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Some chemical environmental factors influencing primary root penetration into the subsoil.

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TIMOTHY OKUNOLA FADAYOMI

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SOME CHEMICAL ENVIRONMENTAL FACTORS INFLUENCING PRIMARY ROOT PENETRATION INTO THE SUBSOIL

^A Dissertation Presented

By

Timothy Okunola Fadayomi

Submitted to the Graduate School of the University of Massachusetts in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

April 1974

Soil Fertility and Plant Nutrition

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> U.S. Department of Agriculture Hatch 305

SOME CHEMICAL ENVIRONMENTAL FACTORS INFLUENCING PRIMARY ROOT PENETRATION INTO THE SUBSOIL

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DEDICATED

to

God, my mother, my brothers, and brochers, for their understanding.

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ABSTRACT

Short-term, split-root experiments were conducted to study the effects of calcium, magnesium, potassium, aluminum, and hydrogen ions on the rates of primary root growth of seedlings of two crop species, namely, romaine lettuce (Lactuca sativa var. longifolia, Lam., cv. Parris Island) and pepper (Capsicum frutescens var. grossum, Bailey, cv. Pennbell), in a growth chamber. The upper portion of the root medium was an adequately limed and fertilized loamy surface soil. The lower portion was either a nutrient solution at various Ca, H, Mg, K, and A1 levels or a subsoil material at various nutrient and pH levels. Two naturally occurring, widely separated acid subsoils were studied. Calcium ion concentrations of ¹ and 72 ppm respectively were required to obtain near maximum lettuce and pepper primary root growth rates in subsurface 1/5-strength Steinberg solutions. Significant reduction in lettuce and pepper primary root growth in subsurface culture solutions occurred when the ratios of molar activities of ^H to Ca exceeded 0.03 and 0.015, the molar ac tivities of Al exceeded 0.1 X 10^{-5} and 0.15 X 10^{-5} , or the ratios of molar activities of Al to Ca exceeded 0.001 and 0.0005 respectively. The toxicity of ^H ions was a factor only at pH values below 6.0. Only 0.25 ppm Al was sufficient to significantly inhibit the primary root growth of lettuce in the presence of either ³⁶ or 100 ppm subsurface solution Ca and that of pepper in the presence of either ²⁰⁰ or 300

V

ppm subsurface solution Ca, although primary roots growing at the higher levels of Ca were less susceptible to Al damage within the range of ⁰ to 1.0 ppm Al studied. ^A phosphorus concentration of 6 ppm was sufficient to significantly inhibit pepper primary root elongation in the presence of adequate amounts of Ca in subsurface culture solutions. However, increased lateral root growth with increasing ^P concentration was observed. Moderate to high concentrations of ^K and Mg significantly increased the primary root growth of lettuce and pepper in the presence of Ca in subsurface culture solutions. In no instance did addition of increasing amounts of ^K or Mg in the presence of Ca result in a decrease in the primary root growth of either lettuce or pepper in subsurface culture solutions. The growth responses of lettuce and pepper primary roots in the subsoils could not be explained by the concentrations and activities of the individual cations per se nor by the H/Ca, Al/Ca, and Ca/total-cation ratios. It was concluded that cognizance be taken of the stimulation of primary root growth by Mg and ^K in addition to the observed inhibition of primary root growth by H, Al, and possibly ^P in an intricate process of determining critical Ca concentrations for optimum primary root growth of these two crop species in subsurface culture solutions and in the subsoil.

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INTRODUCTION

While the importance of the topsoil has been emphasized in plant nutrition, it is, nevertheless, ^a wellestablished fact that the roots of most crop plants penetrate well into the subsoil, that is, below 6 to 8 inches.

Since water movement in unsaturated soil is too slow to be an important factor in supplying the requirements of rapidly transpiring plants, roots must continue to proliferate into unexploited zones throughout the growth cycle of the plant in order to utilize most effectively the moisture and nutrients stored in ^a soil profile. This increased proliferation also has the added advantages of increased resistance of forest species to fire or of grain crops to lodging. At a period of great awareness of environmental pollution such as this, possibilities of utilizing subsoil reserves of moisture and nutrients might obviate the necessity of overfertilizing the surface soil with its implications for underground drainage and pollution of rivers and lakes.

This investigation was suggested by the evidence that roots of certain crops are frequently unable to exploit the lower horizons of soil profiles in the eastern part of the United States. In the more acid subsoils of this region, such chemical factors as Ca deficiency, ^H ion toxicity, or Al toxicity have been cited as responsible for inhibited root growth.

Root penetration into the subsoil may not be due to concentrations of individual ions per se but due to indirect factors such as changes in the chemical environment of the root that are not measurable directly in terms of ionic concentrations. Although the requirements have been defined for very few crops, little else is known of the interactions among these factors or of the variation in requirements for deep rooting among other crop species and varieties.

The objective of this investigation, therefore, was to study the effects of calcium, magnesium, potassium, aluminum, and hydrogen ions on the rate of primary root elongation of two crop species in subsoils and subsurface nutrient solutions. By studying the effects of the interactions among these chemical factors, it was hoped to define the requirement or variation in requirements for deep rooting in the subsoil by these crop species.

LITERATURE REVIEW

Introduction

The rate at which plants can extend their roots under favorable conditions is remarkable. For example, Dittmer (35) reported that total root length increase, including root hairs, of ^a single rye plant (Secale cereale, L.) grown in ¹ cubic foot of soil for ⁴ months averaged 3.1 miles per day.

Different plants and plant varieties possess inherent rooting characteristics (62, 121, 139, 169, 170). Also the rooting patterns of common crop plants vary widely from soil to soil. For example, roots of corn (Zea mays, L.) were found to extend to a depth of nearly ⁵ feet in soils developed from medium- to coarse-textured glacial till in Illinois, but they seldom penetrated beyond 3 feet in fine-textured subsoils (41). Similarly, in New Jersey, effective rooting depth of sweet corn (Zea mays var. saccharata, Bailey) varied from less than 1 to over 3 feet among several soil types (31). Similar variation has been noted for other crops. It would therefore appear that root development in any given crop would proceed according to a genetically determined pattern as modified by both chemical and physical environmental influences.

As regards the influence of physical environment on root behavior, cognizance should be taken of such factors as

soil texture, compaction, mechanical impedance and their indirect effects on other soil physical problems that exert influence on the moisture and aeration requirements for root growth and on continued root exploration of new soil masses deeper in the soil profile.

The Fertility of the Subsoil as it Influences the Rate and Extent of Root Growth

A. The Relative Unproductivity of Subsoils

Both practical observations and scientific investigations have conclusively shown that most subsoils are usually much less productive than surface soils. This is especially true for non-legume crops. However, the nature of the cause of the failure of roots of crop plants to make satisfactory growth in the subsoil or on subsoil material is still ^a disputed question.

Alway et al. (6) found that the subsoil material of Nebraska loess soils was unproductive with corn but showed no "rawness" towards inoculated legumes. They made the further observation that unproductivity of subsoils from humid regions towards inoculated legumes is probably due to lack of availability of phosphorus or potassium or both. Lipman (104), however, questioned the existence of humid subsoils which were sterile towards inoculated legumes and denied that lack of phosphoric acid or potash could be the cause of such unproductivity. Harmer (65) found some of the

humid Minnesota subsoils to be as productive with inoculated alfalfa (Medicaqo sativa, L.) as the corresponding surface soils. Others, however, he found quite unproductive. Such unproductivity was associated with neither an especially low nitrogen content nor a lack of carbonates. McMiller (114) used alfalfa in pot tests to show that certain Minnesota subsoils which previously had been found "raw" towards inoculated legumes were rendered as productive as the corresponding surface soils when soluble phosphorus and potassium fertilizers were added. Millar (117, 118, 119, 120), from work with several soil types, concluded that the poor growth of corn in soil from A_2 and B horizons is due very largely to lack of available nutrients and that very large quantities, particularly of phosphorus, must be added to satisfy the adsorptive capacity of the soil and make plant growth commensurate with that obtained when surface soil is used. Conner (30) carried out pot tests with wheat on surface soil and subsoil horizons of Crosby and Clyde silt loam soils and found that nitrogen and phosphorus were very deficient in all subsoils as compared to surface soils; potassium and lime were less deficient than either nitrogen or phosphorus. He also found that (a) Indiana subsoils show a greater need for phosphorus than do the surface soils for growth of both legumes and non-legumes, this need being often greater the farther from the surface the subsoil is taken; (b) nitrogen is more deficient for grain crops in subsoils than it is in

surface soils; subsoils do not show a deficiency of nitrogen when inoculated legumes are grown; and (c) when more than one crop is grown on the same subsoil, the first crop is rela tively more in need of nitrogen and phosphorus than are the succeeding crops. He then suggested that eroded surfaces and subsoils exposed in regrading operations or in fills using subsoil are in need of liberal phosphate and nitrogen fertilization when seeded down to non-legumes, while legumes on such surfaces should be inoculated and heavily fertilized with phosphates; lime and potash may be added as needed since Indiana subsoils are generally in no greater need of potassium than are surface soils. Comparative data for surface soils and subsoils for 460 soils of the United States (158) subsequently indicated that, for subsoils of the humid regions at least, phosphorus deficiency is an important factor in the unproductivity so often observed. These findings have been substantiated in subsequent investigations (61, 76, 150).

B. Attempts at Improving the Productivity of the Subsoil

In an attempt to make the subsoil a more desirable medium for plant growth, subsoiling has been frequently recommended as a conservation practice on soils. The intensive tillage and traffic associated with continuous row cropping tends to develop a compacted layer below plow depth which may restrict root growth and reduce water movement through the soil. Subsoiling to break up this layer is thought to reduce runoff and erosion and to stimulate

deeper rooting of crops through improved drainage and aeration. Subsoiling has also been proposed as ^a means of applying lime and fertilizer to acid and infertile subsoils to stimulate deeper rooting of crops. Better use of the subsoil water and deep-placed fertilizer in dry years might very well be a means of increasing crop yields.

^A survey by the National Fertilizer Association (115) in 1954 showed considerable research in progress on deep tillage and subsoil fertilization throughout the United States. Results of completed studies were conflicting, but most reports indicated very little benefit from either practice. Duley (36) summarized the research in the Great Plains and found no evidence of the general benefits from subsoiling the semiarid soils. In the North Central States (72, 97) where frost penetration is deep, subsoiling and deep placement of lime and fertilizer did not increase yields in most places. On the claypan soils of Missouri (85) small increases in yields of corn, soybeans, and alfalfa were obtained by subsoiling but were not of practical significance. In Florida (145, 146) and Louisiana (131) where frost action is slight, both subsoiling and deep placement of fertilizer increased the yield of corn and cotton on soils with traffic pans or hardpans. This was particularly evident in years of below—average rainfall. In Connecticut (34) leaf and root growth of tobacco (Nicotiana tabacum, L.) was increased by shattering and fertilizing the subsoil of an intensively

cultivated sandy loam soil. In New Jersey, Nissley (129) found that subsoiling with deep placement of lime and phosphorus frequently increased the yield of vegetable crops in field trials. However, Anderson et al. (7) and Brill et al. (18), using the same practice on loam and sandy loam soils, reported no significant yield increases even in dry years for the vegetable crops grown over 4- and 7-year periods. Insofar as deep tillage and fertilization throughout the entire tilled zone could possibly play an important role in subsoil moisture conditioning and enable crop roots to penetrate deeper, with the years, it should help to increase the organic matter content of the subsoil and therefore contribute to the stability of its aggregates.

C. Rate of Root Growth and Root Activity

Studies in Tanzania and Sudan (98) have indicated that with adequate rainwater penetration, the rate of root penetration of certain annual crops is without regard to texture of the different soil horizons. At one site, the profile consisted of moderately acid sandy loam surface soil, an acid subsoil, and an underlying concretionary or sheet ironstone or acid gneiss at ⁵ to ⁷ feet. At the other site, the profile consisted of heavy clays. Groundnut (Arachis hypo gaea, L.) tap roots, for example, penetrated the profiles at both sites very rapidly in the first ¹¹ days at 1.24 and 1.31 in./day respectively. Slower rates of penetration were observed only when tap roots approached weathered rock in one instance and dry soil in the other. Subsequent root depth

was approximately the same as moisture penetration. Ward (168) in Canada observed that greenhouse tomato roots grew in ^a surface-soil medium at ^a fairly constant rate of about 1.3 in./day; the longest root measured was about 58 inches and it had penetrated to ^a depth of ⁴⁰ inches into the soil. He also observed that different watering schedules in several experiments appeared to have little effect on the distance or rate of growth. Pearson and Lund (133) in the U.S.A. also found that under favorable soil chemical and physical conditions, such as an adequately limed and fertilized sandy loam surface soil, the rate of cotton primary root elongation was remarkably uniform and was 3.3 times as fast as stem elongation rate when root had reached the 170-cm maximum depth at which observation could be made. The period of maximum root proliferation coincided with the maximum rate of plant height increase and occurred during the period immediately following initiation of the reproductive cycle. There were no identifiable periods of suspense in root extension until the onset of final cessation of root growth. This occurred following maturation of the first boll set. Soileau and Engelstad (152) have suggested that adverse chemical or physiological effects related to extreme soil acidity may be dominant over other physical and chemical factors that inhibit cotton root penetration in Dickson fragipan subsoil in Alabama.

While considerable research aforementioned suggest extensive activity of roots in the deeper layers of the soil, the availability of radioactive tracers has made it possible

to calculate the contribution of separate soil layers to crop nutrition. Murdock and Engelbert (124) and Shain and Kashmanova (149) concluded that phosphorus can be absorbed in substantial amounts from the subsoil even from depths of about 100 cm. Far more interesting are the results of experiments where both uptake from and the amount of roots in the deeper layers of the soil have been estimated. Fox and Lipps (43) concluded that where the topsoil became dry, ³ per cent of the roots of alfalfa at ²⁰⁰ to ⁴⁰⁰ cm depth absorbed ⁶² per cent of the mineral uptake. It has also been shown elsewhere that for corn and oats (Avena sativa, L.) the contribution in nutrition from the soil at ⁶⁰ to ⁸⁰ cm depth exceeded that of the percentage of roots, namely, 1.7 per cent roots contributed 9.2 per cent of the uptake of phosphate (174). Nakayama and van Bavel (126) also noted that phosphorus absorption of ⁷ per cent of the total could be accomplished by a ² per cent root mass at 60 cm depth. Fox and Lipps (44) demonstrated that ⁶⁰ per cent of the total root activity of alfalfa took place at depths below ⁷ feet as ^a result of the more favorable moisture conditions. Since root activity, and not necessarily root yield, is considered by some researchers to be the real determinant of root effectiveness, the work of Eck and Davis (38) and Wiersum (174) lend strong support to the idea that roots in the lower depths of a soil profile are just as active as those in the surface soil and are therefore potential absorbing units for water and nutrient

uptake provided aeration is not limiting.

The general conclusion is that the roots in the subsoil certainly are of potential value in feeding crop plants. Their relative performance may even rise to high values depending on the circumstances in the topsoil. Their contribution to plant nutrition will, however, depend on the fertility of the subsoil layers, the moisture content of these layers, and the amount of roots that have been able to develop in these regions.

Nutritional and Toxic Factors Affecting the Subsoil

The failure of roots of crop plants to make satisfactory growth in the subsoil has largely been attributed to hydrogen ion toxicity, aluminum and manganese toxicity, or calcium deficiency.

Hydrogen Ion Toxicity

There is usually the difficulty of interpreting a soil pH value in terms of an isolated and independent variable. Is the failure of plants to thrive in an acid soil due to a high hydrogen ion concentration per se or to such other unfavorable factors of which ^a low soil pH is generally symptomatic; for example, a depletion of Ca and the presence of toxic amounts of Al and Mn in the soil solution? Nevertheless it is clear that variations in hydrogen ion concentration have a significant influence on the absorption of many inorganic ions. However, provided the reaction does not fall

below pH 4.0 to 4.5, there appears to be little detrimental effect on growth of most crop plants when the nutrient supply is adequate and the amounts of Al and Mn are not excessive.

Some nutrient solution experiments have been reported in which secondary nutritional effects and pH drift were minimized. Arnon and Johnson (9) showed that two acid-sensitive crops, lettuce (Lactuca sativa, L.) and tomato(Lycopersicon esculentum. Mill.), and an acid-tolerant crop, bermudagrass (Cynodon dactylon, Pers.), failed to develop to any extent at pH 3.0. Both root and shoot growth of tomato and lettuce decreased sharply between pH 5.0 and 4.0 and also between pH 8.0 and 9.0. On the other hand, good growth of bermudagrass occurred throughout the range pH 4.0 to pH 8.0; root weight was maximum at pH 4.0, although shoot weight was depressed as pH decreased below 6.0.

It has been suggested that the influence of extreme acidity or alkalinity on growth, water uptake, and nutrient absorption is largely the reflection of a primary injury to the absorbing root cells (8, 159). Substantial alleviation of damage was effected as the Ca supply in the nutrient solution was raised. Varying the Ca supply from ¹ to ⁴ to ¹⁴ meq/ liter resulted in progressive increases in growth of lettuce and tomato at pH 4.0. The data suggest that one of the differences in species sensitivity to high acidity is a difference in ability to absorb Ca at low pH.

Burstrom (20), working with wheat (Triticum aestivum,

L.), employed both cell multiplication and elongation as criteria of hydrogen ion-effect on root growth. Growth decreased as pH decreased from about 6.0, with ^a particularly sharp drop below pH 5.0. Both Burstrom and Audus (10) suggested that the growth depression at low pH was because of decreasing dissociation of auxin in the roots as nutrient solution pH was reduced. However, Arnon and Johnson showed that cell sap pH did not change when nutrient solution pH was varied from 4.0 to 9.0.

In conclusion, two general effects may be induced by high acidity. One is ^a competitive action of hydrogen for initial binding reactions of cations (53) while the other is ^a more prolonged and drastic consequence of direct damage to cellular membranes.

Aluminum Toxicity

Early investigators (66, 162) have demonstrated that the toxic effects produced in certain plants grown on acid soils are caused primarily by the presence of aluminum in the soil. With most crop plants, considerable accumulation of Al occurs in the roots and relatively little is transported to the above-ground portions (134, 177). However, the Al content of the leaves of five calcifugous species growing under natural conditions was found to exceed that of the roots (74), and considerable increases in Al content of the above-ground tissue have been obtained in other experiments (77, 78). It has been further shown that crop plant species

and varieties within the same species differ widely in their tolerance to acid soils and nutrient solutions containing high levels of soluble or exchangeable Al (46, 47, 48, 51, 142). The nature of this differential Al tolerance has, however, not been clarified, very likely because the mechanism of Al toxicity is still in question.

Some of the beneficial effects of liming are commonly ascribed to immobilization of Al in the soil thereby preventing Al toxicity from developing (148). In fact, lime applications based on neutralization of the exchangeable Al are found to be ^a suitable criterion for the measurement of lime requirement defined as the amount of lime necessary for maximum crop production on Ultisols and Oxisols, and this amount of lime is only a fraction of the amount required to raise the soil pH to 6.5 (92, 141). Nevertheless, lime additions seldom affect the Al contents of the above-ground portion of crop plants to an appreciable extent (54). On the other hand, Ouellette and Dessureaux (130) found that alfalfa clones which transported less Al to the tops were more tolerant to high Al concentrations and were able to accumulate higher amounts of Ca. Furthermore, species such as mustard can accumulate substantial amounts of Al in leaves and yet remain rather unaffected (88), and toxic effects may result from excess Al with very little change in the Al content of the foliage.

The addition of phosphorus to acid soils often has been found to overcome the toxic effects of Al. Following ^P

additions to an acid soil, Burgess and Pember (19) found that better growth occurred even though the Al contents of barley and lettuce tops on treated and untreated soils were equally great. Experiments in which ^P and Al variables have been studied in culture solutions do not reveal large and consistent decreases in Al contents of the tops upon the addition of phosphorus (45, 134) although root contents usually are considerably increased.

Early studies to determine Al distribution within root tissue were made by McLean and Gilbert (113). Their data indicated high localization of Al in the cortical cells of corn roots. Most of the Al was found in the cytoplasm rather than in the cell walls or vacuoles, and especially dark staining with haematoxylin was observed in the nuclei. In view of other studies, absence of Al in cell walls is subject to doubt. Wright and Donahue (178), using radioactive P, noted considerable amounts of Al on the surface of the epidermal cells of barley (Hordeum vulgare, L.) roots. Al could not be detected, however, in the inner walls of the endodermis or in the vascular system. It was concluded that precipitation of aluminum phosphate occurred internally as well as on the epidermal surfaces. Wallihan (167) used extracting procedures similar to those earlier used by Wright and concluded that almost all of the precipitate was on the external surfaces of ladino clover (Trifolium repens, L.) roots. Clarkson (28) concluded that an absorption-

precipitation reaction was involved and that it was roughly confined to cell walls.

The inhibition of ^P translocation to shoots in the presence of Al has been demonstrated by several workers (45, 46, 69, 138). However, Randall and Vose (138) found that ^P contents of ryegrass shoots increased after ⁸ weeks in toxic Al solutions although the increases were not as large as occurred in the roots. Humphries and Truman (77) showed that ^P contents of Monterey pine shoots were increased in the presence of Al. Other data suggested that ^P content of tops was not affected appreciably by Al even though growth was depressed (130, 140). Tea, a noted Al accumulator, contained considerable amounts of ^P in the aerial portions even when the Al contents were high, suggesting that there was little effect on ^P mobility in this plant (25).

Aluminum has been shown to depress the uptake of Zn by citrus plants (58) and potato varieties (99). Harward et al. (67), however, noted that Fe content of the older leaves of lettuce was increased when severe growth inhibition was induced by Al. The detoxifying influence of Al on the injurious effects of excessive Cu on citrus has been reported by Liebig et al. (102). Low Al concentrations stimulate Cu uptake and transport while high Al concentrations depress Cu uptake and transport in young wheat seedlings (14, 71). Since the Al-stimulated Cu transport was, however, eliminated by adding small amounts of CaSO₄ such as 10^{-4} M, it is

doubtful if this stimulation would be expressed when plants are growing in a complete nutrient solution or in soil culture whereas the depressing effect of A1 may well exist.

The ^K content of both young and old leaves of lettuce was sharply decreased under extreme Al toxicity (67) but little effect of Al on ^K content was noted in spinach, barley, and saltbush (140). Aluminum depressed Rb uptake from acid solutions by 6-day-old excised wheat roots at lower concentrations of Rb but no depression occurred at higher Rb concentrations such as commonly used in nutrient solutions, and a stimulatory effect was noted. There is therefore reason to expect that at the concentrations of ^K existing in the soil solution a depressing effect may occur.

Aluminum strongly depresses the uptake of Ca (86, 130), but there has been little success in overcoming Al toxicity at concentrations of the order ¹⁰ - ²⁰ ppm by increasing Ca concentrations to high levels (24, 26). Clymo (29) found the inhibiting effect on root growth to be greatest when the Ca supply was low. Aluminum has also been associated with decreases in the uptake and utilization of Mg (111) by barley roots. Thus the possibility exists of Al restricting the entry of Ca and Mg into specific reaction loci in root cells. If Al indeed does create an unsuitable distribution of divalent cations in macromolecular configurations at the subcellular level, it would seem reasonable that the stabilizing influence apparently exerted by Ca and Mg on macromolecular
configurations of cellular membranes and ribosomal particles would be drastically modified and this effect would not be totally alleviated by increasing the Ca supply alone. Recently, Lance and Pearson (96) studied the initial effects of A1 on cotton seedling roots and found that prior exposure to low concentrations of Al not in excess of 0.30 ppm subsequently inhibited seedling root uptake of water, Ca, Mg, K, P, and NO₃ from $1/4$ Hoagland's solution. The concurrent inhibition of all types of uptake indicated that the permeability of the plasmalemma was reduced possibly due to interference in a function of Ca.

Roots usually are first affected when plants are exposed to toxic Al concentrations, with damage to the tops occurring later (26, 103, 113). Similarly, at low Al concentrations, root injury may be noted without damage to shoots. Generally, the roots develop a brownish color and lose turgidity. Main roots fail to elongate rapidly and become thick, swollen, and distorted. Laterals are initiated but their development is limited and they largely remain as short abortive stubs. In some cases, new roots are initiated above the solution level but die upon reaching the solution (66, 177). Damage resulting from interference by Al in accumulation of various nutrients is further accentuated in soils by restriction of the root system so that only small volumes of soil are exploited. Aluminum toxicity has been associated with disruption of tissue organization of root

laterals, failure of laterals to penetrate the cortex (66), undifferentiated tumor-like tissue (140), damage of onion root apices and severe disruption of further growth (27), and amitosis in the apical meristem cells of bent-grass (Aqrostis stolonifera, L.) and barley roots (26, 147).

Most experiments have been concerned with Al toxicity on young seedlings and the effects are often severe. Little attention has been paid to the relative degree of tolerance at different stages of plant development. It has, however, been suggested that plants are most susceptible in the early stages during the transition from dependence on seed reserves to dependence on external sources of nutrients. If growth is only partially restricted by Al, the seedlings may be able to survive this crucial stage and develop normally later on. More favorable growth conditions during the seedling stages result in greater resistance to Al toxicity (80).

The presence of Al sometimes results in altered growth which is not necessarily detrimental. For example, Lipman (105) found a large increase in corn ear production resulting from ¹ ppm Al in culture solution. Hortenstine and Fiskell (73) showed root growth of sunflowers to be enhanced slightly by 4 ppm Al, and Harward et al. (67) reported some increases in leaf and root weights of lettuce at low Al concentrations. Young seedlings of four species which vary in their adaptation to acid soils showed greater root development when Al was present during germination (60). Shoot growth, on the other hand, was not stimulated at the Al concentrations which resulted in greater root production. Clarkson (26) also indicated an increase in root elongation of seedlings of bentgrass when Al was added at low concentrations. These observed beneficial effects of Al on growth have been ascribed to prevention of toxic efrects of other ions such as H (40), Cu (14, 58, 71), Zn (57), or the possible inducement of changes in the absorption rates and metabolism of mineral nutrients such that the rate of growth of one plant organ may exceed those of others (80).

The detrimental effects of Al toxicity thus result from a combination of factors, the manner in which they are brought about being different under various experimental conditions and with different plants. Root growth appears to be most affected by severe inhibition of cell division and inducement of other metabolic aberrations which parallel this effect. In the shoots, however, the most common effect is due to a lack of ^P resulting from greatly impaired ^P translocation, and this usually develops only after root growth has been adversely affected.

Manganese Toxicity

Manganese activates a large number of different enzymes including those involved in hydrolysis, oxidationreduction, decarboxylation, and phosphate transfer reactions. There are many instances when Mg and Mn can substitute for each other with variable degrees of efficiency (80). The

importance of Mn in photosynthetic processes and maintenance of chloroplast structure has been well documented (93, 135, 155). Lyttleton (110) found Mn to be especially effective in preserving the stability of ribosomal particles, although for optimal effects Ca and Mg were required as well.

Manganese toxicity may be expressed in two quite different ways, namely, an indirect effect resulting in Fe deficiency and a direct toxic action of excessive Mn. The latter, however, constitutes the main effect of acid injury. Hewitt (70) and Lohnis (106, 107) have described the direct toxicity symptoms of ^a large number of crop species and listed the differences in tolerance to excess Mn. With excess Mn, roots are normally brownish and the older leaves develop a speckled appearance produced by highly localized accumulation of Mn. Older leaves are, however, more tolerant to excessive Mn. With some species a marginal cupping of the leaves occurs. Often ^a distinct chlorosis develops on the leaf margins, and an interveinal chlorosis may be induced. Black necrotic spots may occur on petioles and leaf veins; under severe conditions they coalesce and form long necrotic streaks on the conducting tissue.

Calcium, iron, and ammonium ions exert a detoxifying influence on excessive Mn. High ^P supply has also been shown to decrease Mn toxicity (15) although contrasting effects were observed by Morris and Pierre (122).

Even though the toxic limits of Mn concentration, even in solution, have not been clearly defined and there are known differences in tolerance among plant species, it would seem that for many plants ¹⁰ ppm is about the maximum concentration in solution that can be tolerated without damage (4, 15, 123). As in the case of aluminum, the level of other ions in solution would modify the toxic effect of Mn at a given concentration.

Manganese, unlike aluminum, has little direct effect on roots (15, 56, 144). Nevertheless, Mn toxicity could be masked by the presence of Al toxicity in some acid soils (50). Besides, counteracting effects of Al and Mn levels have also been observed on the growth, mineral nutrition and tuber yield of potato plants (100, 140). However, the opposite Al and Mn tolerances of ''Atlas 66'' wheat variety from North Carolina and "Monon" wheat variety from Indiana strongly suggest that tolerance to one acid soil factor even in ^a given plant genotype does not necessarily mean tolerance to another (52).

Calcium Deficiency

Calcium deficiency is a potential factor in root development in acid soils. Its removal from the ambient medium results in rapid and dramatic abnormalities in plant tissue. An uneven growth pattern results in margins of leaves being restricted. Differential growth rates of the cells produce distinct cupping and hooking patterns.

Collapse of stem and petiole tissue often is noted. Premature flower abscission, collapse of ovules, poor seed development, and collapse of cells at distal portions of fleshy fruits, for example, blossom-end root of tomato and pepper, are further characteristics of low Ca availability. There is dramatic cessation of root growth at the apical meristem, and the development of lateral primordia is seriously arrested. In all cases, the general effects are induced most rapidly in the young meristematic tissue (70).

It appears probable that the fundamental physiological influence of Ca is through an effect on structural characteristics of cellular membrane systems. Light microscopic examinations of tissues exposed to little or no Ca reveal profound effects on cellular and subcellular structure (11, 32, 33, 91, 109, 153, 154, 156). These studies reveal rapid development of abnormal mitoses in which the spindle develops abnormally, separation of chromosomes, aggregation of chromatin near the nuclear membrane, and loss of much of the cytoplasm. Failure to produce new cellwalls after division results in development of some binucleate cells. Alterations in cell wall development are found only after cytoplasmic aberrations become distinct (154). Electron microscopic examinations reveal that cell expansion in the shoot apex of young barley seedlings ceased within ² days after transfer to Ca-free solutions (112). Membrane dissolution and loss of subcellular structure also was noted

in Ca-deficient barley roots, the breakdown of the tonoplast being especially evident. Removal of Ca by prolonged treatment with ethylenediaminetetraacetate (EDTA) resulted in degradation of polynucleotides, loss of selectivity in ion accumulation, restriction in respiratory and phosphorylative activity of the mitochondria presumably by disrupting the membraneous structure of the organelle (64), changes in the initial entry of ions into root cells (42), and leakiness of cell-wall membranes (163). However, Van Steveninck (163) found Sr and Mn to be of some benefit in reversing the leakage caused by loss of Ca.

Calcium--in conjunction with boron--must be present constantly in the entire rooting medium for normal root growth (68) as ^a result of very little translocation within the root tissue to the developing root apex (172). Burstrom (20) has shown that Ca influences root elongation of wheat seedlings through influences both on cell division and on cell elongation and that the influence is moderated by the acidity of the growth medium. Above pH 5.0 cell division was not increased beyond 10⁻⁶M Ca whereas cell elongation still required 10^{-4} M Ca for maximal effects. Subsequently, it was shown that inclusion of boron and iron in the medium modified some of the effects, but that the dominant effect of Ca on root growth at pH values near neutrality was exerted through an effect on cell elongation (21). Retarded elongation of etiolated pea stems as ^a consequence of restricted

cell elongation was also ascribed to an effect of Ca (22). Since growing root tips produce a natural inhibitor of lateral root initiation (160), it would seem reasonable that the destruction of the meristematic cells in root tips resulting from very low Ca supplies, or from interferences in Ca reaching the reactive sites in the cytoplasm (154), could result in some increased lateral root development.

True (161) earlier indicated the importance of Ca in regulating the processes involved in accumulation of various inorganic ions by plant cells. Many other investigators (39, 82, 83, 90, 101, 157, 164) have subsequently shown that accumulation of monovalent ions by root tissue is often accelerated in the presence of Ca. The effect of Ca to increase ^K absorption, to decrease Na absorption, and to prevent Na interference in ^K absorption has also been noted (39). Tanada (157), Handley et al. (63), and Foote and Hanson (42) have indicated that a role of Ca in regulation of ion uptake and leakage is exerted probably by the ion serving as a link in RNA-protein complexes.

High acidity induces the loss of many organic and inorganic cellular components from root tissue and also depresses absorption of many cations. Jacobson et al. (81) observed considerable losses of inorganic P, soluble organic ^P and ^N compounds, ^K and Ca from root tissue at pH 5.0 and below. Hanson (64) observed loss of organic constituents from root tissue when Ca was removed. The data of Rains et

al. (137) clearly confirm the many observations that the injurious effect of ^H ions could be moderated by Ca.

Calcium also exerts effects in many enzyme systems. For example, it activates certain amylases, phospholipases, and kinases (127). Ability of Ca to activate adenosine triphosphatases in root mitochondria (87) and in chloroplasts (12) would suggest some regulation in high energy phosphate production.

The general conclusion to be drawn is that Ca availability in the soil is a function of the kind of cation exchange material and the degree of Ca saturation of this material. However, specific deficient levels of Ca in acid soils are in doubt because soil pH was increased simultaneously with Ca level in many of the reported experiments (5, 116). In such cases, adverse effects of pH and Al may have masked any plant response to Ca (75).

Work Done Specifically on Primary Root Elongation in the Subsoil

Little attention to date has been given to the study of chemical factors influencing root penetration of the subsoil. The results of several split-root experiments confirm the requirement that Ca be present in the subsurface rooting medium even though the surface medium has an adequate amount of Ca (75, 128, 144) since downward translocation of Ca from adequately limed and fertilized surface soil is negligible.

The work of Ragland and Coleman (136) with grain sorghum (Sorghum vulgare, Pers.) and that of Howard and Adams (75) with cotton seedlings suggest that, except for sandy soils, poor root growth in acid subsoils is not generally the result of Ca deficiency. Even though the absolute requirements at the site of root growth are not clear, Ca requirements are very low if other essential ions are in balance and if no toxic ions are present (89, 108). The introduction of another nutrient cation decreases Ca availability to roots through the antagonistic effects of one cation on another (1) Cotton (Gossypium hirsutum, L.) primary root growth at various Ca concentrations in subsurface sulfate solutions was studied. Calcium concentrations ranged from 0.04 to 1.5 meq/liter. It was found that in the absence of other cations Ca was adequate at a concentration of 0.29 meq/liter but was deficient at 0.15 meq/liter. In the presence of Mg and K, however, higher minimum Ca concentrations were recuired for root growth. In fact, Ca deficiency was induced at each of the Ca levels by the addition of sufficient Mg, K, or Mg plus K. The antagonistic effects of Mg and ^K on Ca were evident, with no great difference between the effects of the two (75). Lund (108), working with "Lee" soybeans (Glycine max. Merr.), found that high concentrations of Mg expressed as low equivalent ratios of Ca/(Ca ⁺ Mg) were detri mental to soybean primary root growth. The soybean taproots that grew in the solution with the 0.05 ratio elongated at an

average rate of 1.56 mm/hr for the first ⁴⁸ hours as compared to rates of 2.59 and 3.25 mm/hr in the solutions with the 0.10 and 0.20 ratios. The low ratio of Ca/total cations was, however, less detrimental when ^K was substituted for one-half the Mg applied. Thus replacing half of the Mg with ^K at low ratios of Ca/total cations alleviated root inhibition somewhat but had no effect at the higher ratios of 0.10 and 0.20. It was then suggested that low ratios might be induced in soils low in Ca when fertilized with high analyses non-calcitic fertilizers.

Although the effects of pH per se of subsoil solutions are inadequately defined for most plants, primary roots of cotton seedlings were found to be inhibited only at a solution pH below 4.25 (75). Whereas a Ca concentration of 0.25 ppm was sufficient to obtain maximum soybean root growth rates in a nutrient solution at pH 5.6, progressively increasing Ca levels were necessary for optimum growth as pH values cropped to about 4.0 (108).

The detrimental effect of low subsoil pH on root growth has been demonstrated to be intimately associated with exchangeable or, more particularly, soluble Al (2, 3). Furthermore, the detrimental effect of small amounts or Al on root growth in the subsoil has been well documented (96, 144). However, increasing the Ca level in nutrient solution from 10 to 40 ppm decreased the damage to soybean roots caused by concentrations of 0.5, 1.0, or 2.0 ppm Al in solution (108).

Whereas there was a distinct relationship between root penetration into the subsoil and the factor of subsoil pH, Adams and Lund (2) noted that critical subsoil pH levels and critical levels of toxic Al appear to be different for different soils. Critical levels of Ca also appear to be different for different soils (75) and for different subsurface solution pH's (108). However, in all these reports the subsoils and the subsurface nutrient solutions employed appeared to share a common relationship between root penetration into the subsoil and the molar activities of Ca and Al in the subsurface rooting media. Critical and toxic concentrations of Ca and Al, respectively, in terms of molar activity were quite similar for subsoil solutions in situ and for subsurface nutrient solutions. The Ca requirement for penetration of subsurface rooting media by cotton and soybean primary roots involved Ca/total—cation ratios between 0.10 and 0.15 in subsoil solutions in situ as well as in subsurface nutrient solutions (75) and clearly not below 0.10 in subsurface nutrient solutions (108) respectively. Utilizing two distinct approaches, subsoil or subsurface nutrient solution Al was progressively more toxic to cotton roots as the molar activity of Al exceeded a minimum of about 0.15 x 10⁻⁵ (2), whereas subsurface nutrient solution Al adversely affected soybean roots when the ratio of activities of Al to Ca was greater than 0.02 (108). However, it has recently been shown that the above ratios and critical limits might

be very different for different crops. Peanut (Arachis hypocaea, L.) roots were clearly more tolerant than cotton roots to the low pH and the associated high solution Al of a strongly acid sandy loam subsoil (3).

Dther Nutrients

There is as yet meager evidence of requirement for specific nutrients other than Ca at the point of growth. It has been shown that nutrients in addition to Ca are required for cotton lateral root development, as indicated by the weight of the root systems per unit length, but not for primary root elongation. Elimination of N from the subsurface nutrient solution drastically reduced lateral growth. There was a tendency for lack of P, S, and Mg to reduce lateral development also (132). The effects of N have been observed previously. Bosemark (16) found that ^N deficiency resulted in long, slender roots whereas increasing levels of ^N produced short, stocky roots; the length of wheat root cells increased progressively as the level of ^K in the nutrient solution was decreased from 10⁻² to 10⁻⁴ M. In the reported experiment (132), which extended over a 2—week period, there were some treatment differences in top growth which would suggest that nutrient uptake from the surface soil might not have been adequate for maximum plant growth rate; but it was clear that lack of nutrients other than Ca resulted in more rapid primary root elongation and reduced lateral growth. Subsequently, the observed proliferation of corn roots in

fertilizer bands containing ^N and ^P was ascribed to increased higher order branching of lateral roots in the presence of ^N and P (37). Nitrate, P, and K, in decreasing order of effectiveness, promoted branching of pea roots (173). It was suggested that this effect was exerted through growth substances that regulate root initiation. Evidence supporting this hypothesis was presented by Wilkinson and Ohlrogge (175) who showed that roots of soybean fertilized with N were consistently higher in growth hormone than were roots of unfertilized plants or plants fertilized with ^P alone. However, experiments have been reported in which surface-applied ^N did net depress growth in the subsoil. Increased root development in the lower horizons was found in the case of wheat (95) and several grasses (23, 59). In spite of some apparent inconsistencies, reported observations in general lend credence to the idea that ^N in the subsoil may stimulate root development in that zone even though adequate ^N for plant needs is provided in the surface soil (132).

Because of its immobility in soil and its low level of availability in many subsoils, P would seem to be a possible limiting nutrient for deep root development. However, results of short-term, split-root experiments indicate that P does not have to be provided at the site of root growth for normal root development (144). From deep tillage and fertilization experiments aforementioned, it woulc be reasonable to assume that the presence of available P in the subsoil may

improve root growth in the lower horizons during periods of drought or depressed uptake in the surface soil.

Even though Haynes and Robbins (68) reported that boron should be present in conjunction with Ca throughout the entire rooting medium for normal growth to take place, translocation studies have shown that ^B moves downward in roots (179), which suggests that this element could be translocated from zones of adequate supply in the soil to roots growing in deficient zones.

Rationale for this Investigation

The foregoing would suggest that root penetration into the subsoil may not be due to concentration of individual ions per se but due to indirect factors such as changes in the chemical environment of the root that are not measurable directly in terms of ionic concentrations (166). Indeed, effective counter measures against the detrimental effects of the abovementioned chemical factors on root penetration into the subsoil would therefore depend upon an understanding of the relationships between these factors and retarded root extension or root injury in the subsoil. Although the requirements have been defined for very few crops, little else is known of the interactions among other crop species and varieties.

The main objectives of this investigation, therefore, were (a) to compare the sensitivity of primary root elongation

of six vegetable crop species to high aluminum and hydrogen and low calcium ion concentrations, and (b) thereby to select two vegetable crop species and study the effects of calcium, magnesium, potassium, aluminum, and hydrogen ions on the rate of primary root elongation in subsoils and subsurface nutrient solutions.

MATERIALS AND METHODS

Collection, Processing, and Incipient Analyses of Soils Used

Surface soils, approximately 0" to 7", and subsoils, approximately 9" to 15", were collected in the summer of 1970 from two locations belonging to different parent materials and soil series. The soils were placed in plastic bags, in order to avoid contamination, and subsequently air-dried, sieved with a 2-mm sieve, and stored in 20-gallon cans lined with plastic bags.

The soil from Bristol County, Massachusetts, was taken from the unlimed plot from a ten-year lime experiment. The site was ^a field of alfalfa and grass located within two miles east of Horseneck Beach. The unlimed plot supported only grasses. The soil is a well-drained, very stony fine sandy loam belonging to the Narragansett series. With adequate use of lime and fertilizer, this soil produces all the common crops at ^a high level. Since this area is still important agriculturally, this soil has been in cultivation for much more than a hundred years. The climate in this south coastal region of Massachusetts is distinctly marine with moderate temperatures and high humidity (171). The soil from Franklin County, Massachusetts, was taken from an uncultivated area under secondary forest growth at relatively high elevation adjacent to an alfalfa field which had been

adequately limed and also adjacent to the South Deerfield Experimental Station below. The soil is awell-drained, sandy loam belonging to the Merrimac series. This is ^a part of the Hinckley-Windsor-Merrimac soil association in the central part of the county parallel to the Connecticut River and carrying important agricultural crops such as tobacco, dairy feeds, vegetables, and other cash crops. The climate in this northwestern part of Massachusetts is characterized by warm summers, moderately severe winters, and annual precipitation ranging from 43 to 50 inches; about two-thirds of the precipitation falls during the growing season (151).

The pH of each soil layer was determined in water using a 1:1 soil-water suspension and in $0.01M$ CaCl₂ solution using a 1:2 soil-solution suspension (13) by means of a glass electrode pH meter.

The lime requirement of each soil layer was determined by Woodruff's buffer method (176) using a glass electrode pH meter. The actual amount of lime needed to bring the final pH of each soil layer to the required values upon two wetting-incubation-drying cycles was empirically determined by fractional reduction of the value obtained by Woodruff's buffer method.

The organic matter content of each soil layer was determined by the wet combustion and titration method involving $\overline{1N}$ K₂Cr₂0₇ and 0.5N Fe(NH₄)₂(SO₄)₂.6H₂O (79).

The specific conductivity of the 1:2 soil-water extract from each soil layer was determined with the Solu-Bridge Tester RD 15 Model.

The exchangeable cations of each soil layer were extracted by leaching with $1N N_{A}O$ Ac. Upon washing out the excess salts in each soil-layer sample with electrolytefree isopropyl alcohol, the cation exchange capacity (CEC) of each soil layer was determined by leaching each sample with 1.0N NaCl, distilling the NH $_A^+$ in the NaCl leachate into ²% boric acid solution and titrating it against 0.01 ^N potassium biiodate, KH(IO₃)₂. Exchangeable Ca, Mg, K, Na, and Mn were determined by atomic absorption spectrophotometry.

Exchangeable Al was displaced by 1 N KCl and determined by the aluminon method (55).

The 1/3-bar moisture percentage of each soil layer was determined by the pressure membrane method (143).

The texture of each soil layer was determined by the modified hydrometer method (17) followed by reading the textural triangle.

The incipient chemical and physical analyses of these topsoils and subsoils are presented in Table 1.

Preliminary Subsoil Studies on Six Crop Species

Six test crop species, namely, pea (Pisum sativum, L., cv. Frosty), cucumber (Cucumis sativus, L., cv. Challenger), romaine lettuce (Lactuca sativa var. longifolia, Lam,, cv, Parris Island), spinach (Spinacia oleracea var, inermis, Peterm., cv. America), pepper (Capsicum frutescens var. grossum, Bailey, cv. Pennbell), and tomato (Lycopersicon esculentum var. commune, Bailey, cv. Moreton Hybrid), were used in these studies. The plants were grown in all instances in ^a growth chamber under a 14-hour light period and 10-hour dark period at a continuous temperature of 25^{\pm} 1° C. Light was provided by Sylvania Cool-White fluorescent tubes giving an intensity of 1100 ft-c at the level of plant tops. Relative humidity was 60% \pm 5% as measured continuously by a hygroscope.

(a) Primary Root Growth Studies in Acid Subsoils at Two Lime Levels

Each bulk lot of the dried and screened Narragansett loam and Merrimac sandy loam surface soils was steam sterilized, treated with adequate $CaCO_3$ to bring the final pH upon incubation to 6.5, and mixed with a rotary blender for an hour. The soil was then wetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated moist for ⁵ days in polyethylene bags. The moist surface soil was then spread in a thin layer, air—dried, rewetted to the 1/3-bar moisture percentage with demineralized

water, mixed by hand, and incubated moist for another 5-day period as before. Completion of reaction between the fine CaCO₃ and the surface soils was greatly facilitated by this procedure. The moist soil was then again spread in a thin layer and air-dried. The fertilizer treatments were then imposed on the air-dried, lime-treated surface soil. Solid NH_4 NO₃, KH₂PO₄, and (NH₄)H₂PO₄ were added to the soil to give ⁵⁰ ppm N, 100 ppm P, and 50 ppm K. The surface soil was then blended for ³⁰ minutes, wetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated overnight. Finally, the moist, treated surface soil was spread in ^a thin layer, air-dried, and stored.

Each bulk lot of the dried and screened Narragansett loam and Merrimac sandy loam subsoils was treated with adequate CaCO₃ to bring the final pH upon incubation to about 6.0, and mixed with a rotary blender for an hour. The sub soil was then wetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated moist for ⁵ days in polyethylene bags. The moist subsoil was then spread in a thin layer, air-dried, rewetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated moist for another 5-day period as before. The moist subsoil was then again spread in ^a thin layer, airdried, and stored.

^A vertical split-root technique similar to the one described by Muzik and Whitworth (125), for growing seedlings

in glass-front boxes tilted to form a 15° angle from the vertical, was used to measure the effects of treated subsoils on subsoil primary root growth. This is shown in Figure 1. This technique permits the growth of roots along the glass front and facilitates measurements without disturbing the roots or soils. 2,000-g samples of treated and untreated subsoils that had been moistened to the 1/3-bar moisture percentage with demineralized water were placed in compartments of glass-front boxes, gently settled to a depth of about 18 cm, and covered with 650 ^g of moist, treated surface soil to give ^a surface soil depth of about ³ inches. Ten seedlings of each of the six test crop species were selected for uniformity. Seedlings were then transplanted onto the moist, treated surface soil about ⁴ cm above the subsoil. Each box was then weighed and recorded. The depth to which each primary root extended into the treated and untreated subsoils, as observed through the glass front, was traced with India ink, marked, and measured daily for a period of ¹⁰ days after the roots entered the subsoils. Each box was brought to its initial recorded weight with demineralized water once daily.

^A randomized, complete-block design involving ² subsoil treatments and ⁴ replications was used for this experiment. The results of this experiment are presented in Table 2.

Figure 1.—Glass-front box used in vertical splitroot technique for the periodic observation of primary roots in situ.

Figure 2.—Arrangement of 20-liter plastic cans used for preliminary primary root growth studies on six crop species in culture solutions.

(b) Primary Root Growth Studies in Acid Subsoils at Two Lime-Plus-Boron Levels

Each bulk lot of the dried, screened, and limed Narragansett loam and Merrimac sandy loam subsoils was treated with 0.5 ppm ^B as boric acid, blended for an hour, wetted to the 1/3-bar moisture percentage, mixed by hand, and incubated moist overnight in plastic bags. The moist subsoil was then spread in a thin layer, air-dried, and stored.

The vertical split-root technique aforementioned was used. 2,000-g samples of treated and untreated subsoils that had been moistened to the 1/3-bar moisture percentage with demineralized water were placed in compartments of glassfront boxes. They were then covered with 650 ^g of moist, treated surface soil and the aforementioned test crop species . were studied as previously described.

A randomized, complete-block design involving 2 subsoil treatments and 4 replications was used for this experiment. The results of this experiment are presented in Table 3.

Preliminary Culture Solution Studies On Six Crop Species

(a) Primary Root Growth in Culture Solution as Influenced by Aluminum Ion Concentration

Seeds of the ⁶ aforementioned test crop species were germinated in petri dishes and then transferred to plastic

gauze above 1-liter plastic cups filled with 1/5-strength Steinberg solution. Two-week-old seedlings, selected for uniformity in lengths of labeled primary roots were then transferred to 20-liter plastic cans. These cans had been sprayed on the outside with black enamel paint and covered with perforated, black-sprayed, Plexiglass sheets. The seedlings were supported on perforated, black, plastic sheets wrapped around the bottoms of plastic cylinders and held in place with rubber bands. This is shown in Figure 2. In all instances, solutions were aerated and pH was adjusted to desired values twice daily for ² days before transfer of seedlings to the 20-liter cans in order to help stabilize the pH of the solution medium.

The aerated 1/5-strength Steinberg solution was prepared from the following stock solutions: 300 g of $Ca(NO₃)₂$. 4 H₂O, 70 g of Mg(NO₃)₂.6H₂^O, 19 g of NH₄NO₃, 75 g of K₂HPO₄, 17.6 g of (NH_A) ₂SO₄, 23 g of K₂SO₄, and 58 g of KNO₃ per liter respectively. The micronutrient stock solution consisted of 2.34 g of MnCl₂.4H₂O, 2.04 g of H₃BO₃, 0.88 g of $ZnSO_4$.7H₂O, 0.20 g of CuSO₄.5H₂O, and 0.126 g of Na₂MoO₄.2H₂O per liter. The solution in which plants were grown contained 20 ml each of the Ca(NO₃)₂.4H₂O, Mg(NO₃)₂.6H₂O, and NH₄NO₃ stock solutions, 4.5 ml each of the K_2 HPO₄ and (NH_4) ₂SO₄ stock solutions, 13.3 ml each of the K_2SO_4 and KNO_3 stock solutions, and ⁴ ml of the micronutrient stock solution. The final concentrations of elements in ppm were: 50.8 Ca,

6.6 Mg, 56 N (51.9 as NO₃⁻ and 4.1 as NH₄⁺), 3.8 S (as SO₄²⁻), 29.4 K, 0.01 Na, ³ P, 0.34 Cl, 0.13 Mn, 0.07 B, 0.04 Zn, 0.01 Cu, and 0.005 Mo. The macronutrients were thus supplied in meq/l, as follows: Ca^{++} , 2.53; Mg⁺⁺, 0.54; K⁺, 0.75; N, 4.00 (3.70 as NO_3 ⁻ and 0.30 as NH_4^+); SO_4^2 ⁻, 0.24; and HPO₄²⁻, 0.19. Iron was added separately at ¹ ppm Fe (half as FeEDTA and half as FeSO_4) from freshly prepared solution (49). Aluminum was supplied as $\text{Al}_2(SO_4)_{3}$.18H₂O. The pH was maintained at 4.8 \pm 0.1 by adjusting the pH to 4.8 twice daily with 0.1 N NaOH or 0.1 N $H₂SO₄$ as necessary. After a growth period of 11 days, the primary root lengths were again recorded as labeled.

^A split-plot design, with aluminum ion concentration as whole plot and crop species as subplots, with 2 replications was used for this experiment. The results of this experiment are presented in Table 4.

(b) Primary Root Growth in Culture Solution as Influenced by Hydrogen Ion Concentration

Two-week-old seedlings of the ⁶ aforementioned test crop species, selected for uniformity in lengths of labeled primary roots, were transferred to 20-liter plastic cans of aerated 1/5-strength Steinberg solution as previously described. The pH values were maintained at 4.4 ± 0.05 , 4.8 \pm 0.1, 5.2 \pm 0.1, 5.6 \pm 0.1, and 6.0 \pm 0.15 by adjusting the pH to the desired values twice daily with 0.1N NaOH or

 0.1 N H₂SO₄ as necessary. After a growth period of 11 days, the primary root lengths were again recorded as labeled.

^A split-plot design, with hydrogen ion concentration as whole plot and crop species as subplots, with 2 replications was used for this experiment. The results of this experiment are presented in Table 5.

(c) Primary Root Growth in Culture Solutions as Influenced Calcium Ion Concentration

Two-week-old seedlings of the ⁶ aforementioned test crop species, selected for uniformity in lengths of labeled primary roots, were transferred to 20-liter plastic cans of aerated 1/5-strength Steinberg solution as previously described, with the exception that Ca was supplied as CaSO₄.2H₂O and nitrate was supplied as Mg(NO₃) ₂.6H₂O and KNO₃. The pH was maintained at 4.8 \pm 0.1 by adjusting the pH to 4.8 twice daily with 0.1N NaOH or 0.1N H^2 SO₄ as necessary. After a growth period of 12 days, the primary root lengths were again recorded as labeled.

^A split-plot design, with calcium ion concentration as whole plot and crop species as subplots, with 2 replications was used for this experiment. The results of this experiment are presented in Tables ⁶ and 7.

Subsoil Studies on Two Selected Crop Species

On the basis of the preliminary primary root growth studies on the ⁶ aforementioned test crop species in the subsoil and in culture solutions, two crop species, namely romaine lettuce (Lactuca sativa var. lonqifolia, Lam., cv. Parris Island) and pepper (Capsicum frutescens var. grossum, Bailey, cv . Pennbell), were selected for further investigation having regard to their suggested calcium requirement, aluminum tolerance, and relative ease of husbandry under growth chamber conditions.

(a) Effect of Subsoil Applications of CaCO₂ and MgCO₂ on Subsoil Primary Root Growth

Each bulk lot of the dried and screened Narragansett loam and Merrimac sandy loam subsoils was subdivided and treated with different levels of $CaCO_{2}$ and $MgCO_{2}$. Level No. 1 $CaCO_3$ was the amount of $CaCO_3$ necessary to neutralize the KCl-extractable Al expressed in meq/100 ^g subsoil. Level No. 2 $CaCO_3$ was twice the amount of $CaCO_3$ necessary to neutralize the KCl-extractable Al expressed in meq/100 ^g subsoil. Treatments involving 0% , 40% , and 100% of MgCO₃ equivalent to level No. 1 CaCO₃ were used in fractional or complete substitution of Mg for Ca. Each treated subsoil was mixed with a rotary blender for an hour. It was then wetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated moist for ⁵ days in

polyethylene bags. The moist subsoil was then spread in a thin layer, air-dried, rewetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated moist for another 5-dav period as before. Finally, the moist subsoil was then spread in a thin layer, air-dried, and stored.

The vertical split-root technique aforementioned was used. 2,000-g samples of treated and untreated subsoils that had been moistened to the 1/3-bar moisture percentage with demineralized water were placed in compartments of glass-front boxes and covered with 650 ^g of moist, treated surface soil. Lettuce and pepper primary root growth in the subsoil was then studied as previously described.

^A randomized, complete-block design involving ⁶ subsoil treatments and ² replications was used for this experiment.

At the end of this experiment, a 2,000-g sample of each treated and untreated subsoil was taken for displaced subsoil solution studies. ^A 1,000-g subsample was then wetted to the 1/3-bar moisture percentage with demineralized water, mixed by hand, and incubated for ⁵ days in polyethylene bags. The moist subsoil was tnen placed in a specially constructed Plexiglass column. This is shown in Figure 3. The column, 11 cm in diameter, has a perforated Plexiglass plate at the bottom. ^A small piece of glass wool was placed at the bottom of the column,

wetted with demineralized water, and then dried in place. The moist subsoil was then added in small increments while using the glass rod and rubber stopper, in piston-like combination, to thoroughly pack the column. The subsoil solution was then displaced following Jackson's procedure (79) by eluting the subsoil in successive 10-ml portions with 500 ml of 0.5% potassium thiocyanate solution. The elution process was stopped when the thiocyanate ion could be detected in the eluate by testing a few drops on a spot plate with 4% FeCl₃ solution. The eluate was then centrifuged at 33,000 ^X ^G for ³⁰ minutes to remove any particulate matter, particularly those responsible for Brownian movement, and ¹ drop of concentrated HC1 was added to preserve the highly charged aluminum ions for subsequent analyses.

Displaced subsoil solution concentrations of Ca, Mg, K, Na, and Mn were determined by atomic absorption spectrophotometry. Displaced subsoil solution concentration of Al was determined by the aluminon method (35). The final subsoil pH was determined with the glass electrode pH meter.

The results of this experiment are presented in Tables 8 and 9.

Figure 3.—Specially constructed Plexiglass column for displacing subsoil solutions for subsequent chemical analyses.

Figure 4.—Arrangement of 20-liter cans and cut-andinverted 1-liter yogurt cups used for primary root growth studies on selected crop species in controlled subsurface nutrient solutions.

(b) Effect of Subsoil Applications of CaCO₂ and MgCO₂ on Primary Root Growth in Subsoil Leached with 1.0 N AlCl₃ Solution

Each bulk lot of the dried and screened Narragansett loam and Merrimac sandy loam subsoils was slowly leached with 2 liters of 1.0 N AlCl₃ solution per 2,000-g sample. The sample was then leached with demineralized water until excess AlCl₃ removal was complete, as shown by a negative 0.1 N AgNO₂ test for chloride. Each bulk lot was then air dried and blended for an hour. It was then subdivided and treated with different levels of $CaCO₃$ and MgCO₃ using the same reasoning as in the preceding soil experiment. The treated subsoils were then subjected to two wetting—incubation—drying cycles as previously described.

The vertical split-root technique aforementioned was used. 2,000-g samples of treated and untreated subsoils that had been wetted to the 1/3-bar moisture percentage with demineralized water were placed in compartments of glass-front boxes and covered with ⁶⁵⁰ ^g of moist, treated surface soil. Lettuce and pepper primary root growth in the subsoil was then studied as previously described.

^A randomized, complete—block design involving ⁶ subsoil treatments and ² replications was used for this experiment.

At the end of this experiment, final subsoil pH and displaced subsoil solution studies were undertaken as described in the preceding soil experiment.

The results of this experiment are presented in Tables 10 and 11.

Subsurface Solution Studies on Two Selected Crop Species

The technique employed in these experiments is similar to that developed by Rios and Pearson (144). This is shown in Figure 4. This permitted ^a vertically changing chemical environment and allowed the isolation of the lower roots in controlled media placed directly beneath a layer of adequately limed and fertilized surface soil. Thus, the effect of the nutrient solutions was reflected in the changes r in root elongation by this system and therefore not confounded by differences in top growth. The specific effects of nutrient or toxic ions on primary root behavior in a lower rooting medium simulating the subsoil could thus be easily studied.

(a) Subsurface Primary Root Growth as Influenced by Hydrogen
Ton Concentration in Subsurface Nutrient Medium Ion Concentration in Subsurface Nutrient

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water, as previously described, was placed in ^a soil container made from an inverted 1-liter plastic yogurt cup the lid of which had been perforated and sealed over with 1:2 paraffin-petrolatum membrane. The membrane was made from paraffin wax and clear Vaseline petroleum

jelly. Seeds of lettuce or pepper were then planted in each cup. The cup was then placed over aerated 1/5-strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath. This is shown in Figure 5. Primary roots penetrated the membrane into the solution below. Each liter cup of solution was filled to the top once a day, and fresh 1/5-strength Steinberg solution was supplied every three days. The moisture content of the surface soil was replenished every 24 hours. The soil containers were maintained in this fashion for ¹⁴ days before differential treatments were imposed, at which time the primary roots were 5.5 to 8.0 cm long.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to 20-liter plastic cans—sprayed black on the outside and covered with perforated, blackened Plexiglass sheets—of aerated 1/5-strength Steinberg solution with variable pH values. The solution composition was as previously described. The pH values were maintained at 3.8 = 0.01, 4.0 = 0.01, 4.2 \pm 0.05, 4.5 \pm 0.1, and 4.8 \pm 0.1 by adjusting the pH to desired values twice daily with 0.1 N NaOH or 0.1 N H^1_2 SO₄ as necessary. After growth periods of 48 and 72 hours respectively, the primary root lengths were again recorded as labeled.

A split-plot design, with subsurface nutrient solution pH as whole plot and crop species as subplots, with 4 replications was used for this experiment. The results of

Figure 5.--Arrangement of cut-and-inverted 1-liter yogurt cups used for preparatory primary root growth of selected crop species atop 1-liter cups of aerated 1/5 strength Steinberg solution.

this experiment are presented in Table 12.

(b) Subsurface Primary Root Growth as Influenced by Aluminum Ion Concentration in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup, as previously described. It was then seeded with lettuce or pepper, placed over aerated 1/5-strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated 1/5-strength Steinberg solution with variable concentrations of $\text{Al}_2(SO_4)$ ₃. $18H₂$ O. The solution composition was as previously described. The pH was maintained at 4.8 ± 0.1 by adjusting the pH to 4.8 twice daily with 0.1 N NaOH or 0.1 N H_2SO_4 as necessary. After growth periods of ⁴⁸ and ⁷² hours respectively, the primary root lengths were again recorded as labeled.

A split-plot design, with subsurface Al ion concentration as whole plot and crop species as subplots, with ³ replications was used for this experiment. The results of this experiment are presented in Table 13.
(c) Subsurface Primary Root Growth as Influenced by Calcium Ion Concentration in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in ^a soil container made from ^a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5 strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter cans of aerated, modified 1/5-strength Steinberg solution with variable concentrations of $CaSO_4$. $2H_2O$, in which nitrate was supplied as $Mg(NO_3)_{2}$.6H₂O and KNO_3 . The pH was maintained at 4.8 ± 0.1 by adjusting the pH to 4.8 twice daily with 0.1 N NaOH or 0.1 N H₂SO₄ as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

A split-plot design, with subsurface Ca ion concentration as whole plot and crop species as subplots, with ³ replications was used for the first two sets of trials of this experiment. ^A randomized, complete-block design with ² replications was used for third set of trials of this experiment. The results of this experiment are presented in Table 14.

Subsurface Solution Studies on Two Selected Crop Species Involving Cation Interactions

(a) Subsurface Primary Root Growth as Influenced by Calcium and Hydrogen Ion Concentrations in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans (i) of aerated, modified 1/5-strength Steinberg solution, with variable concentrations of CaSO₄.2H₂O in which nitrate was supplied as Mg(NO₃)₂.6H₂O and KNO₃, and (ii) of aerated solutions consisting of variable concentrations of $CaSO_4$. $2H_2O$ and of 2 ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH values were maintained at 4.2 ± 0.01 , 4.4 \pm 0.05, 4.5 \pm 0.05, 4.6 \pm 0.1, and 4.8 \pm 0.1 by adjusting the pH to desired values twice daily with 0.1 ^N NaOH or 0.1 N H₂SO₄ as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

^A randomized, complete-block design involving a ³ ^X ³ factorial treatment combination with ³ replications was used for this experiment. The results of this experiment are presented in Table 15.

(b) Subsurface Primary Root Growth as Influenced by Calcium, Magnesium, and Hydrogen Ion Concentrations in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5-strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated solutions consisting of variable concentrations of $CaSO_4$. 2H₂O and MgSO₄.7H₂O and of 2 ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH values were maintained at 4.5 ± 0.1 , 4.8 ± 0.1 , 5.4 ± 0.15 , and 6.0 [±] 0.15 by adjusting the pH to desired values twice daily with 0.1 N NaOH or 0.1 N H_2SO_4 as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

^A randomized, complete-block design involving a ³ ^X ³ factorial treatment combination with ² replications was used for this experiment. The results of this experiment are presented in Tables 16, 17, and 18.

(c) Subsurface Primary Root Growth as Influenced by Differen tial Liminq-Plus-Fertilization of Surface Soil and by Calcium and Magnesium Ion Concentrations in Subsurface Nutrient Medium

As previously described, the original, dried and screened Merrimac surface soil was treated with (i) solid NH_4NO_3 , KH_2PO_4 , and $(NH_4)H_2PO_4$ to give 50 ppm N, 100 ppm P, and 50 ppm K, and (ii) solid CaCO₃ to give 1,750 ppm CaCO₃ and to bring the final pH upon two wetting-incubation-drying cycles to 6.5. Another bulk lot of the original, dried and screened Merrimac surface soil was treated with (i) double the above fertilizer rates to give ¹⁰⁰ ppm N, ²⁰⁰ ppm P, and 100 ppm K, (ii) solid $CaCO₃$ to give 1,750 ppm $CaCO₃$ and to bring the final pH upon two wetting-incubation-drying cycles to 6.5 , and (iii) additional MgCO₃ equivalent to this above amount of $CaCO₃$.

²⁵⁰ ^g of each of these differentially treated Merrimac surface soils which had been wetted to the 1/3—bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5-strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated solutions consisting of variable concentrations of $CaSO_4$. $2H_2O$ and $MgSO_A$. $7H_2O$ and 2 ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH values were maintained at 6.0 ± 0.1 by adjusting the pH to 6.0 twice daily with 0.1 N NaOH or 0.1 N H^2 SO₄ as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

^A randomized, complete-block design involving a 3X3 factorial treatment combination was used for this experiment. The results of this experiment are presented in Tables 16 and 19.

At the end of each set of trials within this experiment, the young succulent topgrowth was harvested, washed twice with demineralized water, and dried at 70°C in a forced draft oven for ²⁴ hours. The dried plant sample was weighed into a 110-ml Kjeldahl flask and completely moistened with 5 ml of concentrated HNO₃ in order to control the reaction intensity and eliminate the possibility of explosion. Grinding prior to digestion was unnecessary on account of succulence and size of sample obtained. Five ml of concentrated $HClO_4$ was then added. One ml of demineralized water was used to rinse down the neck of the flask. The sample was then ashed on the digestion rack with condenser attachment under a hood.

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After thorough digestion the sample was cooled for an hour. While still warm, demineralized water was added to volume in order to avoid any crystalline precipitation. The sample was left to set overnight. Portions of the sample solution were taken for subsequent analyses.

Calcium, magnesium, and potassium contents of topgrowth were determined by atomic absorption spectrophotometry. Phosphorus content of topgrowth was determined by the molydovanadophosphoric acid method, the absorbance being measured spectrophotometrically at 470 mu. The results of topgrowth analyses are presented in Tables 20 and 21.

(d) Subsurface Primary Root Growth as Influenced by Calcium and Potassium Ion Concentrations in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5 strength Steinberg solution in ^a 1-liter yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated solutions consisting of variable concentrations of $CaSO^4$. 2H₂O and K₂SO₄ and ² ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH was maintained at 6.0 ± 0.1 by adjusting the pH to 6.0 twice

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daily with 0.1 N NaOH or 0.1 N H^2 SO₄ as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

^A randomized, complete-block design involving a ³ ^X ³ factorial treatment combination with ² replications was used for this experiment. The results of this experiment are presented in Tables 22 and 23.

(e) Subsurface Primary Root Growth as Influenced by Calcium and Aluminum Ion Concentrations in Subsurface Nutrient Medium

250 ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup. It was then seeded with lettuce or pepper, placed over aerated 1/5 strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated solutions consisting of variable concentrations of $CaSO^4$. 2H₂O and $\text{A1}_2(\text{SO}_4)$ ₃.18H₂O, a fixed concentration of MgSO₄.7H₂O, and ² ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH was maintained at 4.8 ± 0.1 by adjusting the pH to 4.8 twice daily with 0.1 N NaOH or 0.1 N H₂SO₄ as necessary. After a growth

period of 72 hours, the primary root lengths were again recorded as labeled,

^A randomized, complete-block design involving a 2X5 factorial treatment combination with ² replications was used for this experiment. The results of this experiment are presented in Tables 24 and 25,

(f) Subsurface Primary Root Growth as Influenced by Calcium and Phosphate Ion Concentrations in Subsurface Nutrient Medium

²⁵⁰ ^g of the regular limed and fertilized Merrimac surface soil which had been wetted to the 1/3-bar moisture percentage with demineralized water was placed in a soil container made from ^a 1-liter plastic yogurt cup. It was seeded with pepper, placed over aerated 1/5-strength Steinberg solution in ^a 1-liter plastic yogurt cup beneath, and maintained for a 14-day period as previously described.

Cups of 2-week-old seedlings, selected for uniformity in lengths of labeled primary roots, were then transferred to black-painted, 20-liter plastic cans of aerated solutions consisting of variable concentrations of $CaSO_4$. $2H_2O$ and $Ca(H_2PO_A)_{2}H_2O$ and of 2 ml of the micronutrient stock solution equivalent to that in 1/10-strength Steinberg solution. The pH was maintained at 6.0 \pm 0.15 twice daily with 0.1 N NaOH or 0.1 N H₂SO₄ as necessary. After a growth period of 72 hours, the primary root lengths were again recorded as labeled.

^A randomized, complete—block design involving a 3X3 factorial treatment combination with ² replications was used for this experiment. The results of this experiment are presented in Table 26.

R E S U L T S

The results of laboratory analyses conducted on the Narragansett and Merrimac soil types are presented in Table 1. Analyses were conducted with a view to characterizing their relevant chemical and physical properties.

The subsoils, which had a pH less than 5.3, were less acid than their corresponding topsoils.

The lime requirement and per cent organic matter content decreased with depth.

The soils were clearly low in salt content and cation exchange capacity, the major exchangeable cation being Ca. All exchangeable cations and KCl-extractable Al decreased with depth, except exchangeable Na which increased with depth in the Narragansett soil type.

Two textural classes were represented. The texture of the Narragansett sample was a loam while that of the Merrimac sample was a sandy loam. As expected, the finer textured soil had the higher 1/3-bar moisture percentage.

Preliminary Subsoil Studies on Six Crop Species

The results of primary root growth studies on the two acid subsoils at two lime levels are presented in Table 2. The studies indicate that response to added lime differed with different subsoils and with different crop

SOME CHEMICAL AND PHYSICAL ANALYSES OF SOILS USED

TABLE 2 PRIMARY ROOT GROWTH IN ACID SUBSOILS AT TWO LIME LEVELS

+ Average of two separate trials.

++ Growth increase expressed as a percentage of the growth with no lime.

* Different at the 5% level of significance.

** Different at the 1% level of significance.

species, as follows:

- (a) The primary root growth of pea was not significantly altered by liming either subsoil.
- (b) The primary root growth of cucumber increased significantly after ⁵ days in the limed Merrimac sandy loam subsoil, whereas it consistently decreased significantly in the limed Narragansett loam subsoil.
- (c) The primary root growth of lettuce increased sig nificantly for the first 5 days in the limed Merrimac sandy loam subsoil, whereas it consistently increased highly significantly in the limed Narragansett loam subsoil.
- (d) The primary root growth of spinach increased significantly only for the first ³ days in the limed Merrimac sandy loam subsoil, whereas it increased significantly only by the tenth day in the limed Narragansett loam subsoil.
- (e) The primary root growth of pepper consistently decreased but not significantly in the limed Merrimac sandy loam subsoil, whereas it consistently increased significantly only for the first ⁵ days in the limed Narragansett loam subsoil.
- (f) The primary root growth of tomato consistently decreased, and significantly only by the fifth day, in the limed Merrimac sandy loam subsoil, whereas it consistently decreased significantly after the

third day in the limed Narragansett loam subsoil.

The results of primary root growth studies on the two acid subsoils at two lime-plus—boron levels are presented in Table 3, as follows:

- (a) The primary root growth of pea was not significantly altered by the lime—plus—boron treatment of either subsoil.
- (b) The primary root growth of cucumber also was not significantly altered by the lime-plus-boron treatment of either subsoil.
- (c) The primary root growth of lettuce increased sig nificantly only by the tenth day as ^a result of the lime-plus-boron treatment of the Merrimac sandy loam subsoil, whereas it consistently increased highly significantly as ^a result of the lime-plusboron treatment of the Narragansett loam subsoil.
- (d) The primary root growth of spinach decreased consistently but significantly only after the third day as ^a result of the lime—plus-boron treatment of the Merrimac sandy loam subsoil, whereas it consistently decreased highly significantly as a result of the lime-plus-boron treatment of the Narragansett loam subsoil.
- (e) The primary root growth of pepper decreased consistently but significantly only by the tenth day as a result of the lime—plus—boron treatment of

TABLE 3 PRIMARY ROOT GROWTH IN ACID SUBSOILS AT TWO LIME-PLUS-BORON (0.5 ppm B) LEVELS

+ Average of two separate trials.

++ Growth increase expressed as a percentage of the growth with no lime.

* Different at the 5% level of significance.

** Different at the 1% level of significance.

the Merrimac sandy loam subsoil, whereas it was not significantly altered by the lime-plus-boron treatment of the Narragansett loam subsoil.

(f) The primary root growth of tomato was not significantly altered by the lime-plus-boron treatment of the Merrimac sandy loam subsoil, whereas it decreased consistently and significantly after the third day as a result of the lime-plus-boron treatment of the Narragansett loam subsoil.

^A comparison of the lime-plus-boron with the lime treatments indicates that the addition of boron to the lime treatment yielded slightly different results for both subsoils, as follows:

- (a) The addition of 0.5 ppm ^B did not significantly alter the nil response of pea roots to liming either subsoil.
- (b) The addition of 0.5 ppm ^B nullified the positive response of cucumber roots to liming the Merrimac sandy loam subsoil and the negative response of cucumber roots to liming the Narragansett loam subsoil.
- (c) The addition of 0.5 ppm ^B did not significantly alter the positive response of lettuce roots to liming either subsoil.
- (d) The addition of 0.5 ppm ^B not only nullified the positive response of spinach roots but also

significantly decreased spinach root growth in either subsoil, particularly the Narragansett loam subsoil.

- (e) The addition of 0.5 ppm ^B not only reduced the root growth of pepper to significant proportion by the tenth day in the limed Merrimac sandy loam subsoil but also nullified the positive response of pepper roots to liming the Narragansett loam subsoil.
- (f) The addition of 0.5 ppm ^B nullified the negative response of tomato roots to liming the Merrimac sandy loam subsoil but did not alter the response of tomato roots to liming the Narragansett loam subsoil.

Preliminary Culture Solution Studies On Six Crop Species

The results of primary root growth studies which indicate that the six crop species have different susceptibilities to Al ion concentration in 1/5-strength Steinberg solutions are presented in Table 4, as follows:

- (a) The primary root growth of pea was not significantly altered within ^a range of ⁰ to 2.5 ppm Al. It, however, increased significantly at the ⁴ ppm Al level.
- (b) The primary root growth of cucumber was not significantly altered within a range of ⁰ to 2.5 ppm Al.

PRIMARY ROOT GROWTH AS INFLUENCED BY ALUMINUM ION CONCENTRATION

++Roots lengths followed by the same letter are not significantly different at the 0.05 probability level, separately for each crop species.

Brownish or dead root tips.

**Dead root tips.

Average of ² replications in essentially 1/5—Steinberg culture solutions over an 11-day period; pH adjusted to 4.8 twice daily.

It, however, increased highly significantly at the the ⁴ ppm level; the primary roots were spindly and brownish with markedly inhibited lateral root development.

- (c) The primary root growth of lettuce was significantly reduced at the 0.5 ppm Al level and above.
- (d) The primary root growth of spinach was significantly increased within a range of ⁰ to 0.5 ppm Al and thenceforth completely inhibited.
- (e) The primary root growth of pepper, like that of lettuce, was significantly reduced at the 0.5 ppm Al level and above.
- (f) The primary root growth of tomato was significantly reduced at the 2.5 ppm Al level and above.

The results of primary root growth studies which indicate that the six crop species have different susceptibilities to hydrogen ion concentration in 1/5-strength Steinberg solutions are presented in Table 5, as follows:

- (a) The primary root growth of pea, cucumber, and tomato was not significantly altered within a pH range of 4.4 to 6.0.
- (b) The primary root growth of lettuce, spinach, and pepper was significantly reduced only at a pH below 4.8.

The results of primary root growth studies which indicate that the six crop species have different requirements

PRIMARY ROOT GROWTH AS INFLUENCED BY HYDROGEN ION CONCENTRATION

** Root lengths followed by the same letter are not significantly different at the 0.05 probability level, separately for each crop species.

****** Brownish or dead root tips.

+Average of ² replications in essentially 1/5- Steinberg culture solutions, over an 11-day period; pH adjusted twice daily.

++Average of ² replications in essentially 1/5- Steinberg culture solutions, over a 12—day period; pH adjusted twice daily.

for Ca in 1/5-strength Steinberg solutions are presented in Tables ⁶ and 7, as follows:

- (a) The primary root growth of pea increased with increasing Ca ion concentration up to about the 12 ppm Ca level,
- (b) The primary root growth of cucumber increased witn increasing Ca ion concentration up to the ⁴ ppm Ca level.
- (c) The primary root growth of lettuce did not significantly increase beyond the 0.6 ppm Ca level.
- (d) The primary root growth of spinach increased with increasing Ca ion concentration up to the maximum 36 ppm Ca level studied.
- (e) The primary root growth of pepper increased with increasing Ca ion concentration up to the ⁶ ppm Ca level•
- (f) The primary root growth of tomato increased with increasing Ca ion concentration up to the 12 ppm Ca level.

Subsoil Studies on Two Selected Crop Species The results of further studies of the rates of primary root growth of lettuce and pepper in the Narragansett loam and Merrimac sandy loam subsoils are presented in Tables 8 and 9. Application of different rates of CaCO₃ and/ or MgCO₃ to the two subsoils resulted in different subsoil

PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM ION CONCENTRATION (a)

Root lengths followed by the same letter are not ROOL Tengths forfowed by the same recter are : separately for each crop species.

Brownish or dead root tips.

+Average of ² replications in modified 1/5—Steinberg culture solutions, with Ca supplied via $CaSO_4$. $2H_2O$ and culture solutions, with Ca supplied via CaSO_4 . Englands in the supplied via Mg(NO₃)₂ and KNO₃, over a 12-day period; nitrace suppried vid ng (nog)?

PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM ION CONCENTRATION (b)

Root lengths followed by the same letter are not ROOL Tengths forfowed by the same fector are in separately for each crop species,

+Average of ² replications in modified 1/5—Steinberg average of 2 reprications in moderated by culture solutions, with Ca supplied via CaSO₄.2H₂O and culture solutions, with ta supprise via ease A^{2} and A^{2} and A^{2} and A^{2} over a l2-day period; pH adjusted to 4.8 twice daily.

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TABLE 8 TABLE 8 EFFECT OF GCO3 AND MgCO3 ADDITIONS ON PRIMARY ROOT GROWTH OF CROP SPECIES, $\sum_{i=1}^{n}$ and an original matrix of calculations of crop species, $\sum_{i=1}^{n}$ and $\sum_{i=1}^{n}$ SUBSOIL SOLUTION COMPOSITION, AND SUBSOIL _PH OF NARRAGANSETT LOAM

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EFFECT OF GaCO3 AND MgCO3 ADDITIONS ON PRIMARY ROOT GROWTH OF CROP SPECIES, EFFECT OF CaC03 AND MgC03 ADDITIONS ON PRIMARY ROOT GROWTH OF CROP SPECIES, SUBSOIL SOLUTION COMPOSITION, AND SUBSOIL PH OF MERRIMAC SANDY LOAM SUBSOIL SOLUTION COMPOSITION, AND SUBSOIL pH OF MERRIMAC SANDY LOAM

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Note: Level No.1 CaCO3 is amount necessary to neutralize the KCI-extractable AI in me./100g subsoil; level No.2 CaCO3
is twice the amount of level No.1 CaCO3; 40 and 100% of MgCO3 equivalent to level No.1 CaCO3 are used in is twice the amount of level No.l CaC03; 40 and 100% of MgC03 equivalent to level No.l CaC03 are used in treat¬ Note: Level No.1 CaCO3 is amount necessary to neutralize the KCI-extractable Al in me./100 g subsoil; level No. 2 CaCO₃ ments involving fractional or complete substitution of Mg for Ca. ments involving fractional or complete substitution of Mg for Ca. 78

solution compositions and final subsoil pH's as well as different primary root growth rates for lettuce and pepper.

Addition of $CaCO₃$ or combinations of $CaCO₃$ and MgCO₃ to the unlimed Narragansett loam subsoil did not significantly increase the primary root growth of lettuce, whereas addition of $MgCO₃$ alone to the unlimed subsoil significantly increased it. The results indicate that while Narragansett loam subsoil was not naturally deficient in Ca for lettuce primary root growth, lettuce primary root growth in it would respond significantly to added MgCO₃ alone.

On the other hand, addition of $CaCO_{3}$, MgCO₃, or combinations of both CaCO₃ to the unlimed Narragansett loam subsoil resulted in a significant increase in primary root growth of pepper particularly after the third day. Pepper root growth positively responded equally as well to liming with MgCO₃ alone as to liming with CaCO₃ alone or with combinations of both $CaCO_{3}$ and MgCO₃. The results indicate, therefore, that the Narragansett loam subsoil was not naturally deficient in Ca for pepper primary root growth and that the observed positive response of pepper primary root growth was probably a result of neutralization of subsoil acidity.

Addition of CaCO₃, MgCO₃, or combinations of both CaCO₃ and MgCO₃ to the unlimed Merrimac sandy loam subsoil did not significantly increase the primary root growth of lettuce. However, the addition of combinations of both

CaCO₃ and MgCO₃ yielded the best rates, whereas addition of twice the amount of CaCO₃ needed to neutralize the KClextractable Al in the unlimed subsoil yielded the poorest rate by the seventh day. The results indicate that the Merrimac sandy loam subsoil was not naturally deficient in Ca for lettuce primary root growth and point up the possibility of injury due to excess $CaCO_{3}$.

on the other hand, addition of CaCO₃, MgCO₃, or combinations of both $CaCO₃$ and MgCO₃ to the unlimed Merrimac sandy loam subsoil did not result in a significant increase in primary root growth of pepper, except in one instance. The combination of 0.70 meq $CaCO_{3}$ and 0.18 meq $MgCO_{3}$ per 100 ^g of subsoil significantly increased the primary root growth of pepper above that in the unlimed subsoil. However, addition of an equivalent amount of $CaCO₃$ alone (0.88 meq/ 100 ^g subsoil) to the unlimed subsoil yielded the poorest rate by the fifth day. The results indicate that while the Merrimac sandy loam subsoil was not naturally deficient in Ca for pepper primary root growth, it would seem that pepper root growth would positively respond to a particular combination of both CaCO₃ and MgCO₃. The results also point up the possibility of injury due to excess $CaCO_{3}$.

The results of further subsoil studies on the two selected crop species are presented in Tables 10 and 11. After leaching the subsoils with AlCl₃ solution, addition of different rates of CaCO₃ and/or MgCO₃ resulted in different

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subsoil solution compositions and final subsoil pH's as well as different primary root growth rates for lettuce and pepper.

Addition of $CaCO₃$ or MgCO₃ sufficient to neutralize the KCl-extractable Al in the leached Narragansett loam subsoil did not result in ^a significant increase in the primary root growth of lettuce. Addition of twice the amount of CaCO₃ needed to neutralize the KCl-extractable Al (1.40 meq/ 100 ^g subsoil) gave the greatest increase in lettuce primary root growth, followed by addition of an equivalent combination of 0.70 meq CaCO₃ and 0.70 meq MgCO₃ per 100 g of subsoil. The results indicate that lettuce primary root growth would respond to added $CaCO₃$ or a particular combination of both CaCO₃ and MgCO₃ only when in excess of that required to neutralize the exchangeable Al present in the leached Narragansett loam subsoil.

On the other hand, addition of different levels of CaCO₃ to the leached Narragansett loam subsoil resulted in a significant increase in the primary root growth of pepper. Addition of combinations of both $CaCO_{3}$ and MgCO₃ equivalent to twice the amount of CaCO₃ needed to neutralize the KClextractable Al in the leached subsoil gave further significant increases in pepper primary root growth. Addition of $MgCO₃$ alone, however, did not significantly alter the primary root growth of pepper in the leached subsoil. The results indicate that pepper primary root growth would respond positively to added Ca alone, but not to added Mg alone, as a

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result of neutralization of the exchangeable Al in the leached Narragansett loam subsoil and yet more positively to further addition of MgCO₃ in combination with CaCO₃ after the exchangeable Al in the leached subsoil had been neutralized with $CaCO₃$.

Addition of sufficient $CaCO₃$ or MgCO₃ to neutralize the KCl-extractable Al in the leached Merrimac sandy loam subsoil and the combination of 0.32 meq CaCO₃ and 0.08 meq $MgCO₃$ per 100 g of subsoil resulted in a significant increase in the primary root growth of lettuce. However, addition of twice the amount of $CaCO₃$ required to neutralize the KClextractable Al in the leached subsoil resulted in a significant decrease in the primary root growth of lettuce. The data indicate that while lettuce primary root growth responded positively to neutralization of exchangeable Al in the leached Merrimac sandy loam subsoil, it was also susceptible to injury in this subsoil upon application of excess $CaCO_{3}$.

On the other hand, addition of varying levels of $CaCO₃$ or combinations of both CaCO₃ and MgCO₃ to the leached Merrimac sandy loam subsoil did not significantly alter the primary root growth of pepper even though combinations of both CaCO₃ and MgCO₃ yielded the best growth rates. In $control, addition of MgCO₃ alone resulted in a significant$ decrease in the primary root growth of pepper and actual death of the growing root tips after the third day.

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In general, upon treating the unlimed subsoils with increasing amounts of $CaCO₃$ and/or MgCO₂, the subsoil solution concentrations of Ca and/or Mg progressively increased accordingly while the subsoil solution concentrations of K, Na, Mn, A1, and ^H ions progressively decreased.

Subsurface Solution Studies on Two Selected Crop Species

The results of some primary root growth studies in subsurface 1/5-strength Steinberg solutions are presented in Tables 12, 13, and ¹⁴ and Figures 6, 7, and 8. The results indicate that lettuce and pepper have different susceptibilities to hydrogen and aluminum ion concentrations and different requirements for Ca.

The results presented in Table 12 and Figure 6 indicate that subsurface primary root growth of lettuce was significantly inhibited at some pH between 4.5 and 4.8, whereas that of pepper was even more markedly inhibited within this same pH range.

The data presented in Table ¹³ and Figure ⁷ indicate that subsurface primary root growth of lettuce and pepper was significantly inhibited at ^a concentration of 0.5 ppm Al. Pepper seemed to be slightly more susceptible within the narrow 0 to 0.5 ppm Al range than lettuce.

The data presented in Table 14 and Figure 8 show that near maximum subsurface primary root growth of lettuce

PRIMARY ROOT GROWTH AS INFLUENCED BY HYDROGEN ION CONCENTRATION IN SUBSURFACE NUTRIENT MEDIUM

Root lengths followed by the same letter are not ROOT Lengths TOITOWED by the same feeter are in separately for each period of observation for each crop species•

+Average of two replications, each treatment mean involving a maximum of 14 subsurface primary roots individually monitored in essentially 1/5-Steinberg culture solutions; pH adjusted twice daily.

PRIMARY ROOT GROWTH AS INFLUENCED BY ALUMINUM ION CONCENTRATION IN SUBSURFACE NUTRIENT MEDIUM

Root lengths followed by the same letter are not ROOL Tengths IOIIOWED by the Same Ieccer die significantly different at the order presenting force,

* Brownish and dead root tips.

+Average of two replications, each treatment mean involving a maximum of 14 subsurface roots individually monitored in essentially 1/5—Steinberg culture solutions; pH adjusted to 4.8 twice daily.

THREE-DAY PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM ION CONCENTRATION IN SUBSURFACE NUTRIENT MEDIUM

Root lengths followed by the same letter are not significantly different at the 0.05 probability level, separately for each trial for each crop species.

+Average of two replications, each treatment mean involving a maximum of 14 subsurface primary roots individually monitored in modified 1/5—Steinberg culture solutions, with Ca supplied via $CaSO_4$. $2H_2O$ and nitrate supplied via Mg(NO₃)₂ and KNO₃; pH adjusted to 4.8 twice daily.

++Separate trial for each crop species.

Figure 7.—Effect of subsurface solution aluminum ion concentration on primary root elongation of lettuce and pepper seedlings for a three-day period.

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Figure 8. — Effect of subsurface solution calcium ion concentration on primary root elongation of lettuce and pepper seedlings for a three-day period.

Figure 9.—Effect of subsurface solution calcium and hydrogen ion concentrations on primary root elongation of lettuce seedlings.

was obtained at a concentration of only ¹ ppm Ca, whereas about 72 ppm Ca was needed for near maximum subsurface primary root growth of pepper.

Subsurface Solution Studies on Two Selected Crop Species Involving Cation Interactions

The results of primary root growth studies on lettuce and pepper as influenced by calcium and hydrogen ion concentrations in subsurface $CaSO_A$ nutrient solutions are presented in Table 15 and Figures ⁹ and 10.

The results indicate that significant increases in primary root growth of lettuce were obtained upon increasing the Ca ion concentration from ⁶ to ³⁶ ppm at pH 4.5, upon progressively increasing the Ca ion concentration from ¹ to 36 ppm at pH 4.8, upon increasing the pH from 4.5 to 4.8 at the ⁶ ppm Ca level, and upon progressively increasing the pH from 4.2 to 4.8 at the 36 ppm Ca level in the subsurface nutrient medium. However, there was no significant interaction between Ca and ^H ions at the ¹ ppm Ca level.

On the other hand, the results also indicate that significant increases in primary root growth of pepper were obtained only upon increasing the Ca ion concentration from ³⁶ to 100 ppm at pH 4.8 in the subsurface nutrient medium. There were no significant interactions between Ca and ^H ions at the ²⁴ and ³⁶ ppm Ca levels.

THREE-DAY PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND HYDROGEN ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

A. LETTUCE⁺

B. PEPPER+

$C.$ PEPPER⁺⁺

+Average of ² replications, each treatment mean involving a maximum of ¹⁵ subsurface primary roots individually monitored in culture solutions consisting of CaSO₄. 2H₂O and 2 ml of the In curture solutions consisting or ease available of the curtum equivalent to $1/10$ Steinberg; pH

TABLE 15—Continued

adjusted twice daily.

++Average of two replications, each treatment mean involving a maximum of ¹⁵ subsurface primary roots individually monitored in modified 1/5-Steinberg culture solutions, with monitored in modified 1/5-Steinberg culture solutions, with
Ca supplied via CaSO₄.2H₂O and nitrate supplied via Mg(NO₃)₂ and KNO₃; pH adjusted twice daily.

*Root lengths followed by the same letter are not significantly different at the 0.05 probability level, separately for each experiment for each crop species.

Figure 11.--Effect of subsurface solution calcium and potassium ion concentrations on primary root elongation of lettuce seedlings.

In a similar experiment, pH and Ca concentration were varied in essentially 1/5-strength Steinberg solutions. The results indicate that significant increases in primary root growth of pepper were obtained upon increasing the Ca ion concentration from 24 to 100 ppm at pH's 4.4 and 4.6, upon progressively increasing the Ca ion concentration from 12 to 100 ppm at pH 4.8, upon increasing the pH from 4.6 to 4.8 at the ¹² and ²⁴ ppm Ca levels, and upon progressively increasing the pH from 4.4 to 4.8 at the 100 ppm Ca level. The rate of pepper primary root growth in the Steinberg solution was clearly much higher than that in the $CaSO_A$ nutrient solution at Ca ion concentrations of 12 and 24 ppm upon increasing the pH from 4.6 to 4.8 and at a Ca ion concentration of 100 ppm upon progressively increasing the pH from 4.4 to 4.8. This would suggest that there was some nutrient element or elements other than Ca stimulating the primary root growth of pepper in subsurface culture solution.

The results of primary root growth studies on lettuce and pepper as influenced by calcium, magnesium, and hydrogen ion concentrations in subsurface nutrient solutions consisting essentially of $CaSO_4$ and MgSO₄ are presented in Tables 16, 17, and 18.

The results in Table 16 indicate that, at pH 4.8, significant increases in primary root growth of lettuce were obtained upon increasing the Ca ion concentration from 18 to ³⁶ ppm at the ⁴ and 12 ppm Mg levels, upon increasing the Ca

THREE-DAY LETTUCE PRIMARY ROOT GROWTH AS INFLUENCED BY DIFFERENTIAL LIMING-PLUS-FERTILIZATION OF SURFACE SOIL AND BY CALCIUM, MAGNESIUM, AND HYDROGEN ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

A. Ca_l N_1 P_1 K_1 ⁺

(a) pH 4.8

(b) $pH 6.0$

ppm Ca^{**}

			18	36	100
		$/cm)^*$			
		2.51 _c	2.84 _c	4.03_{ab}	4.23_a
ppm Mg	12	4.03 _{ab}	3.77_h	4.10 _{ab}	
	36	4.20 _{ab}	4.64 _a	4.43	

B. Ca_1 Mg₁ N₂ P₂ K₂⁺⁺

(b) pH 6.0

Root lengths followed by the same letter are not significantly different at the 0.05 probability level. separately for each trial.

 $^+$ Surface soil treated with (a) solid NH_4NO_3 , KH₂PO₄, and $(NH_A)H_2PO_A$ to give 50 ppm N, 100 ppm P, and 50 ppm²K, and (b) solid CaC_3^3 to give 1,750 ppm CaCO₃ and to bring the final pH upon incubation to 6.5

++Surface soil treated with (a) solid NH₄NO₃, KH₂PO₄, and $(NH_4)H_2PO_4$ to give 100 ppm N, 200 ppm P, and 100 pp, K, (b) solid $CaCO₃$ to give 1,750 ppm CaCO₃ and to bring the final pH upon incubation to 6.5, and (c) additional MgCO₃ equivalent to this amount of $CaCO_{2}$.

* * Each treatment mean involves a maximum of 16 subsurface primary roots individually monitored in culture solutions consisting of CaSO_4 .2H₂O, MgSO₄.7H₂O, and 2 ml of solutions consisting of ease, $\frac{1}{2}$, $\frac{1}{2}$, $\frac{1}{2}$, $\frac{1}{2}$, $\frac{1}{2}$, $\frac{1}{2}$, $\frac{1}{2}$ and $\frac{1}{2}$ and $\frac{1}{2}$ and $\frac{1}{2}$ pH adjusted twice daily.

THREE-DAY PEPPER PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM, MAGNESIUM, AND HYDROGEN ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

A. pH 4.5

B. pH 4.8

 $C.$ pH 5.4

TABLE 17—Continued

D. pH 6.0

Root lengths followed by the same letter are not significantly different at the 0.05 probability level, separately for each trial.

+Each treatment mean involves a maximum of 16 sub surface primary roots individually monitored in separate trials in culture solutions consisting of $CaSO_4$. $2H_2O$, $MgSO_4$.7H₂O, and 2 ml of the micronutrient stock solution equivalent to $1/10$, Steinberg; pH adjusted twice daily.

THREE-DAY PEPPER PRIMARY ROOT GROWTH AS INFLUENCED BY DIFFERENTIAL LIMING-PLUS-FERTILIZATION OF SURFACE SOIL AND BY CALCIUM AND MAGNESIUM ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

ca_1 N_1 P_1 K_1 * 1

B.
$$
Ca_1
$$
 Mg₁ N₂ P₂ K₂^{**}

Surface soil treated with (a) solid and $(NH_4)H_2PO_4$ to give 50 ppm N, 100 ppm P, and (NH₄)H₂PO₄ to give 30 ppm N, 100 ppm 2, and to bring the final pH upon incubation to 6.5. NH_4 NO₃, KH₂PO₄, and 50 ppm K ,

* * Surface soil treated with (a) solid NH_4NO_3 , KH_2PO_4 , and $(NH_4)H_2PO_4$ to give 100 ppm N, 200 ppm P and 100 ppm K, and $(nn_4)n_2$ for give 100 ppm $(n-1)$ and to bring final pH upon incubation to 6.5, and (c) additional MgCO₃ equivalent to this amount of CaCO₃.

+Each treatment mean involves a maximum of 14 subsurface **Each treatment mean involves a maximum of In Suzed
primary roots individually monitored in culture solutions** primary roots individually monitored in curture below
consisting of CaSO₄.2H₂O, MgSO₄.7H₂O, and 2 ml of the

micronutrient stock solution equivalent to 1/10-Steinberg; pH adjusted to 6.0 twice daily.

++Root lengths followed by the same letter are not significantly different at the 0.05 probability level, significantly different at the 0.05 probability of the control of the separately for each surface soil treatment.

ion concentration from ⁶ to ¹⁸ ppm at the ³⁶ ppm Mg level, upon increasing the Mg ion concentration from ⁴ to 12 ppm at the 6 ppm Ca level, and upon increasing the Mg ion concentration from 12 to 36 ppm at the 18 ppm Ca level. At pH 6.0, significant increases in primary root growth of lettuce were obtained upon increasing the Ca ion concentration from 18 to ³⁶ ppm at the ⁴ ppm Mg level, upon increasing the Mg ion concentration from ⁴ to 12 ppm at the ⁶ ppm Ca level, and upon progressively increasing the Mg ion concentration from 4 to 36 ppm at the 18 ppm Ca level. At 36 ppm Ca, no significant interaction was observed between Ca and Mg at either pH 4.8 or 6.0. However, significant increases in subsurface primary root growth of lettuce, averaged over the different levels of Mg, were obtained at the ⁶ and ¹⁸ but not at the ³⁶ ppm Ca levels upon increasing the pH from 4.8 to 6.0.

On the other hand, the results in Table ¹⁷ indicate that, at pH 4.5, significant increases in primary root growth of pepper were obtained upon increasing the Ca ion concentration from 100 to 200 ppm at the ⁴ and ³⁶ ppm Mg levels, upon progressively increasing the Ca ion concentration from 36 to 200 ppm at the 100 ppm Mg level, and upon increasing the Mg ion concentration from ³⁶ to 100 ppm at the ¹⁰⁰ ppm Ca level. There were no significant interactions between Ca and Mg at the ³⁶ and ²⁰⁰ ppm Ca levels. At pH 4.8, significant increases in primary root growth of pepper were obtained upon increasing the Ca ion concentration from 100 to 200 ppm at the ⁴ ppm Mg

level, upon progressively increasing the Ca ion concentration from 36 to 200 ppm at the 36 and 100 ppm Mg levels, upon increasing the Mg ion concentration from ³⁶ to 100 ppm at the ³⁶ and 200 ppm Ca levels, and upon progressively increasing the Mg ion concentration from ⁴ to 100 ppm at the 100 ppm Ca level. At pH 5.4, significant increases in primary root growth of pepper were obtained upon progressively increasing the Ca ion concentration from ³⁶ to ²⁰⁰ ppm at all Mg levels and upon increasing the Mg ion concentration from ³⁶ to 100 ppm at the 36 and 100 ppm Ca levels. There was no significant interaction between Ca and Mg at the ²⁰⁰ ppm Ca level. At pH 6.0, significant increases in primary root growth of pepper were obtained upon progressively increasing the Ca ion concentration from ³⁶ to 200 ppm at the ⁴ and ³⁶ ppm Mg levels, upon increasing the Ca ion concentration from ³⁶ to 100 ppm at the 100 ppm Mg level, upon increasing the Mg ion concentration from ⁴ to ³⁶ ppm at the ³⁶ ppm Ca level, and upon progressively increasing the Mg ion concentration from 4 to 100 ppm at the 100 ppm Ca level. There was no significant interaction between Ca and Mg at the ²⁰⁰ ppm Ca level.

Liming with MgCO₃ in addition to CaCO₃ coupled with doubling the rate of N-P-K fertilization of the surface soil somewhat influenced the extent of the observed interaction between Ca and Mg in the subsurface solution consisting essentially of $caso^A$ and $MgSO^A$.

The results in Table ¹⁶ indicate that addition of $MgCO₃$ and increased fertilization of the surface soil had little effect on lettuce primary root growth in the subsurface solution. Significant increases in lettuce primary root growth were obtained upon progressively increasing the Ca ion concentration from ⁶ to 36 ppm at the ⁴ ppm Mg level and upon increasing the Mg ion concentration from ⁴ to 12 ppm at the ⁶ and ¹⁸ ppm Ca levels. There was no significant interaction between Ca and Mg at the ³⁶ ppm Ca level.

Similarly, the results in Table ¹⁸ indicate that addition of $MgCO₃$ and increased fertilization of the surface soil had little effect on pepper primary root growth in the subsurface solution. Significant increases in pepper primary root growth were obtained upon progressively increasing the Ca ion concentration from ³⁶ to ²⁰⁰ ppm at all Mg levels and upon progressively increasing the Mg ion concentration from 4 to 100 ppm at the 36 ppm Ca level. There were no significant interactions between Ca and Mg at the 100 and ²⁰⁰ ppm Ca levels. However, increases in root lengths for the less heavily limed and fertilized surface soil, averaged over the different levels of Mg, were significantly higher than those for the more heavily limed and fertilized surface soil at the 200 ppm Ca level.

In no instance did addition of increasing amounts of Mg in the presence of Ca result in ^a decrease in primary root growth of either lettuce or pepper in the subsurface

culture solutions.

The results of topgrowth analyses as influenced by differential treatment of the surface soil are presented in Tables 19 and 20,

The results indicate that, with the exception of Ca, there was ^a relative increase in per cent content of Mg, K, and P in the topgrowths with an increase in the Mg, N, P, and ^K applied to the surface soil. This increase had no effect on the per cent content of Ca in lettuce topgrowth, whereas it effected ^a reduction in the per cent content of Ca in pepper topgrowth. The concentrations of each of these elements in the plant tissue were greater than those normally considered critical for the growth of these crop species.

The results of primary root growth studies on lettuce and pepper as influenced by Ca and ^K ion concentrations in subsurface nutrient solutions consisting essentially of $CaSO_A$ and $K₂SO_A$ are presented in Tables 21 and 22 and Figures 11 and 12,

The data presented in Table ²¹ and Figure ¹¹ indicate that significant increases in primary root growth of lettuce in subsurface nutrient solutions were obtained upon increasing the Ca ion concentration from ⁶ to 18 ppm at the ⁰ and ¹⁵ ppm ^K levels, upon increasing the Ca ion concentration from ⁶ to ³⁶ ppm at the ³⁰ ppm ^K level, upon increasing the Ca ion concentration from ³⁶ to 100 ppm at the ⁰ ppm ^K level, and upon increasing the ^K ion concentration from ¹⁵ to ³⁰ ppm

ANALYSIS OF LETTUCE TOPGROWTH AT THE END OF EXPERIMENT, REFERENCE TABLE 16 ANALYSIS OF LETTUCE TOPGROWTH AT THE END OF EXPERIMENT, REFERENCE TABLE 16

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* Dry weight basis

ANALYSIS OF PEPPER TOPGROWTH AT THE END OF EXPERIMENT, REFERENCE TABLE 18 ANALYSIS OF PEPPER TOPGROWTH AT THE END OF EXPERIMENT, REFERENCE TABLE 18

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* Dry weight basis Dry weight basis

THREE-DAY LETTUCE PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND POTASSIUM ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

*Root lengths followed by the same letter are not significantly different at the $0*05$ probability level.

+Each treatment mean involves a maximum of 15 subsurface primary roots individually monitored in culture solutions consisting of $CaSO_4$. $2H_2O$, K_2SO_4 , and 2 ml of the micronutrient stock solution equivalent to 1/10—Steinberg; pH adjusted to 6.0 twice daily.

THREE-DAY PEPPER PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND POTASSIUM ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

Root lengths followed by the same letter are not significantly different at the 0.05 probability level.

+Each treatment mean involves a maximum of 16 sub surface primary roots individually monitored, in two separate trials, in culture solutions consisting of CaSO4.2H₂O, K₂SO₄, and ² ml of the micronutrient stock solution equivalent to l/10, Steinberg; pH adjusted to 6.0 twice daily.

Figure 12.--Effect of subsurface solution calcium and potassium ion concentrations on primary root elongation of pepper seedlings.

Figure 13.--Effect of subsurface solution calcium and aluminum ion concentrations on primary root elongation of lettuce seedlings.

at the ⁶ ppm Ca level. No significant interactions between Ca and ^K were observed at the ¹⁸ and ³⁶ ppm Ca levels.

On the other hand, the data presented in Table 22 and Figure ¹² indicate that significant increases in primary root growth of pepper in subsurface nutrient solutions were obtained upon progressively increasing the Ca ion concentration from ³⁶ to ²⁰⁰ ppm at all ^K levels and upon increasing the ^K ion concentration from ¹⁵ to ³⁰ ppm at the 100 ppm Ca level. No significant interactions between Ca and ^K were observed at the ³⁶ and 200 ppm Ca levels.

In no instance did addition of increasing amounts of ^K in the presence of Ca result in a decrease in primary root growth of either lettuce or pepper in the subsurface culture solutions.

The results of primary root growth studies on lettuce and pepper as influenced by Ca and Al ion concentrations in subsurface solutions consisting essentially of $CaSO_4$, MgSO₄, and $A1_2(SO_4)$ ₂ are presented in Tables 23 and 24 and Figures 13 and 14.

The data presented in Table ²³ and Figure 13 indicate that significant increases in primary root growth of lettuce in subsurface nutrient solutions were obtained upon increasing the Ca ion concentration from 36 to 100 ppm, in the presence of ⁴ ppm Mg, at the 0, 0.05, and 0.25 ppm Al levels. No significant inhibition of lettuce primary root growth occurred within a range of 0 to 0.05 ppm Al at both Ca levels. However,

Ill

THREE-DAY LETTUCE PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND ALUMINUM ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

Root lengths followed by the same letter are not Root lengths forrowed by the same receer are he significantly different at the 0.05 probability level.

+Each treatment mean involves a maximum of 16 sub surface primary roots individually monitored in culture solutions consisting of $CaSO_4$. $2H_2O$, $MgSO_4$. $7H_2O$, $Al_2(SO_4)$ 3. 18H₂O, and 2 ml of the micronutrient stock solution equivalent to 1/10-Steinberg; pH adjusted to 4.8 twice daily.

THREE-DAY PEPPER PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND ALUMINUM ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

*Root lengths followed by the same letter are not significantly different at the 0.05 probability level.

+Each treatment mean involves a maximum of 16 sub surface primary roots individually monitored, in two separate trials, in culture solutions consisting of $CaSO_4$. 2H₂O, trials, in culture solutions consisting or $\text{Cas}\,Q_*\text{Zn}_2\cup$,
MgSO₄.7H₂O, Al₂(SO₄)₃.18H₂O, and 2 ml of the micronutrient stock solution equivalent to 1/10-Steinberg; pH adjusted to 4.8 twice daily.

Figure 14.—Effect of subsurface solution calcium and aluminum ion con centrations on primary root elongation of pepper seedlings.

Figure 15.--Effect of subsurface solution calcium and phosphate ion concentrations on primary root elongation of pepper seedlings.

significant decreases in lettuce primary root growth occurred upon increasing the Al ion concentration from 0.05 to 0.25 ppm at the ³⁶ ppm Ca level and upon progressively increasing the Al ion concentration from 0.05 to 0.50 ppm at the 100 ppm Ca level. Al inhibition at 0.25 ppm Al was attenuated at the higher level of Ca.

On the other hand, the data presented in Table 24 and Figure ¹⁴ indicate that significant increases in primary root growth of pepper were obtained upon increasing the Ca ion concentration from 200 to 300 ppm, in the presence of ⁴ ppm Mg, at all Al levels. No significant inhibition of pepper primary root growth occurred within a range of ⁰ to 0.05 ppm Al at both Ca levels. However, significant decreases in pepper primary root growth occurred upon progressively increasing the Al ion concentration from 0.05 to 0.75 ppm at both Ca levels. Al inhibition at 0.25, 0.50, 0.75, and 1.00 ppm Al was attenuated at the higher level of Ca.

The results of primary root growth studies on pepper as influenced by Ca and phosphate ion concentrations in subsurface solutions consisting essentially of $CaSO_4^-$ and $Ca(H_{2}PO_{4})_{2}$ are presented in Table 25 and Figure 15.

The results indicate that significant increases in primary root growth of pepper in subsurface nutrient solutions were obtained upon progressively increasing the Ca ion concentration from 36 to 200 ppm. In contrast, significant inhibition of primary root growth of pepper in subsurface

THREE-DAY PEPPER PRIMARY ROOT GROWTH AS INFLUENCED BY CALCIUM AND PHOSPHATE ION CONCENTRATIONS IN SUBSURFACE NUTRIENT MEDIUM

*Root lengths followed by the same letter are not significantly different at the 0.05 probability level.

+Each treatment mean involves a maximum of ¹⁶ sub surface primary roots individually monitored, in two separate trials, in culture solutions consisting of CaSO₄. ²H²⁰, Ca(H2P04)2.H2O, and ² ml of the micronutrient stock solution equivalent to 1/10-Steinberg; pH adjusted to 6.0 twice daily.

nutrient solutions was observed upon increasing the phosphate ion concentration from ⁰ to ⁶ ppm ^P at the 100 and ²⁰⁰ ppm Ca levels. No significant interaction between Ca and ^P was observed at the ³⁶ ppm Ca level. There was also no further significant decrease in pepper primary root growth upon increasing the phosphate ion concentration from ⁶ to ¹² ppm P. However, it was observed that lateral roots of pepper increased in number and length with increasing phosphate ion concentration up to the 12 ppm ^P level studied.

DISCUSSION

Preliminary Subsoil and Culture Solution Studies On Six Crop Species

The results of the preliminary studies on the primary root growth of the six crop species in acid subsoils could be partly explained on the basis of the preliminary culture solution experiments.

The solution displaced from the unlimed Narragansett loam subsoil contained ²⁵³ ppm Ca and 0.70 ppm Al; the final pH of the subsoil, averaged for all the crop species, was 5.02. The solution displaced from the limed Narragansett loam subsoil contained ²⁹⁹ ppm Ca and 0.14 ppm Al; the final pH of the subsoil, averaged for all the crop species, was 5.66. The solution displaced from the unlimed Merrimac sandy loam subsoil contained 14.2 ppm Ca and 0.56 ppm Al; the final pH of the subsoil, averaged for all the crop species, was 4.76. The solution displaced from the limed Merrimac sandy loam subsoil contained ¹⁷⁶ ppm Ca and practically zero Al; the final pH of the subsoil, averaged for all the crop species, was 5.66.

The primary root growth of pea was not significantly altered by liming either subsoil. This is consistent with the results of the culture solution experiments. The primary root growth of pea was not significantly altered within a pH range of 4.4 to 6.0 and within a range of 0 to 2.5 ppm Al;

it, in fact, increased significantly at the ⁴ ppm Al level, whereas it did not increase significantly beyond the ¹² ppm Ca level in culture solution.

The primary root growth of cucumber increased significantly after ⁵ days in the limed Merrimac sandy loam subsoil, whereas it consistently decreased significantly in the limed Narragansett loam subsoil. This is somewhat inconsistent with the results of the culture solution experiments. The primary root growth of cucumber was not significantly altered within a pH range of 4.4 to 6.0 and within a range of ⁰ to 2.5 ppm Al. It did not increase significantly beyond the ⁴ ppm Ca level in culture solution. However, the fact that it increased highly significantly at the ⁴ ppm Al level in culture solution might help to explain the significant decrease of cucumber primary root growth in the limed Narragansett loam subsoil. The significant increase in cucumber primary root growth in the limed Merrimac sandy loam subsoil cannot be explained as a result of neutralization of subsoil acidity or increase in solution Ca.

The primary root growth of lettuce increased significantly upon liming either subsoil. This is consistent with the results of the culture solution experiments. The primary root growth of lettuce did not increase significantly beyond the 0.6 ppm Ca level in culture solution. However, it was significantly reduced at 0.5 ppm Al and above, and at a pH below 4.8. The significant increase in lettuce

primary root growth upon liming either subsoil may therefore be explained as ^a result of neutralization of exchangeable A1 and subsoil acidity.

The primary root growth of spinach increased significantly upon liming either subsoil. This is consistent with the results of the culture solution experiments. The primary root growth of spinach increased with increasing Ca ion concentration up to the maximum 36 ppm Ca level studied in culture solution. However, it was stimulated within ^a range of ⁰ to 0.5 ppm Al and then markedly inhibited at upwards of 0.5 ppm Al and at a pH below 4.8. The significant increase in spinach primary root growth upon liming either subsoil may therefore be explained as a result of neutralization of exchangeable Al and subsoil acidity, with the possibility of stimulation of spinach primary root growth at low Al concentrations.

The primary root growth of pepper consistently decreased but not significantly in the limed Merrimac sandy loam subsoil, whereas it increased significantly in the limed Narragansett loam subsoil. This is somewhat consistent with the results of the culture solution experiments. The primary root growth of pepper did not increase significantly beyond the ⁶ ppm Ca level in culture solution. However, it was significantly reduced at the 0.5 ppm Al level and above, and at a pH below 4.8. The significant increase in pepper primary root growth in the limed Narragansett loam subsoil

may therefore be explained as ^a result of neutralization of exchangeable Al and subsoil acidity. The consistent, nonsignificant decrease in primary root growth in the limed Merrimac sandy loam subsoil cannot, however, be explained in this fashion.

The primary root growth of tomato decreased significantly upon liming either subsoil. This is inconsistent with the results of the culture solution experiments. The primary root growth of tomato did not increase significantly beyond the ¹² ppm Ca level in culture solution. It was not significantly altered within ^a pH range of 4.4 to 6.0. It was, however, significantly reduced at the 2.5 ppm Al level and above. The significant decrease in tomato primary root growth upon liming either subsoil cannot therefore be explained as a result of neutralization of exchangeable Al.

Significant reduction in the primary root growth of some of these crop species upon liming either subsoil could be partly due to interference of Ca with the availability of some other nutrient element or elements required in the meristematic regions of the primary roots for increased growth in the subsoil. It is, however, clear from the limeplus-boron experiments that poor primary root growth in these subsoils was not caused by boron deficiency, furthermore, reducible Mn was determined as ^a measure of the relative supplying power of the soil layers for soluble Mn, using the hydroquinone-NH40Ac reducing method (13). Values obtained

were 0.10, 0.03, 0.04, and 0.10 meq/100 ^g soil for Narragansett loam topsoil and subsoil and for Merrimac sandy loam topsoil and subsoil respectively. This also would clearly suggest that poor primary root growth in these subsoils was not caused by Mn toxicity to primary roots.

Preliminary Subsurface Solution Studies on Two Selected Crop Species

Lettuce and pepper primary roots have different susceptibilities to hydrogen and aluminum ion concentrations and different requirements for Ca in subsurface 1/5-strength Steinberg nutrient solutions.

The primary root growth of lettuce was significantly inhibited at some pH between 4.5 and 4.8 and at a concentration of 0.5 ppm Al. Near maximum primary root growth of lettuce in the subsurface nutrient solution was obtained at a concentration of ¹ ppm Ca within the range of ¹ to 100 ppm Ca tested.

The primary root growth of pepper was significantly inhibited at a pH immediately less than 4.8 and at a concentration of 0.5 ppm Al. Near maximum primary root growth of pepper in the subsurface nutrient solution was obtained at a concentration of 72 ppm Ca within the range of ¹ to 100 ppm Ca tested.

The data for lettuce, unlike those for pepper, are in agreement with those of Jones and Lunt (89) and Lund

(108) which show Ca requirements to be very low when other cations are in balance and roots are growing in the absence of toxic ions. The roots in these experiments were growing in well-aerated nutrient solutions and, therefore, Ca supply to the root surface was replenished rapidly.

Subsurface Solution Studies on Two Selected Crop Species Involving Cation Interactions

A. Solution pH and Ca Experiments

The response of lettuce and pepper primary root growth to varying Ca ion concentration was adversely affected by increasing ^H ion concentration or decreasing pH of the subsurface $CaSO_A$ nutrient medium.

At pH 4.2, the toxicity of H ions completely prevented growth of lettuce primary roots at all levels of Ca studied. The toxicity of ^H ions to lettuce primary roots occurred even at the ³⁶ ppm Ca level at pH 4.5; at this pH value, 70% of the maximum rate of lettuce primary root growth was obtained at the ³⁶ ppm Ca level.

Even though there were no interactions between Ca and ^H ions at the suboptimal ²⁴ and ³⁶ ppm Ca levels, the toxicity of ^H ions to pepper primary roots occurred at pH values of 4.4 and 4.6 at the 100 ppm Ca level. At these pH values, less than 50% of the maximum rate of pepper primary root growth was obtained at the 100 ppm Ca level.

However, a rate of pepper primary root growth greater than that in a $CaSO_A$ nutrient medium of the same Ca ion concentration was obtained using a subsurface 1/5-strength Steinberg nutrient medium. The toxicity of ^H ions occurred at all levels of Ca studied at pH values of 4.4 and 4.6. At these pH values, about 55% and 85% respectively of the maximum rate of pepper primary root growth were obtained at the 100 ppm Ca level. This would clearly suggest that pepper primary root growth in the subsurface 1/5-strength Steinberg nutrient medium was being stimulated by some nutrient element or elements other than Ca.

Lund (108) found that the interaction in the effects of Ca and ^H ions on soybean primary root growth was related to the ratio of the molar activities of ^H to Ca.

Cationic concentrations used in subsurface solution 0 studies were converted to molar activities which should more closely approximate effective concentration. The Debye-Huckel equation for single-ion activity coefficients was used:

$$
- \log f = \frac{A z^2 u^{1/2}}{1 + B a u^{1/2}},
$$

where $f =$ activity coefficient of ion, $u =$ ionic strength of the solution, $z =$ valence of ion, $A = 0.509$, $B = 0.329$, and a = effective diameter of the hydrated ion, in Angstrom units (94).

The ionic strengths of the solutions were calculated by assuming that the Mn and Al in solution were present as

the divalent and trivalent ions respectively and that all the complementary anions were monovalent. Having calculated the activity coefficients, the molar activities of individual cations were then computed (2), Consideration of activity coefficients assuming that complementary anions were divalent did not improve the relationship between the molar activities of the other cations and that of Ca as computed by the Debye-Huckel equation.

The toxicity of ^H ions was a factor at pH values of 4.2, 4.5, and 4.8 for lettuce primary root growth in subsurface culture solutions. The data in Figure ¹⁶ show that, over this range of solution pH, lettuce subsurface primary root growth was a function of the a_H/a_C ratio of the subsurface nutrient medium. Lettuce primary root growth in the subsurface nutrient medium was significantly inhibited when the ratio of the molar activities of ^H to Ca exceeded 0.03. Omitted from the graph were the zero root growth values for solutions having a pH of 4.2.

The toxicity of H ions was a factor at pH values of 4.4, 4.6, and 4.8 for pepper primary root growth in subsurface culture solutions. The data in Figure ¹⁷ show that, over this range of solution pH, pepper primary root growth in subsurface culture solution was also a function of the a_H/a_C ratio of the CaSO₄ and 1/5-strength Steinberg nutrient media. The rate of pepper primary root growth in the 1/5 strength Steinberg solution was clearly higher than that in

Figure 16.—Effect of ratio of molar activities of ^H to Ca on lettuce primary root elongation in subsurface nutrient solution.

Figure 17.—Effect of ratio of molar activities of ^H to Ca on pepper primary root elongation in subsurface nutrient solution.

the CaSO, solution at any given a H/^aCa ratio over this range of solution pH, Pepper primary root growth in both kinds of subsurface nutrient media was significantly inhibited when the ratio of the molar activities of ^H to Ca exceeded about 0.015.

B. Solution Mg, pH, and Ca Experiments

Having established the adverse effects of ^H ions on the response of lettuce and pepper primary root growth to varying Ca concentration, experiments were then designed to study the interactions among Ca, Mg, and ^H ions in subsurface nutrient medium. It was found that significant interactions existed among Ca, Mg, and H ions for lettuce and pepper primary root growths in the subsurface nutrient medium consisting essentially of $\text{Cas}\sigma_4$ and MgSO₄.

The data in Table 16 indicate that minimum Ca levels required for lettuce primary root growth were dependent upon the pH and Mg ion concentration of the subsurface nutrient medium. Lettuce primary root growth generally increased upon increasing the Ca ion concentration up to the ³⁶ ppm Ca level, upon increasing the pH from 4.8 to 6.0, and upon increasing the Mg ion concentration from ⁴ to ³⁶ ppm. However, there was no significant interaction between Ca and Mg at the ³⁶ ppm Ca level at either pH 4.8 or 6.0. Also no significant increases in primary root growth of lettuce in the subsurface culture solutions, averaged over the different levels of Mg, were obtained at the 36 ppm Ca level upon increasing

the pH from 4.8 to 6.0.

The data in Table 17 also indicate that minimum Ca levels required for pepper primary root growth were dependent upon the pH and Mg ion concentration of the subsurface nutrient medium. Pepper primary root growth generally in creased upon increasing the Ca ion concentration up to the ²⁰⁰ ppm Ca level, upon increasing the pH from 4.5 to 6.0, and upon increasing the Mg ion concentration from ⁴ to 100 ppm. However, there were no significant interactions between Ca and Mg at the ³⁶ and 200 ppm Ca levels at pH 4.5 and at the ²⁰⁰ ppm Ca level at either pH 5.4 or 6.0.

These data are not at all in agreement with those of Lund (108) which showed suppression of primary root growth of soybeans in subsurface culture solutions as a result of high levels of Mg, those of Walker et al. (166) which evidenced antagonistic effects of Mg on Ca in the adsorbed state in serpentine soils and in culture solutions for sunflower growth, those of Vlamis (165) which showed lettuce rosette symptoms of Ca deficiency to be induced in solutions low in Ca and more severely in solutions low in Ca and high in Mg or K, those of Howard and Adams (75) which also evidenced antagonistic effects of Mg on Ca for cotton primary root growth and wittingly led to the conclusion that effects of Ca on cotton primary root growth could be measured with out regard to possible deficiencies of other macronutrients in the subsoil, and those of Adams (1) and Adams and Lund (2)

which implied the antagonistic effects of cations other than ^H and A1 on Ca for cotton primary root growth in the subsoil.

It would appear from the data in Tables ¹⁶ and ¹⁷ that Mg was needed for the primary root growth of either lettuce or pepper only when the Ca present in the subsurface solution was inadequate for near maximum primary root growth of either crop species. Also, the results of liming with MgCO₃ in addition to $CaCO_3$ coupled with doubling the rate of N-P-K fertilization of the surface soil indicate significant response of lettuce primary root growth to increasing Mg ion concentration at the 6 and 18 ppm Ca levels and significant response of pepper primary root growth to increasing Mg ion concentration at the 36 ppm Ca level in subsurface nutrient solutions. Furthermore, the results of topgrowth analyses indicate that the concentrations of Ca, Mg, N, P, and ^K in the plant tissues were greater than those normally considered critical for the growth of either crop species. Therefore the response of either lettuce or pepper primary root growth to added Mg was not ^a result of Mg deficiency. It would, however, appear that this was not necessarily ^a Mg requirement per se since near maximum primary root growth of either crop species was obtained when solution Ca alone was present in adequate amounts in the subsurface nutrient solutions. It would also appear that Mg did not substitute for Ca in detoxifying ^H ions for more favorable primary root growth. Significant increases in lettuce primary root growth

were obtained upon increasing the Mg ion concentration from ⁴ to 12 ppm at the ⁶ ppm Ca level and upon progressively increasing the Mg ion concentration from ⁴ to ³⁶ ppm at the 18 ppm Ca level at pH 6.0. At the same pH value, significant increases in pepper primary root growth were obtained upon increasing the Mg ion concentration from ⁴ to ³⁶ ppm at the 36 ppm Ca level and upon progressively increasing the Mg ion concentration from ⁴ to 100 ppm at the 100 ppm Ca level.

Heavy liming coupled with heavy N-P-K fertilization tended to minimize the levels of Ca and/or Mg beyond which significant interactions between Ca and Mg ceased to exist. This was probably due to the greater downward translocation of Mg within the plant tissues to the meristematic regions of the primary roots.

The concentrations of the cations were converted to molar activities as previously described. The ratios of the molar activities of ^H to Ca were then plotted as ^a means of explaining the antagonistic effects of ^H on Ca even in the presence of varying amounts of Mg.

The data in Figure ¹⁸ indicate that the toxicity of ^H ions apparently was not a factor at a solution pH of 6.0 for lettuce subsurface primary root growth. At pH 4.8, the data, however, indicate that minimum Ca levels required for lettuce subsurface primary root growth were dependent upon pH even in the presence of varying amounts of Mg in the subsurface nutrient solution.

Figure 18.—Effect of ratio of molar activities of ^H to Ca on lettuce primary root elongation in subsurface nutrient solution.

Figure 19.—Effect of ratio of molar activities of ^H to Ca on pepper primary root elongation in subsurface nutrient solution.

The data in Figure ¹⁹ indicate that the toxicity of ^H ions apparently was also not a factor at a solution pH of 6.0 for pepper subsurface primary root growth. At pH values of 4.5 and 4.8, however, the data indicate that minimum Ca levels required for pepper subsurface primary root growth were dependent upon pH even in the presence of varying amounts of Mg in the subsurface nutrient solution.

Molar activity ratios of Ca/total cations have also been suggested by other investigators as a means of explaining the antagonistic effects of other cations on Ca. Such ratios were therefore computed for these experiments and plotted. The data presented in Figures ²⁰ and ²¹ show clearly that lettuce and pepper primary root growth in subsurface culture solutions could not be explained by these ratios, since the major premise of antagonism of Mg toward Ca on which explanation by such ratios is based was totally false insofar as these two crop species were concerned.

C. Solution ^K and Ca Experiments

These experiments were designed to study the interactions between Ca and ^K ions in subsurface nutrient medium. It was found that significant interactions existed between Ca and ^K ions in subsurface nutrient medium for lettuce and pepper primary root growth.

Lettuce primary root growth was significantly increased upon increasing the ^K ion concentration from ¹⁵ to ³⁰ ppm at the ⁶ ppm Ca level in the subsurface nutrient

Figure 20.—Effect of Ca/totaI-cation molar activity ratio on lettuce primary root elongation in subsurface nutrient solution.

Figure 21.--Effect of Ca/total-cation molar activity ratio on pepper primary root elongation in subsurface nutrient solution.

medium. Similarly, pepper primary root growth was significantly increased upon increasing the ^K ion concentration from 15 to ³⁰ ppm at the 100 ppm Ca level in the subsurface nutrient medium. Even at the smallest Ca concentrations studied, there was no decrease in primary root growth of either lettuce or pepper upon increasing the concentration of ^K in the subsurface culture solution.

These data are clearly not in agreement with those of Lund (108) which showed suppression of primary root growth of soybeans in subsurface culture solutions as a result of high levels of K, those of Vlamis (165) which showed lettuce rosette symptoms of Ca deficiency to be induced in solutions low in Ca and more severely in solution low in Ca and high in Mg or K, and those of Adams (1) and Adams and Lund (2) which implied the antagonistic effects of cations other than than ^H and Al on Ca for cotton primary root growth in the subsoil.

The concentrations of the cations were converted to molar activities as previously described. The effects of molar activities of Ca on lettuce and primary root elongation in subsurface Ca-Mg and Ca-K systems at pH 6.0 were plotted with a view to comparing the contribution of Mg and ^K to lettuce and pepper primary root growth over and above that of Ca alone.

The data in Figure 22 indicate that Mg stimulated lettuce primary root growth in subsurface culture solutions

Figure 22.—Effect of molar activity of Ca on lettuce primary root elongation in subsurface nutrient solution at pH 6.0.

Figure 23.—Effect of molar activity of Ca on pepper primary root elongation in subsurface nutrient solution at pH 6.0.

many times more than ^K did, in addition to the stimulation observed as due to Ca alone.

The data in Figure ²³ indicate that Mg stimulated pepper primary root growth in subsurface culture solutions only several times more than ^K did, in addition to the stimulation observed as due to Ca alone, and only up to a point beyond which clearly no further stimulation of pepper primary root growth by Mg or ^K occurred over and above that observed as due to Ca alone.

It is therefore suggested that stimulation of pepper primary root growth in the subsurface 1/5-strength Steinberg solution over and above that in the subsurface Cas_4 solution was probably the added effect of the Mg and ^K present in a 1/5-strength Steinberg solution that also contained an amount of Ca equal to that in a CaSO $_A$ solution.

D. Solution Al and Ca Experiments

These experiments were designed to study the antagonistic effects of Al on Ca ions in the subsurface nutrient medium. It was found that significant interactions existed between Ca and Al ions in the subsurface nutrient medium for lettuce and pepper primary root growth.

At the 36 ppm Ca level, 0.05 ppm Al did not significantly reduce primary root growth of lettuce in subsurface culture solutions, whereas 0.25, 0.50, or 0.75 ppm Al completely inhibited lettuce primary root elongation. At the 100 ppm Ca level, 0.05 ppm Al did not significantly reduce

primary root growth of lettuce in subsurface culture solutions. 0.25 ppm Al significantly reduced primary root growth, whereas 0.50 or 0.75 ppm Al completely inhibited lettuce primary root elongation. However, lettuce roots growing at the higher levels of Ca were less susceptible to Al toxicity at 0.25 ppm Al and below.

In contrast, at the ²⁰⁰ ppm Ca level, 0.05 ppm Al did not significantly reduce primary root growth of pepper in subsurface culture solutions, whereas 0.25, 0.50, 0.75, and 1.00 ppm Al progressively inhibited pepper primary root elongation significantly. At the ³⁰⁰ ppm Ca level, 0.05 ppm Al did not significantly reduce primary root growth of pepper in subsurface culture solutions, whereas 0.25, 0.50, 0.75, and 1.00 ppm Al progressively inhibited pepper primary root elongation significantly. However, pepper roots growing at the higher levels of Ca were less susceptible to Al toxicity at all levels of Al tested.

The molar activities of solution Al have been suggested by other investigators as a means of explaining the antagonistic effects of subsurface solution Al on Ca. The concentrations of the cations were therefore converted to molar activities as previously described.

The data in Figure 24 indicate that significant reduction in the primary root growth of lettuce in subsurface culture solutions occurred when the molar activity of Al exceeded 0.1 X 10⁻⁵ and that primary root elongation was

Figure 24.—Effect of molar activity of Al on lettuce primary root elongation in subsurface nutrient solution.

Figure 25.—Effect of molar activity of Al on pepper primary root elongation in subsurface nutrient solution.

completely inhibited when the molar activity of Al exceeded 0.5×10^{-5} .

In contrast, the data in Figure ²⁵ indicate that significant progressive reduction in the primary root growth of pepper in subsurface culture solutions occurred when the molar activity of Al exceeded about 0.15 X 10⁻⁵.

These data are remarkably similar to those of Adams and Lund (2) which also showed solution Al to be progressively more toxic to cotton subsurface primary roots as the molar activity of Al exceeded a minimum of about 0.15×10^{-5} .

Lund (108) suggested that the ratios of the molar activities of Al to Ca were more closely related to the susceptibility of primary root growth in subsurface culture solutions to Al damage than molar activities of Al alone.

The data in Figure ²⁶ indicate that significant reduction in the primary root growth of lettuce in subsurface culture solutions occurred when the ratio of molar activities of Al to Ca exceeded about 0.001 and that lettuce primary root elongation was completely inhibited when the ratio of the molar activities of Al to Ca exceeded about 0.005.

In contrast, the data in Figure ²⁷ indicate that significant progressive reduction in the primary root growth of pepper in subsurface culture solutions occurred when the ratio of the molar activities of Al to Ca exceeded about 0.0005.

root elongation in subsurface nutrient solution.

Figure 27.--Effect of ratio of molar activities of Al to Ca on pepper primary root elongation in subsurface nutrient solution.

These data are not in agreement with those of Lund (108) which suggested that A1 in nutrient solution reduced soybean primary root growth in subsurface culture solutions when the ratio of the molar activities of A1 to Ca exceeded ⁰.02.

E. Solution ^P and Ca Experiments

These experiments were designed, out of curiosity, to study the effects of phosphate on Ca ions in the subsurface nutrient medium. It was found that significant interactions existed between Ca and phosphate ions in the subsurface nutrient medium for pepper primary root growth.

When the levels of Ca were adequate, ⁶ ppm ^P was sufficient to markedly inhibit primary root elongation of pepper in subsurface culture solutions. However, lateral roots of pepper increased in number and length with increasing phosphate ion concentration up to the ¹² ppm ^P level tested.

From the foregoing, it is abundantly clear that a critical Ca concentration for optimum primary root growth of either lettuce or pepper in subsurface culture solutions cannot be easily defined because of the other chemical factors beside Ca influencing primary root growth. Cognizance should therefore be taken of the stimulation of primary root growth by Mg and ^K and the inhibition of primary root growth by H, Al, and possibly ^P in an intricate process of determining critical Ca concentrations for optimum primary root growth of these two crop species in subsurface culture solutions

Subsoil Studies on Two Selected Crop Species

^A clear attempt to discuss the results of the subsoil experiments in the light of results obtained for lettuce and pepper primary root growth in subsurface culture solution experiments would necessitate comparisons of the molar activity relationships in both kinds of subsurface media. The molar activity relationships in solutions displaced from the Narragansett loam and Merrimac sandy loam subsoils are presented in Tables 26, 27, 28, and 29.

^A thorough inspection and an exhaustive study and plotting of the results of the subsoil experiments have revealed beyond a reasonable doubt that the response of lettuce and pepper primary root growth in the subsoil to the CaCO₃ and/or MgCO₃ treatments could not at all be explained either by the concentrations or activities of the individual cations per se or by molar activity ratios of Ca/total cations since, as suggested earlier, the major premise of antagonism of cations other than ^H and Al toward Ca on which explanation by such ratios is based was totally false insofar as these two crop species were concerned. Figures ²⁸ and ²⁹ present the effect of molar activity ratios of Ca/total cations on lettuce and pepper primary root elongation respectively in the subsoil.

EFFECT OF CaCO₃ AND MgCO₃ ADDITIONS ON MOLAR ACTIVITIES OF CATIONS IN SOLUTIONS DISPLACED FROM

TABLE 26

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TABLE 29

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Figure 28.--Effect of Ca/total-cation molar activity ratio on lettuce primary root elongation in the subsoil.

Figure 29.--Effect of Ca/total-cation molar activity ratio on pepper primary root elongation in the subsoil.

A. Narragansett Loam Subsoil

(i) Lettuce. Significant reduction in lettuce primary root growth occurred when the ratio of molar activities of ^H to Ca exceeded about 0.03 at pH values below 6.0 in the subsurface culture solutions. However, the ratio of molar activities of ^H to Ca for the ⁶ subsoil treatments did not exceed a maximum of 0.003. Therefore, the differences in treatment results could not be explained thereby.

Significant reduction in primary root growth of lettuce occurred when the molar activity of A1 exceeded 0 .1 X 10 $^{-5}$ or when the ratio of molar activities of Al to Ca exceeded 0.001 in subsurface culture solutions. However, this did not explain the differences in subsoil treatment results. For example, subsoil treatment No. ³ which had the highest molar ^aAl/^aCa ratio of about 0.003 actually gave the most significant increase in lettuce primary root growth.

The range of activities of Ca studied in the subsurface culture solution experiments was from about 12 to 167 X 10^{-5} M Ca. Near maximum lettuce primary root growth was obtained at activities beyond 69 X 10⁻⁵M Ca. The molar activities of Ca in the subsoil treatments are, however, well above this range. This would therefore suggest that there should be no significant response of lettuce primary root growth in the subsoil to added Ca and would also eliminate the possibility that there could be ^a natural deficiency of Ca in this subsoil for lettuce primary root growth. However,

subsoil treatment No. 3, to which 0.54 meq Mg⁺⁺/100 g subsoil had been added, gave the most significant increase in lettuce primary root growth. This would suggest that this increase in root growth was due to stimulation of lettuce subsoil primary root growth by Mg added to an untreated Narragansett loam subsoil that already had an adequate amount of Ca in it.

(ii) Pepper. Significant reduction in pepper primary root growth occurred when the molar aH/dc ratios exceeded 0.015 at pH values below 6.0 in subsurface culture solutions. However, the molar a_H/a_C ratios for the 6 subsoil treatments did not exceed a maximum of about 0.004. Therefore, the differences in treatment results could not be explained thereby.

Significant reduction in pepper primary root growth occurred when the molar activity of Al exceeded about 0.15 ^X 10^{-5} or when the molar a_{A1}/a_{Ca} ratio exceeded 0.0005 in subsurface culture solutions. However, this did not explain the differences in subsoil treatment results. For example, the results of subsoil treatment No. 3, which had the highest molar ^aAl/^aCa ratio of about 0.003, were not significantly different from those of subsoil treatments Nos. 4, 5, and ⁶ but were significantly superior to those of subsoil treatment No. ¹ to which neither Ca nor Mg had been added.

The range of activities of Ca studied in the subsurface culture solution experiments was from about 53 to 407 X 10⁻⁵M Ca. Near maximum response of pepper primary root

growth to added Ca was obtained at activities of about ⁴⁰⁷ ^X 10⁻⁵M Ca at pH values of up to 6.0 in subsurface culture solutions. The molar activities of Ca in the subsoil treatments, however, range from about 209 to 400 X 10^{-5} M Ca. This would therefore suggest that pepper subsurface primary root growth in the subsoil should respond to added Ca. Subsoil treatment No. 1, to which neither Ca nor Mg had been added, yielded significantly less primary root growth than did the other subsoil treatments particularly after the third day. However, subsoil treatment No. 3, to which only Mg had been added, yielded as good a growth as did subsoil treatments Nos. 2, 4, 5, and ⁶ which consisted of varying amounts of Ca and Mg. This indicates that the subsoil was not naturally deficient in Ca for pepper primary root growth and that the positive response of pepper primary root growth to added Ca or Mg was generally as ^a result of neutralization of subsoil acidity. It is indeed interesting to note that the activity of Ca for subsoil treatment No. 3 was equivalent to a concentration between 100 and 200 ppm Ca in the subsurface culture solution experiments. This is therefore consistent with the observation that significant positive interaction between Ca and Mg occurred in pepper primary root growth at the 100 ppm but not at the 200 ppm Ca level at pH values above 4.8 in the subsurface culture solution experiments.

B. Merrimac Sandy Loam Subsoil

(i) Lettuce. Significant reduction in lettuce primary root growth in the subsoil could not be explained on the basis of the molar a_H/a_C ratios. For example, the molar a_{H}/a_{Ca} ratios for subsoil treatments Nos. 1 and 3 exceeded the critical limit of about 0.03 found for lettuce primary root growth in subsurface culture solutions at pH values below 6.0. Yet their results were not significantly different from those of subsoil treatment No. ⁶ which had a subcritical molar a H/ d Ca ratio of about 0.001.

Significant reduction in lettuce primary root growth in the subsoil could not be explained on the basis of the molar activities of Al or on the basis of the molar a_{A1}/a_{Ca} ratios. For example, the molar ^aAl/^aCa ratios for subsoil treatments Nos. 1, 2, and ³ exceeded the critical limit of about 0.001 found for lettuce primary root growth in subsurface culture solutions. Yet their results were not significantly different from those of subsoil treatment No. ⁶ which had a sub-critical molar a_{A1}/a_{Ca} ratio of practically zero.

The molar activities of Ca in the subsoil treatments ranged from about 15 to 283 X 10⁻⁵M Ca. Since near maximum response of lettuce primary root growth was obtained in subsurface culture solutions at activities beyond 69 X 10^{-5} M Ca, the subsoil data would suggest that lettuce primary root growth in the subsoil should respond to added Ca. However,

the results of subsoil treatment No. 1, to which neither Ca nor Mg had been added, were not significantly different from those of the other subsoil treatments. The slight superiority of treatments Nos. ⁴ and ⁵ to the other treatments would suggest that slightly greater lettuce primary root growth could be obtained by simultaneously adding Ca and Mg to the subsoil. In contrast, the slight inferiority of treatment No. ⁶ to the other treatments by the seventh day would suggest a possibility of overliming injury due to excess $CaCO₃$ in the Merrimac sandy loam subsoil. The data indicate that the Merrimac sandy loam subsoil was not naturally deficient in Ca for lettuce primary root growth.

(ii) Pepper. Significant reduction in pepper primary root growth in the subsoil could not be explained on the basis of molar a_H/a_C ratios. For example, the molar a_H/a_C ratios for subsoil treatments Nos. ¹ and ³ exceeded the critical limit of 0.015 found for pepper primary root growth in subsurface culture solutions at pH values below 6.0. Yet their results were not significantly different from those of subsoil treatments Nos. ⁵ and ⁶ which had sub—critical molar $a_{\text{H}}/a_{\text{Ca}}$ ratios of about 0.003 and 0.001 respectively.

Significant reduction in pepper primary root growth in the subsoil could not be explained on the basis of the molar activities of Al or on the basis of the molar ^aAl/^aCa ratios. For example, the molar ^aAl/^aCa ratios for subsoil treatments Nos. 1, 2, and ³ exceeded the critical limit of

about 0.0005 found for pepper primary root growth in subsurface culture solutions. Yet their results were not significantly different from those of subsoil treatments Nos. 5 and 6 which had sub-critical molar a_{A1}/a_{Ca} ratios of about 0.0002 and practically zero respectively.

The molar activities of Ca in the subsoil treatments ranged from about 15 to 283 X 10^{-5} M Ca. Near maximum response of pepper primary root growth was obtained at activities of about 407 X 10^{-5} M Ca at pH values of up to 6.0 in subsurface culture solutions. This would therefore suggest that pepper primary root growth in the subsoil should respond to added Ca. However, after the third day, subsoil treatment No. 1, to which neither Ca nor Mg had been added, gave as good growth as any of subsoil treatments Nos. 2, 3, 5, and ⁶ to which varying amounts of Ca and/or Mg had been added. The significant superiority of treatment No. ⁴ over treatments Nos. ¹ and ⁵ would suggest that while the subsoil is not naturally deficient in Ca for primary root growth of pepper, pepper primary root growth would respond positively to a particular combination of Ca and Mg added to the Merrimac sandy loam subsoil. In contrast, the slight inferiority of treatment No. ⁶ to subsoil treatments Nos. ¹ and ² by the fifth day would suggest ^a possibility of overliming injury due to excess CaCO₃ in the Merrimac sandy loam subsoil.

C. AlCl^-Leached Narragansett Loam Subsoil

(i) Lettuce. Significant reduction in lettuce primary root growth in the $A1Cl_{2}$ -leached subsoil could be explained on the basis of molar a_{H}/a_{Ca} ratios only up to a point. The molar a_H/a_C ratios for subsoil treatments Nos. ¹ and ³ exceeded the critical limit of about 0.03 found for lettuce primary root growth in subsurface culture solutions at pH values below 6.0. Yet their results were not significantly different from those of subsoil treatments Nos. ² and 4 which had sub-critical molar a_{H}/a_{Ca} ratios of about 0.016 and 0.004 respectively. However, subsoil treatment No. 6, which had the lowest molar a_{H}/a_{Ca} ratio of 0.003, gave the highest and most significant growth followed by subsoil treatment No. 5 which had a sub-critical molar a_H/a Ca ratio of 0.008.

Significant reduction in lettuce primary root growth in the AlCl₃-leached subsoil could be explained on the basis of the molar activities of Al only up to a point. The molar activities of Al in subsoil treatments Nos. 1, 2, 3, 4, and 5 far exceeded the critical limit of 0.1 X 10⁻⁵ found for lettuce primary root growth in subsurface culture solutions. Yet the results of subsoil treatment No. 5 were significantly superior to those of subsoil treatments Nos. 1, 2, 3, and 4. However, subsoil treatment No. 6, which had a sub—critical molar activity of Al of 0.09 X 10⁻⁵, gave the highest and most significant lettuce primary root growth. In contrast,

reduction in lettuce primary root growth in the AlCl₃-leached subsoil could not be explained on the basis of the molar $a_{\rm Al}/a_{\rm Ca}$ ratios. The molar $a_{\rm Al}/a_{\rm Ca}$ ratios for all the subsoil treatments exceeded the critical limit of about 0.001 found for lettuce primary root growth in subsurface culture solutions. Yet the results of subsoil treatments Nos. ⁵ and ⁶ were distinctly superior to those of the other treatments. It is, however, noteworthy that subsoil treatment No. 6, which had a near-critical molar a_{A1}/a_{Ca} ratio of about 0.002 , gave the highest and most significant lettuce primary root growth.

The molar activities of Ca in the $AICl_{3}-l$ eached subsoil treatments ranged from about 3 to 49 X 10⁻⁵M Ca. Since near maximum response of lettuce primary root growth was obtained in subsurface culture solutions at activities beyond 69 X 10⁻⁵M Ca, the subsoil data would suggest that lettuce primary root growth in the leached subsoil should readily respond to added Ca. Nevertheless, the results of subsoil treatments Nos. ² and 4, that had been treated with variable amounts of Ca and Mg, were not significantly different from those of subsoil treatment No. ¹ to which neither Ca nor Mg had been added. However, subsoil treatments Nos. ⁵ and ⁶ were distinctly superior to the rest. This would suggest that lettuce primary root growth in the leached Narragansett loam subsoil would respond to added CaCO₃ or to a particular combination of $CaCO_3$ and MgCO₃ only when in excess of that

required to neutralize the exchangeable Al present in the leached subsoil.

(ii) Pepper. Significant reduction in pepper primary root growth in the $AICl_{3}-leached$ subsoil could be explained on the basis of molar a_H/a_C ratios only up to a point. The molar a_H/a_C ratios for subsoil treatments Nos. ¹ and ³ exceeded the critical limit of 0.015 found for pepper primary root growth in subsurface culture solutions at pH values below 6.0; and these subsoil treatments yielded significantly the poorest pepper primary root growths. However, the results of subsoil treatments Nos. 4 and 5 were significantly superior to those of subsoil treatments Nos. ² and ⁶ despite the fact that these latter four subsoil treatments had sub-critical molar ^aH/^aCa ratios.

Significant reduction in pepper primary root growth in the AlCl₃-leached subsoil could not be explained on the basis of the molar activities of Al or on the basis of the molar ^aAl/^aCa ratios. For example, the molar ^aAl/^aCa ratios for all the subsoil treatments exceeded the critical limit of about 0.0005 found for pepper primary root growth in subsurface culture solutions. Yet the results of subsoil treatments Nos. ⁴ and ⁵ were significantly higher than those of subsoil treatment No. ⁶ which had the lowest and the only sub-critical molar activity of 0.09×10^{-5} and the lowest molar ^aAl/^aCa ratio of about 0.0018.

The molar activities of Ca in the subsoil treatments ranged from about 3 to 49 X 10^{-5} M Ca. Near maximum response of pepper primary root growth was obtained at activities of -5 about 407 X 10^{-5} M Ca at pH values of up to 6.0 in subsurface culture solutions. This would readily suggest that pepper primary root growth in the leached subsoil would respond to added Ca. The results of subsoil treatments Nos. ² and 6, that had been treated with 0.70 and 1.40 meg Ca⁺⁺/100 g subsoil respectively, were significantly higher than those of subsoil treatment No. ¹ that had been treated with no Ca and no Mg and those of subsoil treatment No. ³ that had been treated with 0.70 meq Mg⁺⁺/100 g subsoil alone. This would suggest that primary root growth of pepper in the leached subsoil would respond to added Ca but not to added Mg alone. The fact that subsoil treatments Nos. 4 and 5 were distinctly superior to subsoil treatments Nos. ² and ⁶ would further suggest that pepper primary root growth in the leached subsoil was further stimulated by added Mg only after the KC1 extractable A1 in the leached subsoil had been neutralized with CaCO₃.

D. AlCl₂-Leached Merrimac Sandy Loam Subsoil

(i) Lettuce. Significant reduction in lettuce primary root growth in the subsoil could not be explained on the basis of the molar a_H/a_C ratios. For example, the molar a_H/a Ca ratios for subsoil treatments Nos. 1 and 3 exceeded the critical limit of about 0.03 found for lettuce primary

root growth in subsurface culture solutions at pH values below 6.0. Yet, by the seventh day, their results were significantly superior to those of subsoil treatment No. ⁶ which had the smallest sub-critical molar a_H/a_C ratio of 0.004.

Significant reduction in lettuce primary root growth in the subsoil could not be explained on the basis of the molar activities of Al or on the basis of the molar a_{A1}/a_{Ca} ratios. For example, the molar a_{A1}/a_{Ca} ratios for subsoil treatments Nos. 1, 2, 3, and ⁵ exceeded the critical limit of 0.0001 found for lettuce primary root growth in subsurface culture solutions. Yet, by the seventh day, their results were significantly superior to those of subsoil treatment No. 6 which had a sub-critical molar a_{A1}/a_{Ca} ratio of practically zero.

The molar activities of Ca in the subsoil treatments ranged from about 2 to 65 X 10⁻⁵M Ca. Since near maximum response of lettuce primary root growth was obtained in subsurface culture solutions at activities beyond 69 X 10^{-5} M Ca, the subsoil data would suggest that lettuce primary root growth in the leached subsoil should respond to added Ca. Subsoil treatments Nos. 2, 3, and 4, to which variable rates of Ca and/or Mg had been added, significantly outyielded subsoil treatment No. ¹ only by the seventh day. In contrast, subsoil treatment No. 6, which had the highest activity of about 65 X 10 $^{-5}$ M Ca, gave significantly the poorest growth.

This would suggest that while lettuce primary root growth responded positively to neutralization of exchangeable Al in the leached Merrimac sandy loam subsoil by either Ca or Mg, it was susceptible to overliming injury in this leached subsoil.

(ii) Pepper. Significant reduction in pepper primary root growth in the subsoil could not be explained on the basis of the molar a_H/a_C ratios. For example, the molar ^aH/^aCa ratio for subsoil treatment No. 1 exceeded the critical limit of 0.015 found for pepper primary root growth in subsurface culture solutions at pH values below 6.0. Yet its results were not significantly different from those of subsoil treatments Nos. 4, 5, and ⁶ which had sub-critical molar a_H/a_C ratios.

Significant reduction in the primary root growth of pepper in the subsoil could not be explained on the basis of the molar activities of Al or on the basis of the molar a_{A1}/a_{Ca} ratios. For example, the molar a_{A1}/a_{Ca} ratios for subsoil treatments Nos. 1, 2, and 5 exceeded the critical limit of about 0.0005 found for pepper primary root growth in subsurface culture solutions. Yet their results were not significantly different from those of subsoil treatments Nos. 4 and 6 which both had a sub-critical molar a_{A1}/a_{Ca} ratio of practically zero.

The molar activities of Ca in the subsoil treatments ranged from about 2 to 65 X 10^{-5} M Ca. Since near maximum

response of pepper primary root growth was obtained at activities of about 407 X 10^{-5} M Ca at pH values of up to 6.0 in subsurface culture solutions, this would suggest that pepper primary root growth in the leached subsoil should readily respond to added Ca. However, subsoil treatment No. 1, consisting of neither added Ca nor added Mg, gave as good a growth as subsoil treatments Nos. 2, 4, 5, and 6. Subsoil treatments Nos. ⁴ and 5, to which combinations of Ca and Mg had been added, gave the greatest non-significant growth, whereas subsoil treatment No. 3, consisting of only added Mg, gave significantly the poorest growth by the fifth day. This would suggest that neutralization of the exchangeable Al in the leached subsoil by MgCO₃ alone was detrimental to pepper primary root growth in the leached Merrimac sandy loam subsoil.

SUMMARY AND CONCLUSIONS

The differences in penetration of the primary roots of the six crop species into the two acid subsoils could be partly explained on the basis of relative toxicities of ^H and Al ions to the crop species and probable Ca requirements of the crop species in culture solution experiments. Poor primary root growth in these acid subsoils was caused neither by boron deficiency nor by manganese toxicity.

Subsurface Solution Studies on Two Selected Crop Species

^A calcium ion concentration of ¹ ppm was required to obtain near maximum primary root growth of lettuce in subsurface culture solution when other cations were in balance and the primary roots were growing in the absence of toxic ions. In contrast, a Ca ion concentration of ⁷² ppm was required to obtain near maximum primary root growth of pepper under a similar favorable condition.

High concentrations of ^H ions depressed the primary root growth of lettuce and pepper in subsurface culture solutions. The toxicity of ^H ions was not a factor at a solution pH of 6.0 for either crop species; but increased Ca levels were necessary for optimum primary root growth of either crop species as solution pH progressively dropped below 6.0. The interaction between ^H and Ca ions at pH values below
6.0 was a function of the molar activity ratio of the two ions•

Moderate and high concentrations of magnesium ions significantly increased the primary root growth of either crop species in subsurface culture solutions even in the presence of moderate to adequate amounts of Ca. In no instance did addition of increasing amounts of Mg in the presence of Ca result in ^a decrease in the primary root growth of either crop species in subsurface culture solutions.

Heavy liming with $CaCO^2$ plus MgCO₂ coupled with heavy N-P-K fertilization of the surface soil did not alter the response of lettuce and pepper primary root growth to varying Ca ion concentration in subsurface culture solutions. However, it tended to reduce the levels of subsurface solution Ca and/or Mg beyond which significant positive interaction between Ca and Mg ceases to exist for lettuce and pepper primary root growth. In no instance did addition of increasing amounts of Mg in the presence of Ca result in a decrease in the primary root growth of either crop species in subsurface culture solutions.

As regards further stimulation of primary root growth by Mg over and above that due to Ca alone, there was no clearcut relationship with regard to Ca/Ca+Mg or Mg/Ca+Mg in subsurface culture solutions for the two crop species studied.

A moderate concentration of potassium ions significantly increased the primary root growth of lettuce and pepper

in subsurface culture solutions in the presence of somewhat inadequate amounts of Ca. In no instance did addition of increasing amounts of ^K in the presence of Ca result in a decrease in the primary root growth of either crop species in subsurface culture solutions.

Within the range in which significant interactions occurred between Ca and Mg and between Ca and K, the stimulatory effect of Mg on the primary root growth of either crop species was much greater than that of ^K in subsurface culture solutions.

Increasing low concentrations of A1 progressively depressed the primary root growth of lettuce and pepper in subsurface culture solutions. However, lettuce and pepper primary roots growing at the higher levels of Ca were less susceptible to Al damage. This was a function of both the molar activity of Al and ^a molar activity ratio involving Al and Ca.

A small concentration of P was sufficient to significantly depress the primary root elongation of pepper in subsurface culture solutions in the presence of somewhat inadequate and adequate amounts of Ca. However, subsurface lateral roots of pepper increased in number and length with increasing ^P concentration.

Critical Ca concentrations for optimum primary root growth of lettuce and pepper in subsurface culture solutions could, therefore, not be easily defined because of the other

chemical factors beside Ca influencing primary root growth therein. Cognizance should therefore be taken of the stimulation of primary root growth by Mg and ^K in addition to the observed inhibition of primary root growth by H, A1, and possibly ^F in an intricate process of determining critical Ca concentrations for optimum primary root growth of these two crop species in subsurface culture solutions. This tantamounts a striking contrast between these two crop species and cotton and soybean which had been previously studied by other investigators.

Subsoil Studies on Two Selected Crop Species

The response of lettuce and pepper primary root growth to applied $CaCO_3$ and/or MgCO₃ in the acid subsoils could not at all be explained by (a) the concentrations or activities of the individual cations per se, (b) the ratios of the molar activities of ^H to Ca, (c) the ratios of the molar activities of Al to Ca, or (d) the ratios of the molar activities of Ca to all the cations combined. This was largely due to the fact that the major premise of antagonism of cations other than ^H and Al toward Ca on which explanation by such ratios or activities is based was totally false insofar as these two crop species were concerned. Consequently, the differences in the response of lettuce and pepper primary root growth to applied CaCO₃ and/or MgCO₃ in the acid subsoils were attributed to differing chemical factors.

^A significant increase in lettuce primary root growth in the Narragansett loam subsoil was due to Mg being added to a subsoil that already had an adequate amount of Ca in it. ^A significant increase in pepper primary root growth in the same subsoil was a result of neutralization of the exchangeable A1 and the acidity of the subsoil. The absence of a natural deficiency of Ca for lettuce and pepper primary root growth was observed in this subsoil.

No significant increase in the primary root growth of lettuce and pepper was obtained upon adding varying amounts of Ca and/or Mg to the Merrimac sandy loam subsoil, with only one exception. Pepper primary root growth responded significantly to a particular combination of Ca and Mg in excess of that required to neutralize the exchangeable Al and the acidity of this subsoil. The absence of ^a natural deficiency of Ca for lettuce and pepper primary root growth was also observed in this subsoil.

^A significant increase in lettuce primary root growth in the AlCl₃-leached Narragansett loam subsoil was a result of applying either CaCO₃ or a particular combination of CaCO₃ and MgCO₃ in excess of that required to neutralize the exchangeable Al and the acidity of the leached subsoil. ^A significant increase in pepper primary root growth in the same leached subsoil was due to added CaCO₂ at least sufficient to neutralize the exchangeable Al and the acidity of the leached subsoil. ^A further significant increase in pepper primary

root growth was due to added combinations of $CaCO_{3}^-$ and MgCO₃ in excess of that required to neutralize the exchangeable A1 and the acidity of the leached subsoil.

^A significant increase in lettuce primary root growth in the AlCl₃-leached Merrimac sandy loam subsoil was a result of neutralization of the exchangeable Al and the acidity of the subsoil with applied $CaCO^2$ or MgCO₂. Overliming injury to lettuce primary roots was due to excess $CaCO₃$ applied to the leached subsoil. In contrast, a significant decrease in pepper primary root growth was ^a result of neutralization of the exchangeable Al and the acidity of the leached subsoil with applied MgCO₃ alone.

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