime application. There were no differences in pH values between limed and unlimed plots at depths of 45 cm or greater, the average being 5.1. Exchangeable Al ranged from 21 to 186 mg kg^{-1} at increasing depths in unlimed soils (Table 4). Surface application of lime reduced the Al by 2 to 20 fold in the top 15 cm. At depths of 95 cm and greater there were no significant differences in Al concentration

Table 4. Influence of application date of surface-applied agricultural limestone on soil pH, exchangeable Al and Ca in an Ernest silt loam soil.

Date of planting	Lime application		Depth (cm)					
	Rate	Date	0-5	5-10	10-15	45-50	95-100	145-150
	Mg ha^{-1}		pH					
Apr. 1981	0.0	—	5.9	5.6	5.6	5.0	—	—
	6.7	Apr. 1981	6.9	6.7	6.2	5.1	—	—
	6.7	Aug. 1980	6.9	6.8	6.7	5.1	—	—
LSD(0.05)			0.4	0.6	0.3	NS+	—	—
CV(%)			2.5	4.0	2.2	1.4	—	—
Apr. 1982	0.0	—	5.4	5.3	5.2	5.0	5.0	5.1
	6.7	Apr. 1982	6.6	6.4	6.0	5.4	5.2	5.2
	6.7	Aug. 1980	7.0	6.7	6.1	5.2	5.2	5.1
LSD(0.05)			1.0	0.6	0.5	NS+	NS	NS
CV(%)			6.8	4.2	3.8	3.4	2.1	2.9
			Al (mg kg^{-1})					
Apr. 1981	0.0	—	23	36	42	99	—	—
	6.7	Apr. 1981	1	2	7	69	—	—
	6.7	Aug. 1980	1	3	9	62	—	—
LSD(0.05)			4	10	24	NS	—	—
CV(%)			27	32	54	71	—	—
Apr. 1982	0.0	—	22	38	97	186	166	166
	6.7	Apr. 1982	7	6	8	48	166	182
	6.7	Aug. 1980	2	2	1	60	126	190
LSD(0.05)			12	15	NS	38	NS	NS
CV(%)			54	44	118	9	28	17
			Ca (mg kg^{-1})					
Apr. 1981	0.0	—	305	276	271	330	—	—
	6.7	Apr. 1981	870	498	408	415	—	—
	6.7	Aug. 1980	732	414	282	364	—	—
LSD(0.05)			141	203	62	NS	—	—
CV(%)			5.2	11.9	4.5	6.8	—	—
Apr. 1982	0.0	—	395	329	304	354	486	426
	6.7	Apr. 1982	632	384	356	408	444	348
	6.7	Aug. 1980	752	432	316	360	390	380
LSD(0.05)			203	NS	NS	NS	NS	NS
CV(%)			15.1	17.6	10.0	8.8	31.1	34.2

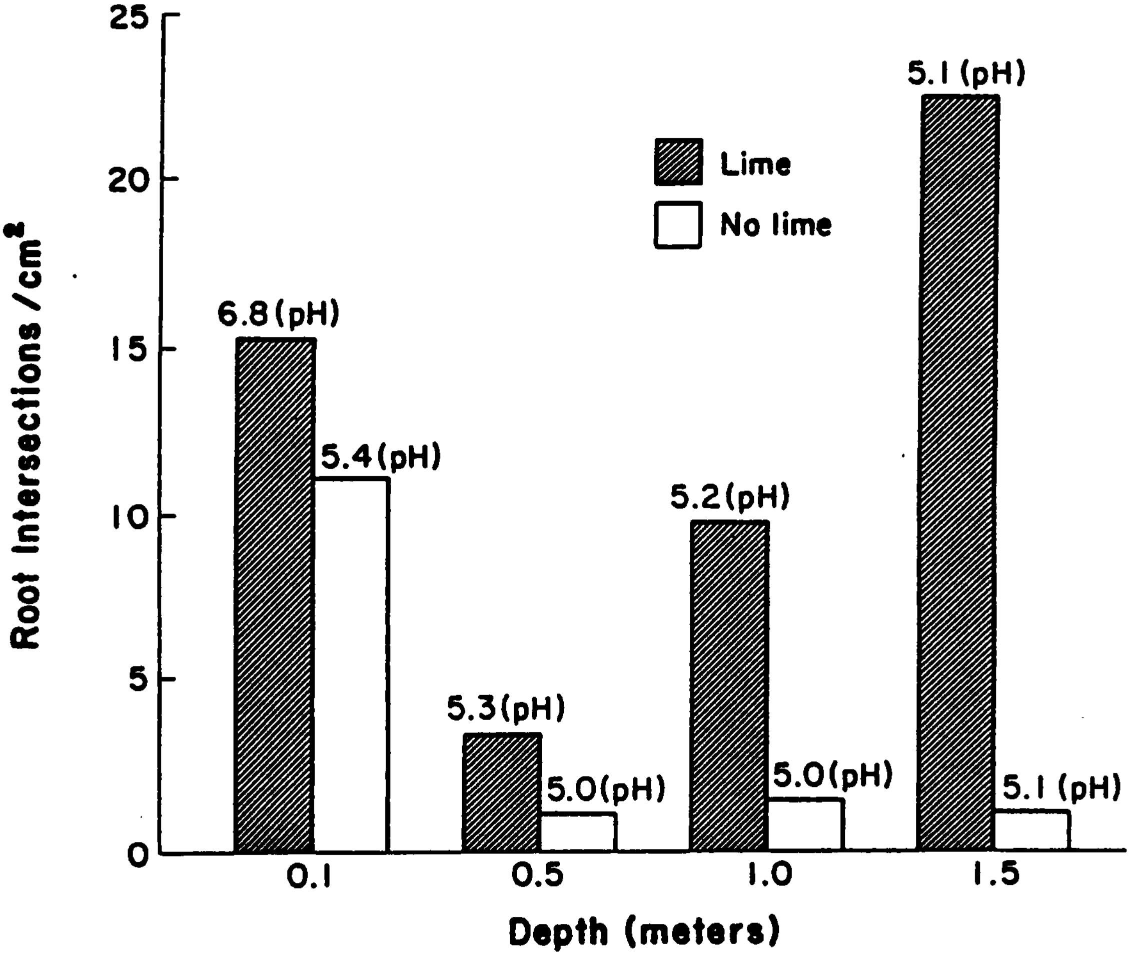
+ Not significant at the 0.05 level using Fisher's protected LSD test.

between limed and unlimed plots. Aluminum increased with increasing depth regardless of lime application. Time of lime application (6 or 18 months prior to planting, or at planting) did not affect Al concentration in the soil.

Calcium ranged from a low of 271 to a high of 870 mg kg^{-1} for unlimed and limed plots, respectively in the top 15 cm (Table 4). Time of lime application resulted in no changes in soil Ca levels.

5.4.2 Root Growth

Roots were more prevalent at the 1.0 and 1.5 m depths in surface limed plots in comparison to the unlimed plots (Fig. 9). Roots penetrated to the 1.5 m depth under acidic conditions. This is in agreement with earlier work conducted by Rechcigl and Reneau (1984), who showed alfalfa roots penetrated to the 152 cm depth in a Tatum clay loam soil (Typic Hapludult; clayey, mixed, thermic) in the presence of 225 mg kg^{-1} exchangeable Al when surface limed. This is contrary to the findings of other investigators who reported stunting of roots in nutrient solutions with as little as 1 mg kg^{-1} Al (Simpson et al., 1978). At the 0.5 m depth, roots were reduced in comparison to other depths regardless of lime application, due to the presence of a fragipan (Fig. 9). Roots which penetrated the fragipan branched at greater depths.



LSD (0.05) =	NS	NS	7.6	23.3
CV (%) =	26.4	44.4	9.0	11.5

Fig. 9 Influence of surface limestone (applied 8 months before planting) on alfalfa root abundance at various soil depths for experiment 2. Numbers above each bar indicate pH values at different depths.

Alfalfa roots from treatments with and without surface lime were tested by the Quinalizarine method to detect the presence of Al on the root surface. Whitish roots from unlimed soil changed to red-violet indicating the presence of Al at all depths. In contrast, surface roots (0 to 15 cm) from plots that received surface lime appeared blue under comparable conditions indicating absence of Al at the 0 to 15 cm depth. At depths greater than 15 cm, all roots appeared red-violet indicating the presence of Al.

5.4.3 Crop Yield

Alfalfa yields were increased by surface application of lime (Fig. 10 and Table 5). The 1981 planting of alfalfa showed a 2 to 3 fold yield increase from surface lime application compared to the unlimed plots. The time of lime application whether applied immediately prior to planting or 6 or 18 months before planting did not influence yields. In the 1982 alfalfa planting there were 3 and 2 fold yield increases in the 1983 and 1984 harvests, respectively, due to surface-lime application. Again, there were no differences in yield due to time of lime application. Rainfall after spring planting may have benefited lime response but different results might occur with late August planting. When rainfall after planting is limited, then

Fig. 10 Influence of surface limestone on alfalfa as compared to control (center).

Table 5. Influence of application date of surface-applied agricultural limestone on alfalfa yield and purity of stand.

Date of planting	Lime application		Year of alfalfa harvest			Year of alfalfa harvest		
	Rate	Date	1982	1983	1984	1982	1983	1984
	Mg ha^{-1}		---- Yield (Mg ha^{-1}) ----			-- Purity of stand (%)--		
Apr. 1981	0.0	--	2.0	4.9	5.0	33	25	51
	6.7	Apr. 1981	6.1	10.0	10.6	72	45	71
	6.7	Aug. 1980	6.8	11.3	10.3	76	46	74
LSD(0.05)			1.6	4.8	4.0	26	NS[+]	NS
CV(%)			25.2	24.6	20.3	19	26	18
Apr. 1982	0.0	--	--	2.6	4.3	--	8	47
	6.7	Apr. 1982	--	8.0	10.5	--	32	74
	6.7	Aug. 1980	--	7.6	8.9	--	31	67
LSD(0.05)			--	2.0	3.9	--	15	14
CV(%)			--	14.2	21.6	--	29	10

[+] Not significant at the 0.05 level using Fisher's Protected LSD test.

lime application 6 to 18 monthes before planting might be necessary.

Yield increases were related to increased root penetration. Increased root penetration may enable the plants to obtain needed nutrients and water. This is in agreement with the previous work of Rechcigl et al. (1985) who reported increased yield with increased root penetration as a result of plowsole lime application.

5.4.4 Tissue Composition

There were no differences in tissue content of Ca, P, Al and Mn between limed and unlimed soil (Table 6). There were trends toward increased Mn concentrations in the absence of surface lime, though not in the toxic range. Nitrogen concentration was increased by lime application. In the absence of lime, N averaged 29 g kg^{-1} while in the presence of lime, N averaged 32 g kg^{-1}. Soil acidic conditions may reduce N fixation and thus limit N available for root and top growth. This is in agreement with the findings of other investigators (Skerman, 1977; Keyser and Munns, 1979). A reduced level of N fixation in the unlimed soil may also account for decreased root penetration resulting in reduced yields.

Table 6. Influence of application date of surface-applied agricultural limestone on nutrient concentration and calculated N uptake of alfalfa herbage. Concentrations are averages of three hay harvests in 1984.

Date of planting	Lime application		Nutrient concentration					
	Rate	Date	N	Ca	P	Al	Mn	N uptake[‡]
	$Mg\ ha^{-1}$		$g\ kg^{-1}$			$mg\ kg^{-1}$		$kg\ ha^{-1}$
Apr. 1981	0.0	—	29	13	4.4	157	91	105
	6.7	Apr. 1981	33	12	4.3	131	68	246
	6.7	Aug. 1980	32	12	4.4	221	77	231
LSD(0.05)			2	NS[+]	NS	NS	NS	114
CV(%)			3	7	3.0	39	26	26
Apr. 1982	0.0	—	30	12	4.2	132	90	75
	6.7	Apr. 1982	32	12	4.2	147	61	227
	6.7	Aug. 1980	32	12	4.5	156	70	188
LSD(0.05)			NS	NS	NS	NS	NS	104
CV(%)			6	10	5.2	17	16	28

[+] Not significant at the 0.05 level using Fisher's Protected LSD test.

[‡] N Uptake = N concentration x yield.

5.5 CONCLUSION

The present study demonstrated that surface application of lime may be adequate for the growth of alfalfa in acidic soils, with the most important factor being N fixation generated in the upper portion of the soil profile.

5.6 LITERATURE CITED

Baylor, J. E. 1983. Agronomy guide. p. 56-58. College of Agriculture Ext. Serv. Pennsylvania State University.

Bohm, W. 1979. Methods of Studying Root Systems. Springer-Verlag Berlin Heidelberg.

Brann, D. E., E. S. Hagood, and J. E. Roberts. 1983. Successful no-till corn production. Va. Coop. Ext. Serv. Pub. 424-004.

Council, K. A. and J. P. Sall (ed.). 1982. SAS User's Guide. Statistical Analysis Systems Institute Inc., Cary, NC.

Donohue, S. J. and S. W. Gettier. 1979. Laboratory procedures, Virginia Polytechnic Institute and State University Soil Testing and Plant Analysis Laboratory. Virginia Polytechnic Institute and State University Pub. 853.

Jayman, T. C. Z. and S. Sivasubramaniam. 1974. The use of ascorbic acid to eliminate interference from iron in the aluminon method for determining aluminum in plant and soil extracts. Analyst. 99:296-301.

Kalavoulos, I. M. and N. D. Misopolinas. 1977. Aluminum qualitative determination on plant roots by quinalizarine. Aristotelian University of Thessaloniki, Scientific Annals of the School of Agriculture and Forestry. K:595-597.

Kauffman, M. 1977. The effect of soil acidity and lime placement on root growth and yield of winter wheat and alfalfa. Ph.D. Diss. Oregon State University.

Keyser, H. H. and D. N. Munns. 1979. Effects of calcium, manganese and aluminum on growth of rhizobia in acid media. Soil Sci. Soc. Am. J. 43:500-503.

Mueller, J. P. and D. S. Chamblee. 1984. Sod-seeding of ladino clover and alfalfa as influenced by seed placement, seeding date, and grass suppression. Agron. J. 76:284-289.

Rechcigl, J. E. and R. B. Reneau, Jr. 1984. Effect of subsurface acidity on alfalfa in a Tatum clay loam. Commun. Soil Sci. Plant Anal. 15: 811-818.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985. Effect of subsurface amendments and irrigation on alfalfa growth. Agron. J. 77: 72-75.

Simpson, J. R. and A. Pinkerton. 1978. Effects of subsoil acidity and liming on the root behavior and shoot growth of some leguminous plants. Inf. Ser-N. Z. Dep. Sci. Ind. Res. 134 (Plant Nutr., V. 2) p. 449-455.

Skerman, P. J. 1977. Tropical forage legumes. Food and Agriculture Organization of the United Nations. Rome. p. 103-120.

Sperow, C. B., Jr. 1983. No-till systems for West Virginia Agronomy Mimeo. 325. West Virginia University.

White, H. E., D. D. Wolf, H. T. Bryant, E. S. Hagood, J. M. Luna, and E. S. Smith. 1982. No-till establishment of alfalfa. Va. Coop. Ext. Serv. Pub. 418-263.

Chapter VI

THE INFLUENCE OF CA, N AND PH ON ALFALFA ROOT GROWTH AND NITROGEN FIXATION USING THE IMPLANTED SOIL MASS TECHNIQUE

6.1 ABSTRACT

Soil acidity is a major growth limiting factor responsible for stunting of roots and reduction of yield in alfalfa (*Medicago sativa* L.). In addition, N fixation of alfalfa is decreased on acid soils eliciting N deficiency of the legume. Because of the complexity and interrelationships of various environmental factors, it is difficult in field situations to study the cause-and-effect relationships and to determine the influence of subsurface amendments on root systems and N fixation.

The implanted soil mass (ISM) technique, as employed in this study, consisted of mixing a study soil with chemical treatments of interest, burying this mixture in mesh bags 15 cm below an existing alfalfa stand, removing the bags after 1 y and determining root mass and N fixation via acetylene reduction.

The mixture placed in the mesh bags consisted of Tatum clay loam (Typic Hapludult; clayey, mixed,thermic) soil with an initial pH of 4.4 and either Ca, N, Ca+N, KOH or $Ca(OH)_2$. Mesh bags with these mixtures were buried in an existing

alfalfa stand on an Aquic Fragiudult that had received either 0 or 6.7 Mg/ha of surface applied dolomitic limestone.

The ISM with $Ca(OH)_2$ contained the most root mass and was more than doubled in the presence of surface liming as compared to the ISM containing the unamended control. Surface liming increased the root mass in the mesh bags for only the $Ca(OH)_2$ and KOH subsurface treatments. These increases were 7.5 and 5 fold for the $Ca(OH)_2$ and KOH treatments, respectively. For the unlimed soil only the ISM containing $Ca(OH)_2$ had increased root mass.

With the exception of $Ca(OH)_2$, ISM treatments had no influence on N fixation. In the ISM containing $Ca(OH)_2$ there was a 4 fold increase in N fixation in the surface limed soil and a 20 fold increase in the unlimed soil.

These results demonstrate that increased pH, as well as the presence of Ca are required for optimal root growth and N fixation.

6.2 INTRODUCTION

Soil acidity has been shown to be a major factor limiting alfalfa (Medicago sativa L.) growth (Foy, 1984; Rechcigl et al., 1986a; Joost and Hoveland, 1986). In particular, soil acidity has been reported to cause root stunting, resulting in reduced moisture and nutrient uptake (Joost and Hoveland, 1986; Rechcigl et al., 1985; Adams and Moore, 1983). Some investigators have shown the stunting of roots to result from Al toxicity (Rechcigl et al., 1986b, Adams, 1984), or N deficiency due to reduced N fixation on acid soils (Munns, 1965). Others have attributed the syndrome to the lack of Ca (Adams and Moore, 1983; Clark, 1984).

There is a general consensus that the problem of acidity can be alleviated through liming (Adams, 1984). Investigators, however, still disagree on the proper time and placement of limestone. Some investigators recommend that lime be incorporated into the soil, while others have shown lime to increase yields when applied to the plow layer (Rechcigl et al., 1985). There is also evidence that surface lime application may be adequate for establishment of no-till alfalfa on acid soils (Rechcigl et al., 1986a; Koch and Estes, 1986).

Because of the complexity of the soil system, it is difficult to study the influence of subsurface amendments on root growth and N fixation in a field situation. This paper describes a practical method which helps to alleviate some of these difficulties. An "implanted soil mass (ISM) technique", as used by Lund et al. (1970) in herbicide studies, was adapted to study the influence of various amendments on root growth and N fixation for alfalfa grown in an acid soil. The technique described has an advantage in that many treatments can be studied in a small area with similar field environmental conditions. In addition, the ISM can be easily extracted with minimal destruction of nodules and roots.

6.3 MATERIALS AND METHODS

'Arc' alfalfa established in 1982 on surface limed and unlimed Ernest silt loam (fine-loamy, mixed, mesic Aquic Fragiudult) an acid soil located at the Agronomy Research Farm near Blacksburg, Virginia was used for this study (Rechcigl et al., 1986a). Broadcast application of 616 kg/ha/yr of 0-17-35-B fertilizer have been applied to both surface limed and unlimed soil since alfalfa was established.

The experiment was conducted as a randomized split plot design with surface applied lime serving as the main plots. Mixtures of Tatum clay loam (Typic Hapludult, clayey, mixed, thermic) soil and either Ca, N, Ca+N, $Ca(OH)_2$ or KOH served as the subplots. These mixtures were made by spraying 1200 g subsamples of Tatum clay loam soil with the above chemicals dissolved in water. The mixture was moistened, sealed in plastic bags, and allowed to equilibrate for two weeks. The mixtures were then air dried, sieved through a 2.2 cm hardware cloth, and placed into mesh bags made of loosely woven saran material sold as shade screen for the ISM technique used by Lund et al. (1970). The shade screen had 12 strands per 2.54 cm and was cut into 30.48 by 40.64 cm rectangles. Screens were then folded in half and the seams sealed with staples and plastic tape. Once the bags were filled with the mixtures, the openings were sealed. In July 1985 the mesh bags were randomly buried 15 cm below the soil surface in a hole excavated with a 10.2 cm bucket auger. The soil was firmed carefully around the mesh bags to ensure continuity between the soil inside and outside of the bags. The bags were then covered with soil and firmly packed in place. One year later, the mesh bags (ISM) were carefully removed and roots protruding from the sides of the bags discarded.

Acetylene reduction measurements were obtained by placing each ISM soil into a mylar bag. Air was replaced with purified acetylene to achieve a 10 percent acetylene environment. The gas inside the mylar bags was sampled in duplicate for ethylene production after one h of exposure. Ethylene samples were analyzed in duplicate using a Varian model 3700 gas chromatograph equipped with a flame ionization detector. The 1.5 m glass column had an internal diameter of 2 mm and was packed with poropak T (80-100 mesh). The carrier gas was N flowing at 30 mL/min. The injector, oven, and detector temperatures were maintained at 80, 70 and 150 C, respectively. Ethylene sample peak areas were compared against ethylene standards.

Roots present in the ISM were separated from the soil by washing the bag content through an 8 mesh sieve and oven dried at 65 C for 48 h prior to weighing.

Soil pH was determined on 1:1 soil to water slurries. Five g soil samples were extracted with 50 mL of 1 M KCl and shaken for 48 h and Al determined on an Inductively Coupled Plasma Spectrophotometer (ICP). In addition, Ca, K, Mg and P were determined on the ICP using the procedures reported by Donohue and Gettier (1979). Soil chemical analyses are given in Tables 7 and 8.

Statistical analyses consisted of analyses of variance (ANOVA) and Fisher's protected least significant differences test for mean separation as outlined by Steel and Torrie (1980). All statistical inferences were made at the 0.05 level of significance.

Table 7. Chemical composition of the implanted soil mass (Tatum clay loam soil + amendment) after 1 y of equilibrium in the field.

Soil Mass Amendment	Rate	pH	Nutrient K	Nutrient Ca	Nutrient Mg	Nutrient P
	Mg ha^{-1}		————	g kg^{-1}	————	
None	——	4.7	63	138	74	0.7
$Ca(OH)_2$	13.0	6.8	36	1191	55	1.0
$CaSO_4$	23.7	4.3	50	864	32	1.5
KOH	19.3	6.3	157	165	71	1.0
$CaSO_4$	23.7	4.1	50	918	39	1.5
+ N	0.2					
N	0.2	4.4	64	117	49	0.5
$LSD_{(0.05)}$		0.6	26	115	27	NS^{+}

$^{+}$Not significant at the 0.05 level using Fisher's protected LSD test.

Table 8. Influence of surface applied limestone on soil pH, exchangeable Al, Ca, and P in an Ernest silt loam soil three years after application.

Lime rate	pH		K		Ca		Mg		P		Al	
	Depth (cm)											
	0-5	45-50	0-5	45-50	0-5	45-50	0-5	45-50	0-5	45-50	0-5	45-50
Mg ha^{-1}			g kg^{-1}								mg kg^{-1}	
0.0	5.3	5.0	56.8	40	369	330	81	64	5.0	4.7	23	99
6.7	6.3	5.1	58.3	49	632	364	74	69	4.3	4.4	1	62
LSD$_{(0.05)}$	*				*						*	

---

[*] Significant at the 0.05 level using Fisher's protected LSD test.

6.4 RESULTS AND DISCUSSION

6.4.1 Root Weight

Root weights were dramatically increased when the ISM included $Ca(OH)_2$ in both the surface limed and unlimed soils when compared to the controls (Fig. 11). Addition of KOH to the ISM also increased root weights, but only in the presence of surface lime. The response to KOH in the ISM in surface limed soils was not as pronounced, however, as was inclusion of $Ca(OH)_2$ in the ISM. Of the various other ISM treatments, only with $CaSO_4$ was there a trend toward increased root mass.

As shown in Table 7, inclusion of $Ca(OH)_2$ in the ISM not only supplied Ca but increased the soil pH (from 4.7 to 6.8). Inclusion of gypsum in the ISM did not significantly alter the soil pH.

Surface lime application, compared to no lime application, increased ISM root weight by two fold with inclusion of $Ca(OH)_2$ and six fold with KOH, respectively (Fig. 11). The increased ISM root weight in limed vs. unlimed soil with KOH suggests that the plant is unable to respond to a favorable pH environment without adequate Ca supply. This is further substantiated by the failure of KOH in unlimed soil to increase ISM root weight as compared with the unlimed control.

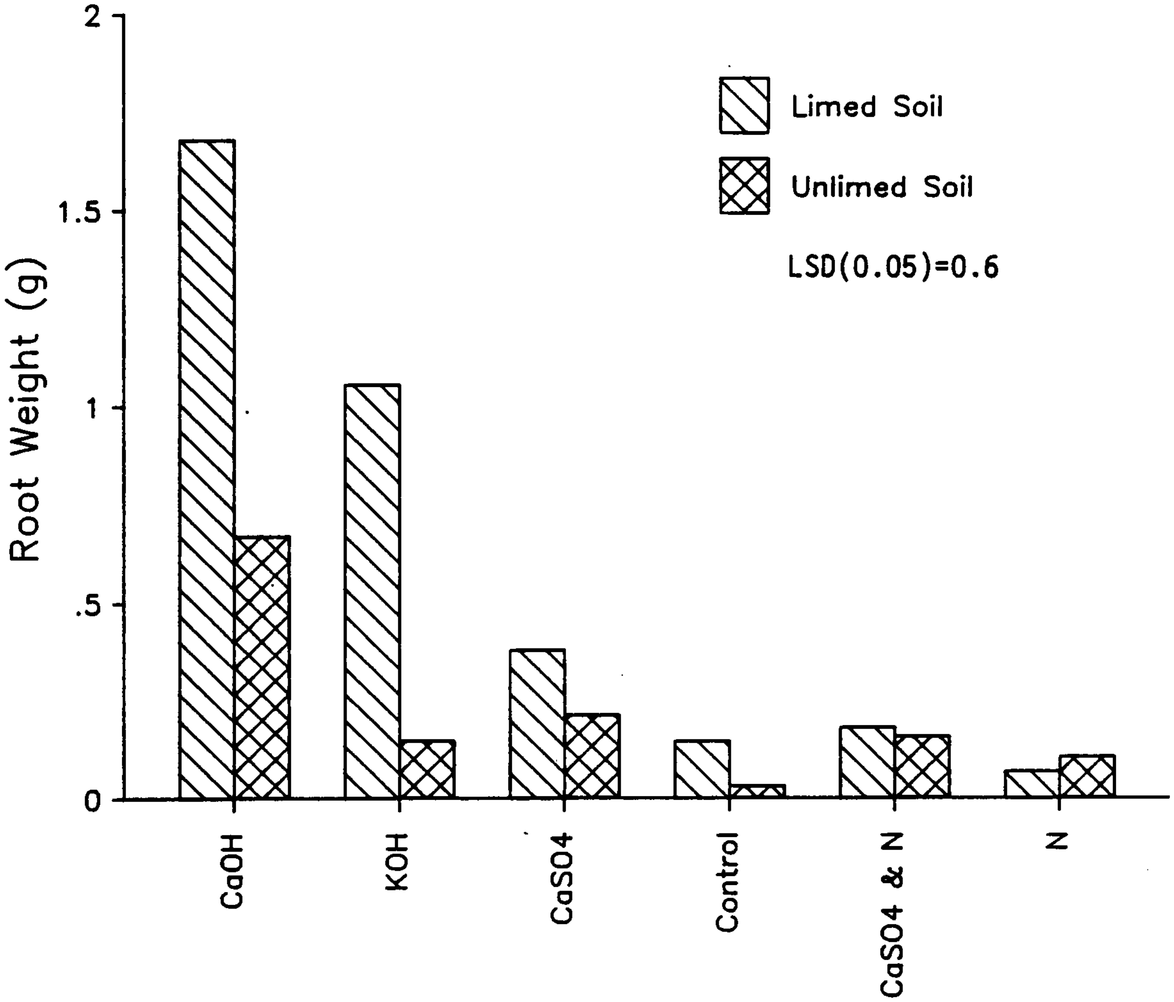


Fig. 11 Root mass in ISM as influenced by surface lime and various amendments.

These results suggest that both the reduction in acidity and the availability of Ca are prerequesites for maximum root growth. The failure of $CaSO_4$ to support maximum root growth is a clear indication that Ca alone is insufficient for this purpose. Similarly studies with the KOH treatment prove that increasing the alkalinity without the addition of Ca does not result in maximum root growth. In this respect it should be noted that some investigators have attributed the stunting of the roots and reduced yields on acid soils solely to Ca deficiency (Clark, 1984; Foy, 1974; Adams and Moore, 1983).

Alfalfa studies conducted previously at this site exhibited increased tissue N levels and forage yield on the limed vs. unlimed soils (Rechcigl et al., 1986a).

Alfalfa plants on the unlimed soil appeared to be N deficient judging from the yellowish green color of the foliage. Nitrogen deficiency in alfalfa on unlimed acid soil has been shown to result from reduced N fixation (Munns, 1965; Skerman, 1977). When this is considered in conjunction with the high pH requirement for rhizobia viability, N was included as a variable in the present work. The current study showed no increase in root weights with N or Ca+N treatment. Any benefits from N application may have been offset by the soil pH level of 4.4 and 4.1 respectively

(Table 7). This situation could have been further compounded by increased acidity due to enhanced nitrification.

6.4.2 N Fixation

Acetylene reduction was significantly increased in ISM treated with $Ca(OH)_2$ in both the limed and unlimed soil (Fig. 12). This is expected since a neutral pH and adequate Ca are required for viability of rhizobia and N fixation (Fred, 1932). In addition, the neutral pH enhances the availability of Mo, which is a component of nitrogenase, the enzyme required for N fixation.

The ISM treated with N did not influence N fixation. This was expected, since N fixation would not normally take place at the soil pH (4.4) used. Under a neutral pH environment, N fixation has been reduced in the presence of added inorganic N (Fred et al., 1932).

Of the various treatments added to the ISM only the $Ca(OH)_2$ affected N fixation. Contrary to what one might expect, there was significantly greater N fixation in $Ca(OH)_2$ treated ISM in unlimed soil than in soils treated with lime.

It is believed that the surface-limed soil provided a zone near the surface favorable for N fixation which used

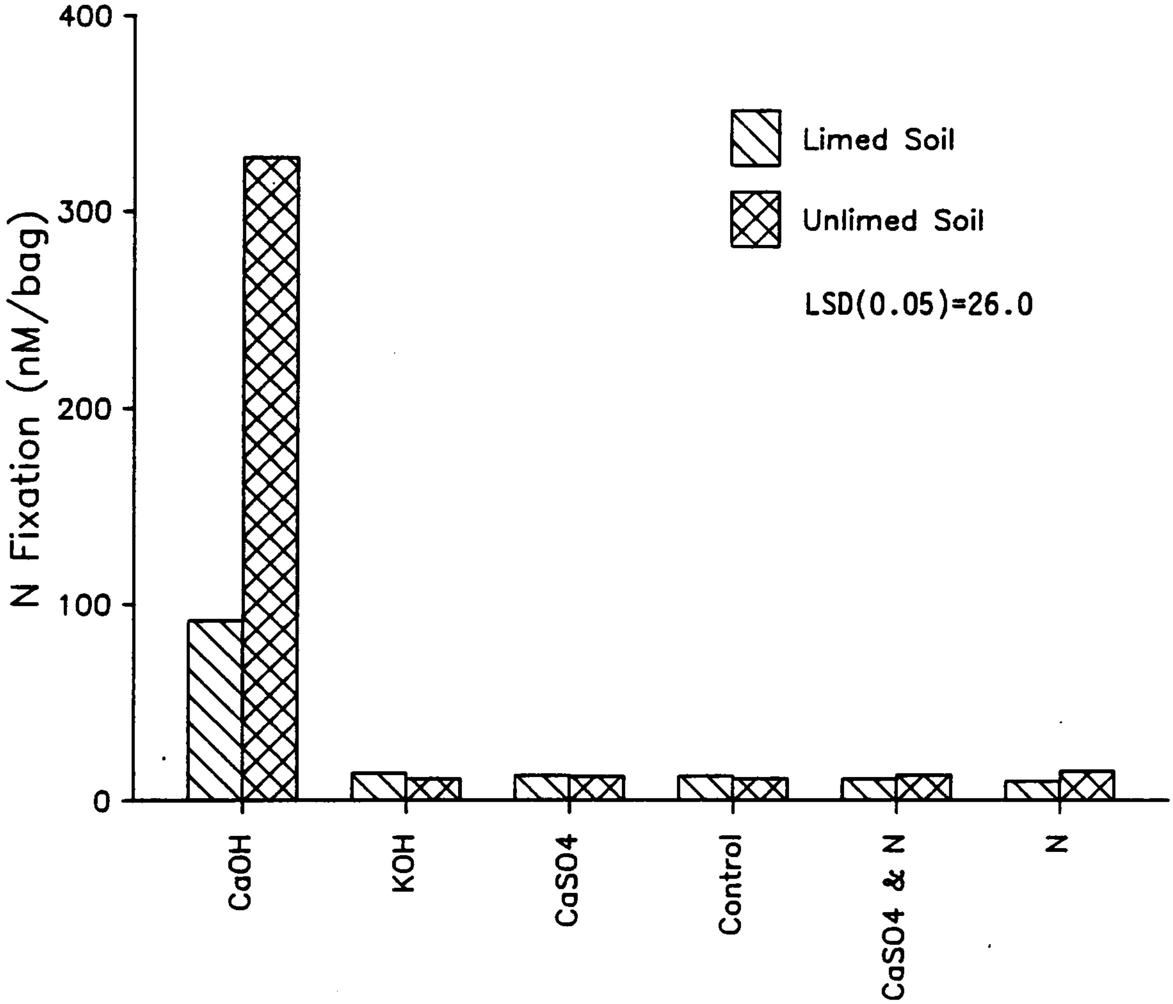


Fig. 12 Nitrogen fixation in ISM as influenced by surface lime and various amendments.

all the precursors necessary for this process (Havelka et al., 1982). As a consequence, only limited N fixation could occur in the $Ca(OH)_2$ treated ISM even though the conditions were otherwise favorable for nodule development.

Investigators have shown increased nodulation in different soil horizons in the presence of lime. One of the most striking results was reported by Pohlman (1946) on the influence of liming subsurface layers on nodulation of alfalfa. He demonstrated that the nodules concentrated in the 40.6-51 cm layer which was limed to neutrality below the acid surface soil layer (0-40 cm). In another experiment, nodules occurred in the 20.3-40.6 cm layer when lime was applied to that layer. A relationship between nodulation and lime application is in agreement with our research as well as the results of Bryan (1923) and Karraker (1927). In previous studies Rechcigl et al.(1985) reported increased N content in alfalfa in the presence of plowsole limestone as compared to the unlimed plowsole. This increase in tissue N was attributed to enhanced N fixation in the plowsole due to presence of plowsole lime.

Many investigators have shown N fixation to be enhanced in soils with a neutral pH and where adequate Ca is present (Adams, 1984). In the present study, increasing the pH without the addition of Ca or the addition of Ca, without increasing the pH, did not enhance N fixation. These results suggests that both adequate pH and Ca are necessary for N fixation.

6.5 LITERATURE CITED

Adams, F. and B. L. Moore. 1983. Chemical factors affecting root growth in subsoil horizons of coastal plain soils. Soil Sci. Soc. Am. J. 47:99-102.

Adams, F. 1984. Soil acidity and liming. A.S.A. Monograph Agronomy 12, 2nd Edition. Amer. Soc. Agron. Madison WI.

Bryan, O. C. 1923. Effect of acid soils on nodule forming bacteria. Soil Sci. 20:37-40.

Clark, R. B. 1984. Physiological aspects of calcium, magnesium, and molybdenum deficiencies in plants. p. 99-147. In F. Adams (ed.) Soil Acidity and Liming. A.S.A. Monograph Agronomy 12, 2nd Edition. Amer. Soc. Agron. Madison, WI.

Donohue, S. J. and S. W. Gettier. 1979. Laboratory procedures, Virginia Polytechnic Institute and State University Soil Testing and Plant Analysis Laboratory. Virginia Polytechnic Institute and State Univ. Pub. 853.

Foy, C. D. 1974. Effects of aluminum on plant growth. p. 601-642. In E. W. Carson (ed.) The plant root and its environment. Univ. Press of Virginia, Charlottesville, Va.

Foy, C. D. 1984. Physiological effects of hydrogen, aluminum, and manganese toxicities in acid soil. p. 57-86. In F. Adams (ed.) Soil acidity and liming. A.S.A. Monograph Agronomy 12, 2nd Edition. Amer. Soc. Agron. Madison, WI.

Fred, E. B., I. L. Baldwin, and E. McCoy. 1932. Root nodule bacteria and leguminous plants. Univ. of Wisconsin, Madison WI.

Havelka, U. D., M. G. Boyle, and R. W. F. Hardy. 1982. Biological nitrogen fixation. p. 365-422. In F. J. Stevenson (ed.) Nitrogen in Agricultural Soils. A.S.A. Monograph Agronomy 22, Amer. Soc. Agron. Madison, WI.

Joost, R. E. and C. S. Hoveland. 1986. Root development of Sericea Lespedeza and alfalfa in acid soils. Agron. J. 78:711-714.

Karraker, P. E. 1927. Production of nodule and different parts of the root systems of alfalfa plants growing in soils of different reaction. Soil Sci. 24:103-107.

Koch, D. W. and G. D. Estes. 1986. Liming rate and method in relation to forage establishment - Crop and soil chemical responses. Agron. J. 78:567-571.

Lund, Z. F., R. W. Pearson, and G. A. Buchanan. 1970. An improved soil mass technique to study herbicide effects on root growth. Weed Sci. 18:279-281.

Munns, D. N. 1965. Soil acidity and growth of a legume. I. Interactions of lime with nitrogen and phosphate on growth of Medicago sativa L. and Trifolium subterraneum L. Aust. J. Agric. Res. 16:733-741.

Pohlman, G. G. 1946. Effect of liming different soil layers on yield of alfalfa and on root development and nodulation. Soil Sci. 62:255-266.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985. Effect of subsurface amendments and irrigation on alfalfa growth. Agron. J. 77:72-75.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., and W. Kroontje. 1986a. Influence of surface liming on the yield and nutrient concentration of alfalfa established with no-tillage techniques. Agron. J. 77:956-958.

Rechcigl, J. E., R. B. Reneau, Jr., D. D. Wolf, W. Kroontje, and S. W. Van Scoyoc. 1986b. Alfalfa seedling growth as influenced by Al, Ca, and pH. Commun. Soil Sci. Plant Anal. 17:24-44.

Skerman, P. J. 1977. Tropical forage legumes. Food and Agriculture Organization of the United Nations, Rome.

Steel, G. D. and J. H. Torrie. 1980. Principles and procedures of statistics; A biometrical approach. McGraw-Hill, New York.

Chapter VII

RESPONSE OF ALFALFA GROWN ON ACID SOIL TO DIFFERENT CHEMICAL AMENDMENTS

7.1 ABSTRACT

Soil acidity is a major cause of reduced yields of alfalfa (Medicago sativa L.) in the southeast. The purpose of this study was to determine if alfalfa could be grown successfully on acid soils if sufficient Ca, N, Mo and surface lime is provided. 'Arc' alfalfa was planted in March 1985 in a split plot design on an Aquic Fragiudult. Treatments included surface and incorporated lime at either 6.5 or 13.0 Mg/ha, gypsum at 13.0 Mg/ha and foliar Mo applied at 560 g/ha. In addition each plot was split with half of the plot receiving 224 kg N/ha and half receiving no N. Surface application of lime increased yields of alfalfa for both 1985 and 1986. Yields increased in response to addition of either N or Mo, indicating the importance of Mo in establishing alfalfa nodulating system for N assimilation. Nitrogen increased yields on control plots but had little influence on alfalfa that received Mo or lime. Soil pH ranged from 6.7 to 4.8 and exchangeable Al from 2 to 21 mg/kg in the surface 5 cm of the limed and unlimed plots, respectively. This study indicates that

surface lime is adequate for growing alfalfa on acid soils and that reduced N-fixation appears to be a major limiting factor to growth of alfalfa on acid soils.

7.2 INTRODUCTION

Alfalfa (Medicago sativa L.), a perennial legume, is of major economic importance to the farmers in the U.S. The major obstacles in alfalfa production in the East are Al toxicity and Ca deficiency. Adams and Hatchcock (1986) have reported improved crop yields following the addition of Ca on acid soils, while other investigators have shown successful alfalfa performance in the presence of lime on such soils. The beneficial response to lime occurred regardless of whether lime was applied at the plowsole (Rechcigl et al., 1985) or at the surface (Rechcigl et al., 1986a; Joost and Hoyt, 1986). Foliar application of Mo, which is known to enhance N fixation, may increase alfalfa yields on acid soils (Doerge et al., 1985; Gupta, 1969).

Because of the complexity of factors involved, it is difficult to reach definitive conclusions about the cause-effect relationships unless the various factors are studied concurrently. The current investigation was therefore undertaken to determine the comparative response of alfalfa on an acid soil following the application of foliar Mo, surface and incorporated limestone, gypsum and/or N application.

7.3 MATERIALS AND METHODS

'Arc' alfalfa was planted at a rate of 22.4 kg/ha in an Ernest silt loam soil (fine-loamy, mixed, mesic, Aquic Fragiudult) at the Agronomy Research Farm near Blacksburg, Virginia in March of 1985. The experimental design was a split plot design replicated four times.

Main plot treatments included surface and incorporated limestone at either 6.5 or 13.0 Mg/ha, gypsum at 13.0 Mg/ha and foliar Mo applied at 560 g/ha. The sub-plot treatment was either 0 or 224 kg N/ha. All plots received a broadcast application of 616 kg/ha/yr of 0-17-35-B fertilizer.

7.3.1 Sample Collection and Analysis

Alfalfa was harvested three times annually during the 1985 and 1986 growing seasons, using a sickle bar mower. Plant samples from two harvests in 1986 were air dried at 65 C for 24 h and ground in a stainless steel mill to pass a 0.84 mm sieve. Nitrogen, Ca, P, Al and Mn were determined as described by Donohue and Gettier (1979).

Soil samples were collected in June, 1985 from depth increments of 0 to 2.5, 2.5 to 5, 5 to 10 and 10 to 15 cm.

All soil samples were air dried and crushed to pass a 2-mm sieve. Soil pH, Ca and Mn measurements were carried out by the method of Donohue and Gettier (1979). For Al determination, 5 g of soil were shaken with 50 mL of M KCl for 48 h and the solution analyzed on the Inductively Coupled Plasma Spectrophotometer (ICP).

7.3.2 Statistics

Statistical analysis consisted of analysis of variance (ANOVA) and Fisher's protected least significant difference test for mean separation, as outlined by Steel and Torrie (1980). All statistical inferences were made at the 0.05 level of significance.

7.4 RESULTS AND DISCUSSION

7.4.1 Soil Factors

Surface applied and incorporated limestone increased the soil pH in the upper part of the soil profile (Table 9). Where limestone was incorporated there was an increase in soil pH in the upper 10 cm and an increase in soil pH with increased lime application. Surface lime application increased the pH of the 0 to 2.5 cm layer and was not influenced by rate of lime application. There was a trend toward increased soil pH with surface lime application in

Table 9. Influence of dolomitic lime, gypsum, and foliar applied Mo on pH, exchangeable Al, Ca, and Mn in an Ernest silt loam soil.

			Depth (cm)			
Amendment	Placement	Rate	0-2.5	2.5-5	5-10	10-15
		Mg ha^{-1}	pH			
None	—	—	5.8	5.2	5.0	5.1
Lime	Incorporated	6.5	6.3	5.9	5.5	5.2
		13.0	6.7	6.5	5.5	5.0
	Surface	6.5	6.7	5.7	5.2	5.2
		13.0	6.5	5.5	5.3	5.4
Gypsum	Incorporated	13.0	5.6	4.9	4.7	4.8
	Surface	13.0	5.1	4.8	4.6	4.7
Mo	Foliar	++	5.5	5.3	5.2	5.2
LSD$_{(0.05)}$			0.3	0.4	0.3	0.3
			Al (mg kg^{-1})			
None	—	—	15.7	21.4	32.3	33.9
Lime	Incorporated	6.5	0.9	1.4	3.0	18.0
		13.0	0.6	2.3	2.1	18.0
	Surface	6.5	1.0	3.1	19.2	50.7
		13.0	1.4	2.9	10.7	11.7
Gypsum	Incorporated	13.0	14.3	24.4	28.7	32.1
	Surface	13.0	20.4	17.0	41.0	35.5
Mo	Foliar	++	24.7	31.7	41.0	32.9
LSD$_{(0.05)}$			15.4	16.5	18.3	$^{+}$NS
			Ca (g kg^{-1})			
None	—	—	368	268	304	362
Lime	Incorporated	6.5	500	388	390	364
		13.0	764	692	396	342
	Surface	6.5	712	372	330	338
		13.0	888	348	360	384
Gypsum	Incorporated	13.0	480	488	666	482
	Surface	13.0	1024	420	416	432
Mo	Foliar	++	372	332	326	360
LSD$_{(0.05)}$			272	124	102	78
			Mn (mg kg^{-1})			
None	—	—	14.5	14.1	11.2	10.5
Lime	Incorporated	6.5	15.2	12.0	9.2	10.2
		13.0	14.0	11.2	9.0	10.0
	Surface	6.5	16.1	13.4	9.5	8.7
		13.0	15.5	11.9	8.5	8.5
Gypsum	Incorporated	13.0	15.0	12.3	10.7	10.2
	Surface	13.0	15.4	14.4	11.8	12.0
Mo	Foliar	++	15.4	12.9	11.2	10.8
LSD$_{(0.05)}$			NS	NS	NS	NS

[+] Not significant at the 0.05 level using Fisher's protected LSD test.

[++] Mo foliar applied at 560 g ha^{-1}.

the upper 10 cm. The pH tended to decrease where gypsum was applied. This decrease in pH may be the result of hydrolysis of exchangeable Al (Adams, 1984).

Soil exchangeable Al levels in the surface 10 cm ranged from a low of 0.6 to a high of 41 mg/kg in the limed vs. unlimed soils, respectively. Highest levels of exchangeable Al were present in samples from plots which received gypsum. This is consistent with the lower soil pH values for these treatments. Lime application reduced Al levels by 20 fold in the surface 10 cm and is in agreement with the work of Coleman and Thomas (1967).

Calcium levels in the surface 10 cm were substantially higher on plots which received either limestone or gypsum than control plots. At the 2.5-5 cm depths, Ca levels were higher in plots receiving incorporated lime or gypsum than in plots receiving amendments at the surface.

There were no significant differences in Mn levels with respect to different treatments regardless of depth. The average level of Mn was 15 mg/kg in the surface 15 cm.

7.4.2 Crop Yields

Alfalfa yields were increased as a result of surface applied and incorporated lime in both 1985 and 1986 (Table 10). This is in agreement with an earlier study which

Table 10. Influence of dolomitic lime, gypsum, foliar applied Mo and N on alfalfa yields in 1985 and 1986 grown on an Ernest silt loam soil.

			N (kg ha^{-1})			N (kg ha^{-1})		
Amendment	Placement	Rate	0	224	Mean	0	224	Mean
			Mg ha^{-1}					
			1985 Yield[+]			1986 Yield[+]		
None	---	---	5.9	7.6	6.8	3.6	5.0	4.3
Lime	Incorporated	6.5	7.9	9.4	8.7	4.9	5.0	5.0
		13.0	8.0	9.5	8.8	5.9	6.0	6.0
	Surface	13.0	7.7	8.8	8.2	5.5	6.3	5.9
		6.5	7.4	8.5	8.0	5.8	6.0	5.9
Gypsum	Incorporated	13.0	6.3	8.4	7.4	5.0	5.7	5.4
	Surface	13.0	7.6	8.0	7.8	4.5	5.4	5.0
Mo	Foliar	[++]	7.2	8.1	7.7	4.9	5.1	5.0
Mean			7.2[*]	8.5		5.0[*]	5.6	
$LSD_{(0.05)}$					0.9			0.7

[*] Means are significantly different at the 0.05 level using Fisher's protected LSD test.

[+] Alfalfa yield is a summation of 3 cuttings.

[++] Mo foliar applied at 560 g ha^{-1}.

showed successful performance of alfalfa on acid soil when surface-limed (Rechcigl et al., 1986a). This observation is of significance to producers since planting no-till alfalfa has not been recommended on acid soils unless the lime was incorporated a year prior to planting (White et al., 1982).

There were no differences in yields when incorporated lime was half the recommended rate in 1985, however in 1986 yields were reduced where half the recommended rate of incorporated lime was applied. Alfalfa yields in 1985, as well as in 1986, were not reduced significantly when surface lime application was half of the recommended rate, suggesting that the surface lime recommendations for the state of Virginia could be lowered (Rechcigl et al., 1986a).

In 1986 where lime was incorporated at half the recommended rate yields were reduced by 1 Mg ha^{-1}. Alfalfa yields also increased in response to N regardless of other treatments.

Δ Foliar application of Mo increased yields of alfalfa on unlimed soil indicating the importance of Mo in establishing alfalfa nodulating system for N assimilation (Doerge et al. 1985). Molybdenum may be unavailable for plant use under acidic soil conditions (Adams 1984). Thus, foliar application of Mo may increase yields under such conditions. This has been substantiated in the present study by comparing the yield response due to N application for the control and the Mo treatment in 1985.

Eventhough the interaction between N and other treatments was not significant, there were several interesting trends. Nitrogen increased yields on soils that received no other amendments but had little influence on alfalfa that received Mo. Yield increases, resulting from N application, were higher in 1985 than in 1986. This may be because lime had more time to react to less favorable environmental conditions.

Gypsum produced yield increases when applied to the surface in the absence of N, however, it was without any apparent effect when N was present, in both years of the study. This may imply the importance of N in the N fixation process. Surface applied gypsum seemed to be more effective then gypsum incorporated into the soil.

The present study demonstrates the importance of Ca for optimal alfalfa production grown on acid soils. This is substantiated by previous work (Rechcigl et al., 1986b). It is, however, apparent that Ca is not the only yield-limiting factor. Besides Ca, Mo and an increased pH were clearly indicated as prerequisites to assure optimal yields.

7.4.3 Tissue Analyses

None of the treatments changed the tissue P and K concentration in the first two harvests (Tables 11 and 12). Calcium concentration tended to be higher in alfalfa that received gypsum as compared to all other treatments. This is expected since the gypsum amendment provided the largest addition of Ca. Lime did not significantly increase the levels of Ca in the tissue. Magnesium levels in the tissue were increased where dolomitic limestone was applied. Magnesium tissue levels were in the sufficiency ranges regardless of treatments.

Tissue N was higher on plots receiving supplementary N as compared to those not receiving N. For treatments that did not receive N, tissue N content was increased only in the first cutting on the high lime and foliar Mo treatments. Increased N levels in the tissues may be a reflection of increased N fixation with these treatments.

Tissue Mn levels were reduced in alfalfa grown on plots receiving either lime or gypsum application when compared to the controls. The reduction in Mn content in alfalfa following liming has been observed by other investigators (Fried and Peech, 1946; Foy et al., 1974; MacLean et al., 1972). This reduction in Mn correlates well with the increased soil pH and larger alfalfa yields on these

Table 11. Influence of dolomitic lime, gypsum, foliar applied Mo and N on nutrient concentration of 1st cutting alfalfa in May 1986.

Nitrogen	Amendment	Placement	Rate	P	K	Ca	Mg	N	Mn	Al
				Nutrient						
kg ha^{-1}			Mg ha^{-1}	g kg^{-1}					mg kg^{-1}	
0	None	—	—	2.7	35.1	12.7	1.9	28.7	70.9	99.5
	Lime	Incorporated	6.5	2.6	35.8	11.4	2.0	28.0	37.6	52.8
			13.0	2.6	33.2	11.6	2.1	31.8	33.1	59.8
		Surface	6.5	2.4	34.1	12.8	2.2	27.2	40.8	86.2
			13.0	2.7	35.1	12.4	2.2	31.5	34.7	66.8
	Gypsum	Incorporated	13.0	2.6	32.7	12.8	1.5	28.3	49.7	107.7
		Surface	13.0	2.5	31.1	15.2	1.5	29.9	47.1	62.5
	Mo	Foliar	[++]	2.7	33.6	11.5	1.6	31.7	72.1	81.0
	LSD$_{(0.05)}$			NS[+]	NS	2.0	0.2	3.3	28.2	NS
224	None	—	—	2.4	30.9	12.2	1.6	29.4	62.6	72.0
	Lime	Incorporated	6.5	2.5	30.8	11.8	1.9	28.9	40.6	51.2
			13.0	2.5	32.0	11.4	2.1	32.1	33.4	59.9
		Surface	6.5	2.4	31.4	12.1	2.0	30.0	38.7	59.6
			13.0	2.4	31.5	11.8	2.0	31.3	35.6	51.0
	Gypsum	Incorporated	13.0	2.5	32.0	13.2	1.4	32.7	52.0	77.6
		Surface	13.0	2.3	28.4	14.6	1.4	31.9	58.8	90.3
	Mo	Foliar	[++]	2.4	31.6	11.9	1.5	31.3	52.9	78.6
	LSD$_{(0.05)}$			NS	NS	1.8	0.2	NS	12.0	NS

[+] Not significant at the 0.05 level using Fisher's protected LSD test.

[++] Mo foliar applied at 560 g ha^{-1}.

Table 12. Influence of dolomitic lime, gypsum, foliar applied Mo and N on nutrient concentration of 2nd cutting of alfalfa in June 1986.

				Nutrient						
Nitrogen	Amendment	Placement	Rate	P	K	Ca	Mg	N	Mn	Al
kg ha^{-1}			Mg ha^{-1}	g kg^{-1}					mg kg^{-1}	
0	None	---	---	2.3	28.2	8.7	1.4	24.2	51.9	35.1
	Lime	Incorporated	6.5	2.4	28.4	7.6	1.5	24.3	35.6	23.4
			13.0	2.5	29.5	7.4	1.5	23.6	29.4	26.4
		Surface	6.5	2.2	28.5	8.2	1.5	24.0	35.8	30.0
			13.0	2.4	28.7	8.1	1.5	25.5	33.8	34.0
	Gypsum	Incorporated	13.0	2.3	29.5	8.3	1.2	24.6	42.2	29.5
		Surface	13.0	2.3	28.4	9.3	1.1	24.8	38.4	25.6
	Mo	Foliar	++	2.1	28.6	8.0	1.2	24.9	41.1	33.4
	LSD$_{(0.05)}$			0.2	+NS	NS	0.2	NS	9.2	NS
224	None	---	---	2.1	28.0	7.8	1.2	23.0	49.1	30.3
	Lime	Incorporated	6.5	2.4	28.4	8.4	1.6	24.2	36.8	33.9
			13.0	2.4	28.2	8.5	1.6	25.1	30.6	28.0
		Surface	6.5	2.4	29.7	8.8	1.6	24.5	34.1	31.7
			13.0	2.3	28.3	8.4	1.6	26.3	33.5	38.1
	Gypsum	Incorporated	13.0	2.2	28.8	8.9	1.2	25.4	46.4	29.5
		Surface	13.0	2.1	27.6	9.4	1.2	23.9	44.2	34.4
	Mo	Folair	++	2.2	30.0	8.6	1.3	27.0	46.1	27.3
	LSD$_{(0.05)}$			NS	NS	NS	0.3	NS	8.6	NS

+Not significant at the 0.05 level using Fisher's protected LSD test.

++Mo foliar applied at 560 g ha^{-1}.

treatments. In this connection, it may be noted that Mn toxicity is frequently involved when legumes are grown on acid soils.

There were no differences in Al levels in the tissue with respect to the various treatments. This is in agreement with the work of many investigators (Adams, 1984; Rechcigl et al., 1986a,b).

7.5 LITERATURE CITED

Adams, F. 1984. Soil acidity and liming. A.S.A. Monograph Agronomy 12, 2nd Edition. Amer. Soc. Agron., Madison WI.

Adams, F. and P. J. Hatchcock. 1986. Aluminum toxicity and calcium deficiency in acid subsoil horizons of two coastal plain soil series. Soil Sci. Soc. Am. 48:1305-1309.

Coleman, N. T. and G. W. Thomas. 1967. The basic chemistry of soil acidity. p. 1-34. In R.W. Pearson and F. Adams (ed.) Soil Acidity and Liming. A.S.A. No. 12. Amer. Soc. Agron., Madison WI.

Donohue, S. J. and S. W. Gettier. 1979. Laboratory procedures, Virginia Polytechnic Institute and State University Soil Testing and Plant Analysis Laboratory. Virginia Polytech. Inst. and St. Univ. Pub. 853.

Doerge, T. A., P. J. Bottomley, and E. H. Gardner. 1985. Molybdenum limitations to alfalfa growth and nitrogen content on a moderately acid, high phosphorus soil. Agron. J. 77:895-901.

Gupta, U. 1969. Effect and interaction of molybdenum and limestone on growth and molybdenum content of cauliflower, alfalfa, and bromegrass on acid soils. Soil Sci. Soc. Amer. Proc. 33:929-932.

Foy, C. D., R. G. Orellana, J. W. Schwartz, and A. L. Fleming. 1974. Response of sunflower genotypes to aluminum in acid soil and nutrient solution. Agron. J. 66:293-96.

Fried, M. and M. Peech. 1946. The Comparative effects of lime and gypsum upon plants grown on acid soils. J. Am. Soc. Agron. 38:614-623.

Joost, R. E. and C. S. Hoveland. 1986. Root development of Sericea Lespedeza and alfalfa in acid soils. Agron. J. 78:711-714.

MacLean, A. J., R. I. Halstead, and B. I. Finn. 1972. Effects of lime on extractable aluminum and other soil properties and on barley and alfalfa grown in pot tests. Can. J. Soil Sci. 52:427-438.

Rechcigl, J. E., R. B. Reneau, Jr., and D. E. Starner. 1985. Effect of subsurface amendments and irrigation on alfalfa growth. Agron. J. 77:72-75.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., and W. Kroontje. 1986a. Influence of surface liming on the yield and nutrient concentration of alfalfa established with no-tillage techniques. Agron. J. 77:956-958.

Rechcigl, J. E., R. B. Reneau, Jr., D. D. Wolf, W. Kroontje, and S. W. Van Scoyoc. 1986b. Alfalfa seedling growth as influenced by Al, Ca, and pH. Commun. Soil Sci. Plant Anal. 17:24-44.

Steel, G. D., and J. H. Torrie. 1980. Principles and procedures of statistics: A biometrical approach. McGraw-Hill, New York.

White, H. E., D. D. Wolf, H. T. Bryant, E. S. Hagood, J. M. Luna, and E. S. Smith. 1982. No-till establishment of alfalfa. Virginia Coop. Ext. Serv. Pub. 418-263.

Chapter VIII

SOIL SOLUTION AL AS A MEASURE OF AL TOXICITY IN ALFALFA ON ACID SOILS

8.1 ABSTRACT

Alfalfa (Medicago sativa L.), a legume known for extreme sensitivity to Al toxicity, has recently been reported to grow successfully on acid soils high in exchangeable Al. In order to more fully understand this seeming contradiction, the present study was undertaken. It was hypothesized that most of the exchangeable Al may be insoluble or may be present at low activity levels and consequently not toxic to the plant.

To test this hypothesis, concentrations and activities of Al, Ca, pH and other major macro and micro elements were determined in Ernest silt loam (fine-loamy, mixed, mesic Aquic Fragiudult) soil solution and compared with soil exchangeable levels. Soil samples were taken from the surface (0-5 cm) and 1 m depth in soils that had received surface lime, gypsum and no amendments.

Aluminum activities were as low as 0 and 0.05 uM in soil solution from the surface and 1 m depth, respectively, for surface limed soils. Calcium activities in the soil solution from the surface 5 cm ranged from a high of 4.5 to

a low of 0.67 mM in soils which received gypsum or no amendment, respectively. Calcium activities in soil solution from the subsurface (1 m) ranged from 0.69 to 1.2 mM for the unlimed and limed soils respectively.

Despite the high exchangeable Al present in the Ernest silt loam soil, relatively low Al activities were present in soil solution, which may explain why alfalfa does not show characteristic Al toxicities when grown on these soils.

8.2 INTRODUCTION

For the past century, soil acidity has been a major obstacle faced by farmers growing alfalfa (Medicago sativa L.). In spite of the enormous research effort exerted in this area, the problem remains with us to date. Aluminum is assumed responsible for much of the adverse effects observed with acid soils.

Aluminum toxicity elicits a dramatic response in alfalfa readily detectable by the stunted roots (Simpson and Pinkerton, 1978; Foy, 1984). In nutrient solutions, the effect of Al toxicity on alfalfa is very pronounced, having been reported at a concentration of 1 mg/L (Rechcigl et al., 1986b). Other investigators have also shown the occurance of root stunting in red clover (Trifolium pratense L.) in nutrient solution in the presence of 0.027 mg/L Al (Kinraide et al., 1985).

The direct correlation between exchangeable Al and poor crop performance not withstanding, there have been recent reports indicating that alfalfa could be grown successfully at least on some acid soils containing high levels of exchangeable Al which have received surface lime. Excellent performance of alfalfa has been demonstrated under such conditions, even though the subsoil had a pH of 4.5 and exchangeable Al levels of 300 mg/kg (Rechcigl et al., 1986a). Roots in this soil were shown to penetrate to a depth of 1.5 m. The purpose of this study has been to investigate this seeming contradiction. Despite the high levels of exchangeable soil Al reported in the earlier work (Rechcigl et al., 1986a), it was felt plausible that the activity of Al in soil solution was relatively low, which could account for the lack of Al toxicity. In addition, the form of Al in soil solution may be important in determing if Al is toxic to the plant. Some investigators report that Al is most likely present in soil solution in different chelated forms and that that these forms of Al may not be toxic to plants (Bartlett and Riego, 1972; Adams and Moore, 1983).

Soil solution Al has been suspected to cause poor growth of crops as early as 1920 (McGeorge, 1924). Magistad (1952) has been given credit for being the first to show soil solution Al to be toxic to plants at both high and low pH. Adams and Lund (1966) showed Al activity values of 2 mM caused stunting of cotton roots grown in nutrient solution. This was substantiated by the work of Richburg and Adams (1970).

Calcium deficiency is known to result in reduced crop growth (Foy, 1974); however few studies have addressed the question of the Ca concentration in soil solution required for optimal alfalfa growth on acid soils. Many Ca deficiencies are actually a result of poor transport of Ca (Foy, 1984). Rios and Pearson (1964) have demonstrated that downward transport of Ca in cotton roots was inadequate to support root growth in the Ca deficient nutrient solution portion of a split medium. Lack of Ca transport down the root though the phloem has been substantiated by other investigators (Foy, 1974). Thus there is a need to determine whether adequate Ca is present in acid subsoil solution for crop growth and if not whether surface lime will increase subsoil Ca levels for adequate crop performance.

8.3 MATERIALS AND METHODS

In June, 1985, three replicate soil samples were collected at the surface (0-5 cm) and 1 m depths from soils that had received either 13.0 Mg/ha surface application of lime, gypsum or no amendment. A measured amount of distilled water was applied to bring these soils to 'field capacity', defined as the amount of water the soil sample would hold after equilibration over night. Soil samples were allowed to equilabrate for 3 months in a polypropylene bag. Subsamples (200 g) were centrifuged, using Butner funnels at 1000 g for 1 h to displace soil solution (Adams et al., 1980). Solutions were immediately analyzed using an ICP for cation and an anion chromatograph for anion concentrations (Donohue and Gettier, 1979) and ion activities calculated using the computer program GEOCHEM (Sposito and Mattigod, 1980). The pH of soil solutions was determined using an ion microprocessor equipped with pH electrodes.

8.4 RESULTS AND DISCUSSION

Average soil solution pH of the control treatments was 4.7 in the surface 0-5 cm (Table 13). The comparable pH value where limestone was applied was 7.3. In contrast to the stimulatory effect of lime, gypsum application actually

Table 13. Influence of surface applied limestone and gypsum at 13 Mg ha^{-1} on soil solution pH and ionic strength at different depths.

Depth	Amendment	pH	Ionic strength
cm			mM
0-5	None	4.73	3.70
	Lime	7.26	7.97
	Gypsum	4.26	19.90
100-105	None	4.83	3.00
	Lime	5.47	5.77
	Gypsum	4.50	3.04

lowered the pH by 0.4 units to 4.3 at the surface. At the 1 m depth, the control treatments had a pH of 4.8 while the gypsum and lime treatments had pH values of 5.5 and 4.5, respectively. It is evident that gypsum application lowers the soil pH due to the displacement of exchangeable Al into solution by Ca, thereby promoting hydrolysis and increasing acidity, as was demonstrated in earlier studies by Adams (1984).

Calcium concentrations were higher in the gypsum treatment than in the lime or control treatment (Fig. 13). The Ca concentration in the control soil solution was 50 mg/L as compared to 500 mg/L for the gypsum treatment at the 0-5 cm depth. At the 1 m depth, the average Ca concentrations for the control and the gypsum treatment were approximately 40 mg/L while those for the limestone treatment were about 80 mg/L (Fig. 14). From the comparison of the surface and the subsurface concentrations, it is apparent that Ca is not leached readily from the soil surface, though Ca concentrations in soil solutions were dramatically increased on surface limed soils at the 1 m depth. Concentrations of Ca in soil solution (40-80 mg/L) at the 1 m depth, were similar to the Ca concentrations used in nutrient solutions which were shown adequate for alfalfa growth (Rechcigl et al., 1986b). Magnesium concentrations

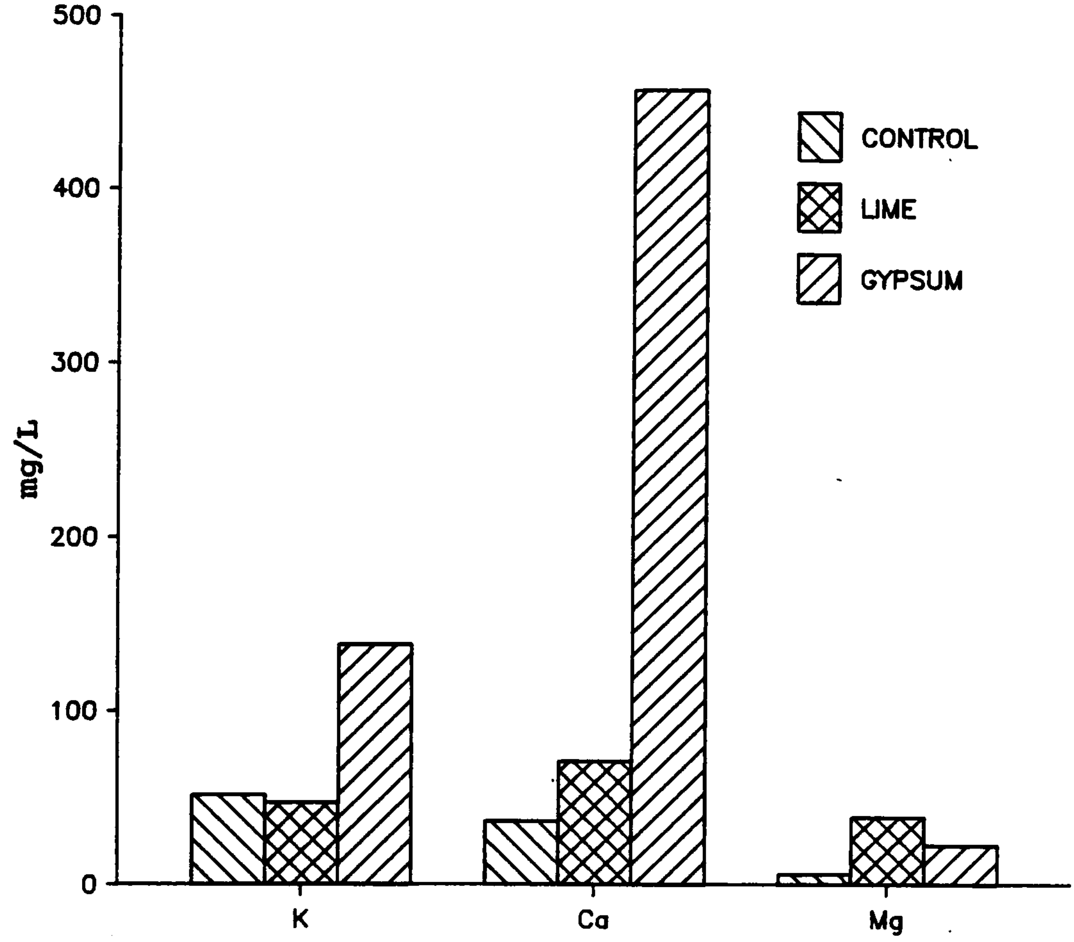


Fig. 13 Influence of lime and gypsum on soil solution K, Ca and Mg in the surface 5 cm of an Ernest silt loam soil.

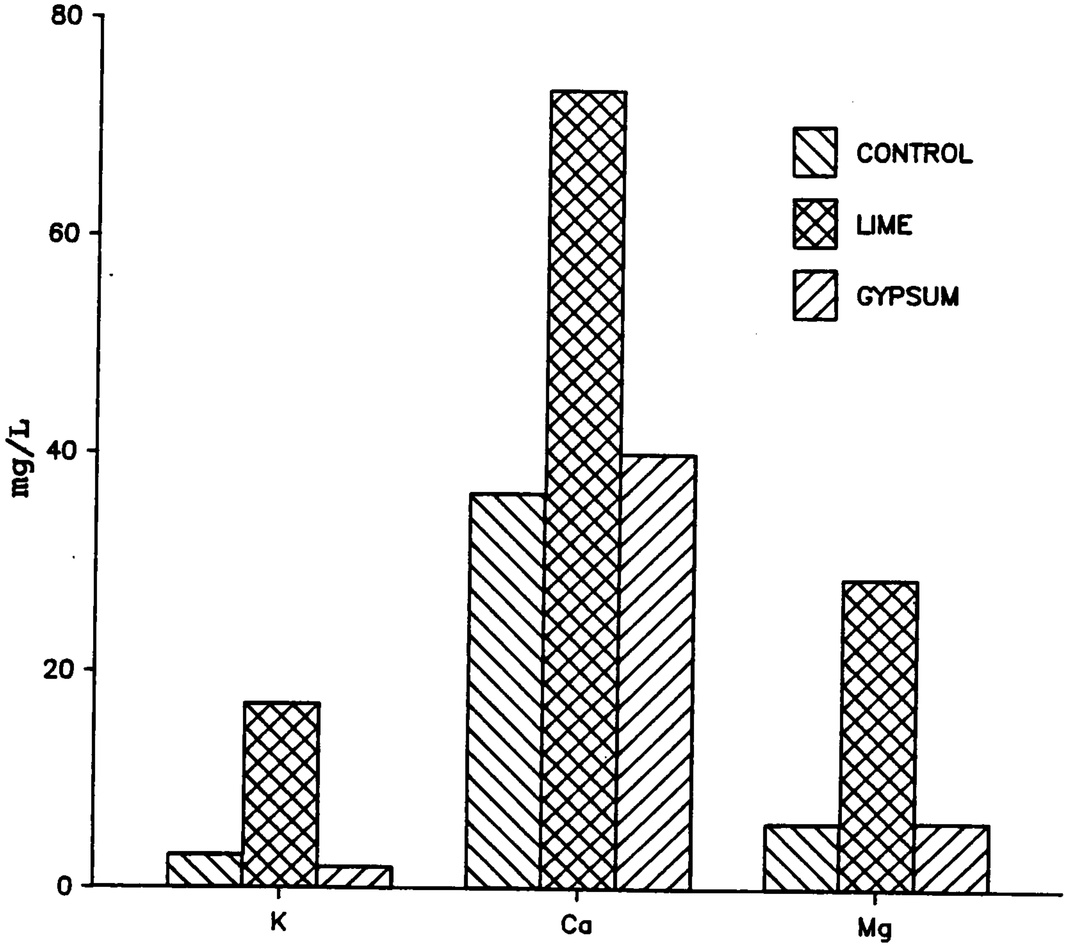


Fig. 14 Influence of lime and gypsum on soil solution K, Ca and Mg at the 1 m depth in an Ernest silt loam soil.

were highest in soils which received dolomitic lime at the surface and 1 m depth, i.e. 40 and 30 mg/L, respectively.

There were no differences in soil solution concentrations for Zn, Fe and Cu in the surface between treatments (Fig. 15). Manganese and B concentrations in the surface layer were lower on soils which received lime application in comparison to control or gypsum treatment. This probably results from increased pH in the presence of limestone, since many studies have demonstrated that the availability of these micronutrients decrease with increasing pH (Bohn et al., 1985).

Aluminum concentrations in the surface 5 cm of the soil solution ranged from a low of 0.3 mg/L in the control and lime treatments to a high of 2.7 mg/L in the gypsum treatment (Fig. 15). At the 1 m depth, Al concentrations were highest in the control soils with an average level of 0.13 mg/L (Fig. 16). This relatively low concentration of Al in solution may explain why the element did not adversely affect the growth of alfalfa on this soil, even though exchangeable Al concentrations were as high as 100 ug/g. The exchangeable Al is obviously a composite figure representing various ionic forms of the element, some of which may be less toxic or nontoxic to the plant. As a consequence, exchangeable Al may not be a good indicator of

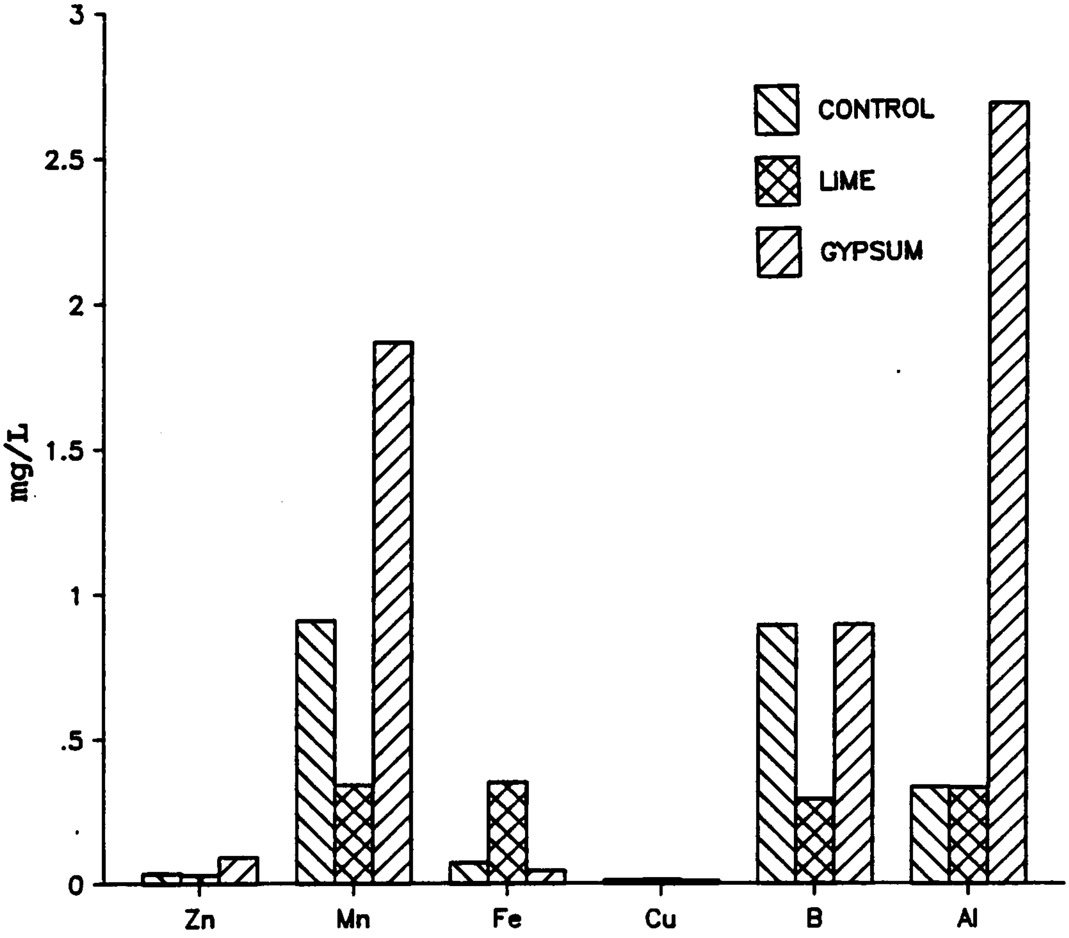


Fig. 15 Influence of lime and gypsum on soil solution micro-nutrients in the surface 5 cm of an Ernest silt loam soil.

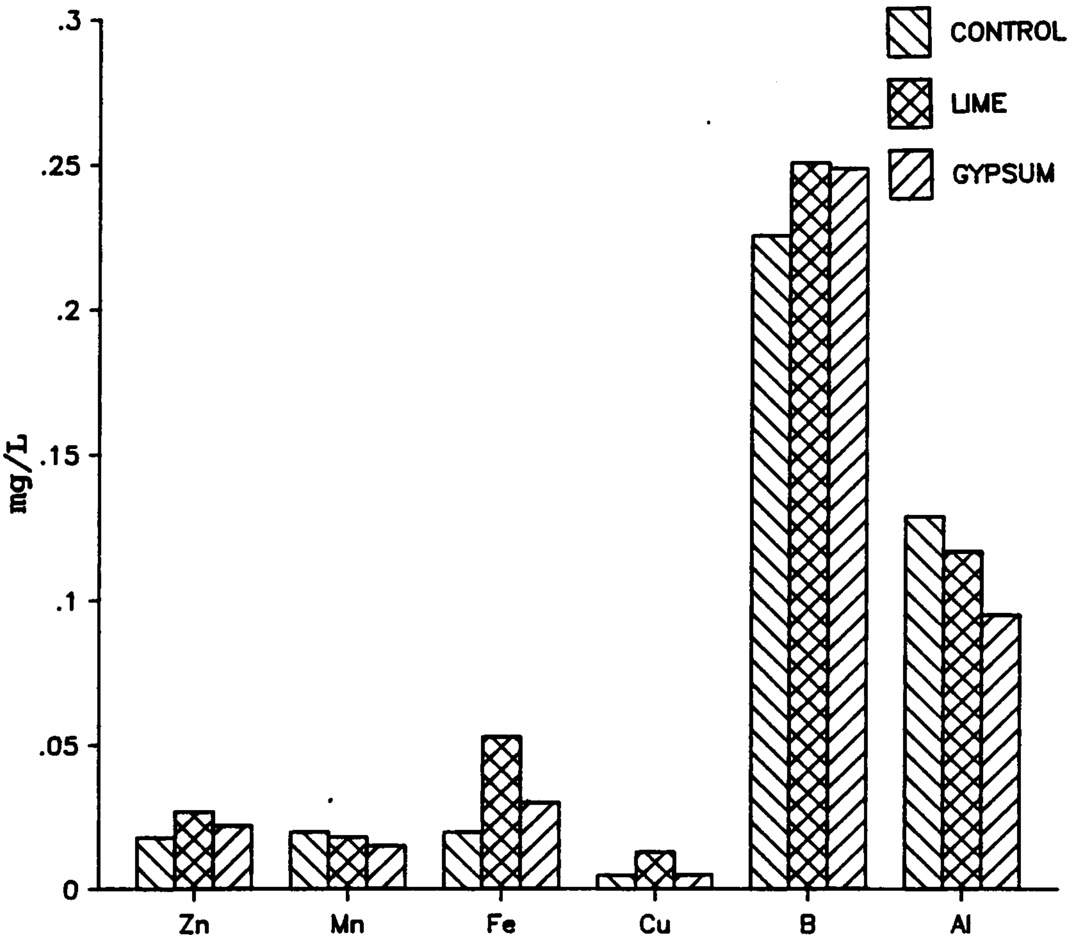


Fig. 16 Influence of lime and gypsum on soil solution micronutrients at the 1 m depth in an Ernest silt loam soil.

Al toxicity of alfalfa grown on acid soils and soil solution Al may be a better measure of this condition. It should be noted that in the nutrient solution studies (Rechcigl et al., 1986b) Al concentrations as low as 2 mg/L were toxic to alfalfa growth, while Al concentration in soil solution in this study were present at lower concentrations.

In order to better describe the relationship between solution Al and crop growth the activities of the various soil solution constituents were also determined. Table 14 depicts the average activity of Al and Ca in soil solution from the surface and the 1 m depths. Aluminum activity values decreased in the surface by 10^{-6} fold following lime application in comparison to the control. As noted earlier, the concentration of Al was not influenced by this treatment. Gypsum application resulted in a 7 fold increase in Al activity at the surface when compared to the control. This response is similar to the effect of gypsum on the Al concentration in the soil solution.

Calcium activity in soil solution averaged 0.67 and 0.69 mM at the 0-5 cm and 1 m depths on the control soils respectively (Table 14). Where lime and gypsum were applied, the Ca activity was increased by 2 and 7 fold, respectively, at the 0-5 cm depth. At 1 m depth, lime produced a similar response but gypsum was without any effect.

Table 14. Influence of surface applied limestone and gypsum at 13 Mg ha^{-1} on Al and Ca activity in soil solution at different depths.

	Al		Ca	
	Depth (cm)			
Amendment	0-5	100-105	0-5	100-105
	uM		mM	
None	2.70	0.78	0.67	0.69
Lime	0.00	0.05	1.20	1.2
Gypsum	18.00	1.20	4.50	0.78

Speciation of Al is important criterion for determining if Al occurs in a form that may be toxic to the plants. Forty percent of the Al in the control treatment was predicted to be trivalent in the surface 5 cm of the soil profile, while over 50% of the Al was estimated to be in a polymeric form (Table 15). Where lime was applied 100% of the Al was present in polymeric forms. In the gypsum treatment 81% of the Al was in trivalent form and 12% in polymeric forms.

At the 1 m depth, in the control treatment 31% of the Al was in trivalent form and 66% in the polymeric form. Lime application reduced the trivalent Al by 10 fold, 97% being in the polymeric form. Aluminum present in the gypsum treatment was 64 % trivalent and 36% polymeric.

It is essential that the relative proportions of the various Al forms in soil is known in order to predict the potential for Al toxicity to plants. As a rule, trivalent Al is generally considered to be very toxic to plants in comparison to its polymeric forms (Adams, 1984).

Table 15. Influence of surface applied limestone and gypsum at 13 Mg ha^{-1} on Al speciation in soil solution at different depths as predicted by Geochem.

Depth	Amendment	Al^{3}	$AlSO_4$	$AlBO_3$	$Al(OH)_x$
cm		%			
0-5	None	39.8	4.5	3.1	52.6
	Lime	0.0	0.0	0.0	100
	Gypsum	81.3	4.7	1.5	12.4
100-105	None	31.4	2.2	0.8	65.5
	Lime	2.6	0.3	0.3	96.9
	Gypsum	63.7	0.0	0.8	35.5

8.5 LITERATURE CITED

Adams, F. 1984. Soil acidity and liming. ASA Monograph Agronomy 12, 2nd edition. Amer. Soc. Agron., Madison, WI.

Δ Adams, F., C. Burmester, N. V. Hue, and F. L. Long. 1980. A comparison of column displacement and centrifuge methods for obtaining soil solutions. Soil Sci. Soc. Am. J. 44:733-735.

Adams, F. and B. L. Moore. 1983. Chemical factors affecting root growth in subsoil horizons of Coastal Plain soils. Soil Sci. Soc. Am. J. 47:99-102.

Adams, F. and Z. F. Lund. 1966. Effect of chemical activity of soil solution aluminum on cotton root penetration of acid subsoils. Soil Sci. 101:193-198.

Bartlett, R. J. and D. C. Riego. 1972. Effect of chelation on the toxicity of aluminum. Plant Soil 37:419-423.

Bohn, H. L., B. L. McNeal, and G. A. O'Connor. 1985. Soil chemistry. 2nd edition. John Wiley & Sons, New York, NY.

Donohue, S. J. and S. W. Gettier. 1979. Laboratory procedures, Virginia Polytechnic Institute and State University Soil Testing and Plant Analysis Laboratory. Virginia Polytechnic Institute and State University, Pub. 853.

Foy, C. D. 1984. Physiological effects of hydrogen, aluminum, and manganese toxicities in acid soil. p. 57-86. In F. Adams (ed.) Soil acidity and liming. ASA Monograph Agronomy 12, 2nd edition. Amer. Soc. Agron., Madison, WI.

Foy, C. D. 1974. Effects of aluminum on plant growth. p. 601-642. In E. W. Carson (ed.) The plant root and its environment. University Press of Virginia.

Kinraide, T. B., R. C. Arnold, and V. C. Baligar. 1985. A rapid assay for aluminum phytotoxicity at submicromoliar concentrations. Physiol. Plant 65:245-250.

Magistad, D. C. 1925. The aluminum content of the soil solution and its relation to soil reaction and plant growth. Soil Sci. 20:181-228.

McGeorge, W. T. 1924. Iron, aluminum, and manganese in the soil solution of Hawaiian soils. Soil Sci. 18:1-11.

Rechcigl, J. E., D. D. Wolf, R. B. Reneau, Jr., and W. Kroontje. 1986a. Influence of surface liming on the yield and nutrient concentration of alfalfa established using no-tillage techniques. Agron. J. 77:956-958.

Rechcigl, J. E., R. B. Reneau, Jr., D. D. Wolf, W. Kroontje, and S. W. VanScoyoc. 1986b. Alfalfa seedling growth as influenced by Al, Ca, and pH. Commun. Soil Sci. Plant Anal. 17:24-44.

Richburg, J. S. and F. Adams. 1970. Solubility and hydrolysis of aluminum in soil solutions and saturated paste extracts. Soil Sci. Soc. Amer. Proc. 34:728-734.

Rios, M. A. and R. W. Pearson. 1964. The effect of some chemical environmental factors on cotton root behavior. Soil Sci. Soc. Amer. Proc. 28:232-235.

Simpson, J. R. and A. Pinkerton. 1978. Effect of subsoil acidity and liming on the root behavior and shoot growth of some leguminous plants. Inf. Ser-N.Z. Dep. Ind. Res. 134 (Plant Nutr., V.2) p. 449-455.

Sposito, G. and S. V. Mattigod. 1980. GEOCHEM: A computer program for the calculation of chemical equilibria in soil solution and other natural water systems. Kearney Foundation of Soil Sci., Univ. of California, Riverside.

Chapter IX

SUMMARY AND CONCLUSIONS

Soil acidity is a major cause of low yields of alfalfa in the Southeastern United States. In vitro and field studies were conducted to investigate selected limiting and/or toxic factors in an attempt to determine conditions which would permit establishment and production of no-till alfalfa on acidic soils. Based on this study the following conclusions can be made:

1. pH 4.5 is not detrimental to alfalfa growth in nutrient solution unless Al is present.
2. Addition of Ca to a nutrient solution containing Al helps alleviate toxic effects of Al on alfalfa growth.
3. No-till alfalfa can be grown on acid Ernest silt loam soil, provided it is surface limed.
4. Tissue N levels were increased in alfalfa grown on soils which received surface lime indicating increased N fixation on surface limed soils.
5. Roots penetrated to the 1.5-m depth under acidic conditions and were more prevalent at the 1.0 and 1.5 m depths in surface limed soils compared with unlimed soils.

6. Root penetration into acid soil is increased by a pH of 6.5 and the presence of Ca.
7. Nitrogen fixation was increased on acid soil containing adequate Ca and a pH of 6.8.
8. Calcium addition enhances alfalfa yields on an acid Ernest silt loam.
9. Alfalfa yields increased in response to Mo or N indicating the importance of Mo in establishing alfalfa nodulating system for N assimilation.
10. Alfalfa yields were not reduced when the surface lime application was half of the recommended rate, suggesting that the lime recommendation for the state of Virginia could be lowered when surface lime is employed.
11. Soil exchangeable Al is not a good indicator of Al toxicity since some of its forms may be insoluble and thus non toxic.
12. The activity and speciation of Al in soil solution is important for ascertaining the suitability of Al-containing soils for growing alfalfa.

The two page vita has been removed from the scanned document. Page 1 of 2

The two page vita has been removed from the scanned document. Page 2 of 2