INFLUENCE OF PHOSPHORUS, SULFUR, AND MOLYBDENUM FERTILIZATION ON THE SEEDLING VIGOR OF SELECTED LEGUMES ADAPTED TO THE APPALACHIAN REGION

by

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Thesis submitted to the Faculty of the

Virginia Polytechnic Institute and State University

in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

in

Agronomy

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March, 1985

Blacksburg, Virginia

10/15/85 MCR

INFLUENCE OF PHOSPHORUS, SULFUR, AND MOLYBDENUM ON THE SEEDLING VIGOR OF LEGUMES ADAPTED TO THE APPALACHIAN REGION.

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(ABSTRACT)

Legume establishment is difficult on many moderately acid, infertile soils in the humid northeastern United States. Legume seedling vigor as influence by P, S, and Mo fertilization was studied in order to determine fertilizer needs for improved establishment. A Gilpin silt loam was fertilized with 0, 22, 67, and 201 mg P kg^{-1} in combination with 0, 22, 67, and 201 µg Mo kg^{-1} in a greenhouse experiment in 1983. Legumes studied in this experiment were red clover (Trifolium pratense), white clover (T. repens), and birdsfoot trefoil (Lotus corniculatus). Field experiments with red clover, birdsfoot trefoil, and flatpea (Lathyrus sylvestris) were initiated in the spring of 1983 and 1984 using 0, 50, 150, and 450 kg P ha^{-1}, 0 and 60 kg S ha^{-1}, and 0 and 874 g Mo ha^{-1}. Seedling vigor as measured by plant height, trifoliate leaf count, dry weight, and trifoliate leaf area increased the greatest in the greenhouse using 22 mg P kg^{-1} relative to the higher rates of P fertilization. Seedling vigor without applied P was poor, which clearly indicated the essential need for P in the early stages of legume growth. Molybdenum applied at 201 µg kg^{-1} increased the growth of the greenhouse

grown legumes the greatest above the 0 µg Mo kg^{-1} rate within each added P treatment. Seedling vigor however, was not enhanced with Mo fertilization until the P deficiency was corrected. Field established legumes increased in height and dry matter yield the most using 50 kg P ha^{-1} with respect to the additional increments of applied P, but the increase was not as great as that obtained in the greenhouse using 22 mg P kg^{-1}. This was attibuted to a higher extractable P level before fertilization within the field experiments. Sulfur fertilization generally did not enhance seedling vigor in the field studies; although, red clover yield was increased using 60 kg S ha^{-1} in the 1984 field experiment. Molybdenum applied at 874 g ha^{-1} increased seedling vigor as measured by plant height and yield in the field experiments with or without P or S fertilization. Phosphorus uptake and Mo concentrations were increased in the plant tissues with either P or Mo fertilization.

ACKNOWLEDGEMENTS

The author is indebted to Dr. Marcus M. Alley for serving as the committee chairman, and to Dr. Robert J. Wright for serving as co-chairman. Their encouragement, guidance, and friendship has been very beneficial through the course of this study. An expression of gratitude is extended to Dr. Orus L. Bennett for providing funds and facilities through the USDA-ARS, Appalachian Soil and Water Conservation Research Laboratory which made this investigation possible. This gratitude is also extended to Dr. Thomas B. Hutcheson, and Dr. David C. Martens for their time and assistance in serving on the committee.

Special thanks are expressed to Dr. Marvin Lentner and Mr. Walter Winant for their assistance with statistical analysis of data; to Mr. Nick Jones for his counsel during the greenhouse experiment; to Dr. Douglas Perry who advised the design and implementation of the field studies; to Dr. Douglas Boyer for providing rainfall data during the field research; to Dr. Joyce Foster for her time and consideration; to Doris Fuller, Michelle Harmon, and especially Antoinette Green for their concern and patience in typing this manuscript; and to the rest of the specialists and technicians whose assistance was very helpful.

Finally, the author thanks his wife for her constant encouragement and understanding during the course of this research.

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INTRODUCTION

Grasslands of the northeastern United States many times produce low yields and low quality forage. Use of legumes can increase the productivity of grasslands and improve forage quality by supplying companion grass species with nitrogen through N_2-fixation. The success of introducing legumes into pastures depends upon the vigor of the seedlings. Increasing seedling vigor lowers legume mortality, increases the rate of stand development, and usually increases yields. Acid, phosphorus deficient soils are major limiting factors in legume establishment, (Plucknett, 1970), particularly in many northeastern United States grasslands.

Soils in the Appalachian region of the northeastern United States are generally moderately acid, shallow to bedrock, leached, and deficient in essential nutrients such as P, S, and Mo that are required for maximum forage production. Soil acidity reduces microbial activity and may result in low levels of plant available P and S mineralized from the organic soil fraction. Iron and aluminum hydrous oxides also exist under acid soil conditions and can specifically adsorb, or "fix" $H_2PO_4^-$, MoO_4^{2-}, and SO_4^{2-} from the soil solution, rendering them unavailable for plant uptake. Proper legume fertilization can overcome these adverse soil conditions.

Many experiments have been conducted with P, S, and Mo fertilization of legumes. These experiments, however, have focused mainly on total yields, forage quality, and the residual properties of the elements within soil. Depending on the soil and environmental condition, increases in legume yields have been reported with P, S, and Mo applications in many regions. However, until 1971 there had been no recorded crop responses to S fertilization in West Virginia (Beaton et al., 1971), and since then few studies have been conducted with S. Moreover, there has been no research in this region on the effect of P, S, and Mo fertilization on legume seedling vigor with adequate soil K.

Seedling vigor has been correlated with leaf area expansion rate (Shibles and MacDonald, 1962), seedling establishment, and ultimately with forage yields (Qualls and Cooper, 1968). Therefore, it is in the context of this experiment to determine seedling vigor as measured by selected legume growth parameters with respect to P, S, and Mo fertilization.

The objectives of this study were:

1. To measure the influence of P and Mo fertilization on plant height, trifoliate leaf count, trifoliate leaf area, and total dry weight of red clover (Trifolium pratense), white clover (T. repens), and birdsfoot trefoil (Lotus corniculatus) seedlings grown in the greenhouse;

2. To measure the influence of P, S, and Mo fertilization on plant height, and total dry matter yields of red clover, birdsfoot trefoil, and flatpea (Lathyrus sylvestris) seedlings established in the field; and

3. To determine the influence of P, S, and Mo fertilization on legume plant contents of P, S, Mo, and N.

REVIEW OF LITERATURE

Soil Phosphorus

Phosphorus exists in soil as inorganic P compounds, organic P compounds, and soil solution P (Follet et al., 1981). Twenty-five to 90% of total soil P is in organic form and unavailable for plant uptake. Thirty to 50% of organic P compounds have been identified as phytic acid, phytin, nucleic acids, and phospholipids with the remainder being "humus." Nucleic acids account for 3% of total soil P and this is the material most readily decomposed, or "mineralized" by microorganisms to inorganic, plant available P compounds. Only small quantities of available P result from mineralization in most soils (Alexander, 1977; Follet et al., 1981).

Inorganic plant available forms of soil P are the primary and secondary orthophosphate anions, $H_2PO_4^-$ and HPO_4^{2-} (Follett et al., 1981). According to Barrow (1978), the pK 1 for H_3PO_4 is approximately 2 and the pK 2 is about 7. Therefore, most plant available P occurs as $H_2PO_4^-$ in acid soils with the proportion of HPO_4^{2-} in solution increasing as pH increases to neutrality. Moderately acid soils in high rainfall areas adsorb anions from soil solution (Barrow, 1978). Oxyanions such as PO_4^{3-} are subject to slow adsorption reactions that decrease their long-term availability. Adsorption occurs in two steps: (1) a rapid initial stage of one day or less, which is a combination of nonspecific adsorption and ligand exchange on mineral edges; and (2) a slow reaction (weeks) which probably consists of a complex combination of mineral dissolution and/or precipitation of added P with exchange sites or mineral lattice cations (Bohn, 1979).

Iron and Al hydroxides and oxides along with kaolinite silicate clay are minerals which adsorb soil solution P. The Fe and Al hydrous-oxides exist in moderately acid soils and tend to be the dominant minerals adsorbing large amounts of added P from soil solution. The surfaces of these minerals contain metal atoms that are not fully coordinated and may complete their shell with OH^- groups or water molecules. The presence of H^+ ions favor the conversion of some of the OH^- groups to water thus leading to a net positive charge (Barrow, 1978). Barrow (1978) also explained that as anion adsorption continues, the rate of adsorption decreases and the surfaces become more negative. Additional fertilizer applications of P may overcome soil adsorption or "buffer capacity" and result in large quantities of P in solution. Phosphorus in soil solution may be readily absorbed by plant roots or lost by leaching. Soil adsorption of P restricts leaching losses, and thus retains P in the root zone. Adsorbed P may be slowly released as P is absorbed from soil solution by plants (Barrow, 1978; Mengel and Kirkby, 1978).

Increased forage production requires P fertilization of most soils (Brady, 1974). As stated by Mengel and Kirkby (1978), soil buffering capacity can determine the quantity of fertilizer P needed for optimum P availability. Levels of available P in soils can be evaluated by methods such as double-acid (HCl + H_2SO_4) and Bray-1 (HCl + NH_4F) (Mehlich, 1953; Olsen and Sommers, 1982). Values obtained from these soil tests are used as an index of P availability which, when related to crop growth makes it possible to estimate a fertilizer rate that is required for optimum plant growth (Kroontje, 1981).

Legumes and Phosphorus

"Phosphorus is essential for plant life" (Wallingford, 1978a). The most important compounds containing P within plants are nucleic acids, phospholipids, adenosine triphosphate (ATP), and the coenzymes nicotinamide adenine dinucleotide (NAD), and nicotinamide adenine dinucleotide phosphate (NADP). These compounds serve a vital role in photosynthesis, protein synthesis, fat metabolism, cell nucleus formation, cell division and energy transfer. Oxidation-reduction reactions in plant cells depend on NAD and NADP (Rhykerd and Overdahl, 1972; Schreiber, 1978). Phosphorus is the most important nutrient in starter fertilizers. Young rapidly growing plants need large quantities of P for energy production and transfer and older plants can not reach their maximum growth potential or complete their reproductive process if P is deficient (Wallingford, 1978b). According to Mays et al., (1980) low native soil P combined with low past P applications have resulted in P being the most yield limiting factor in legume production. Phosphorus deficient legumes have also exhibited depressed nodulation and seedling vigor and growth. Reports by Charlton and Brock (1980), and Shoop et al., (1961) indicated that P fertilization increased seedling vigor as measured by plant height and establishment. Normal root development also requires adequate available P (Shreiber, 1978).

Alfalfa (Medicago sativa), soybean (Glycine max), white clover (Trifolium repens), red clover (T. pratense), and subterranean clover (T. subterrane) yields have been increased with P fertilization (Asher and Loneragan, 1967; Hart et al., 1981; Holford and Gleeson, 1976; Parsons and Davis, 1960; Sherrell and Saunders, 1974). Phosphorus applications of 50 kg ha^{-1} or greater increased alfalfa yields by 20% on a Fredrick silt loam originally containing 8 mg P kg^{-1} of double acid extractable P (Lutz, 1973). McLean and Ssali (1977) observed a 400% increase in alfalfa yields during a growth chamber study using 37 kg P ha^{-1} on a moderately acid loam which originally had 2.7 mg P kg^{-1} of Bray-1 extractable P. They did not obtain a yield increase from a soil with a Bray-1 level of 12.5 mg P kg^{-1}. Kamprath and Miller (1958) reported that soybean dry weight increased 300% on a Coastal Plain soil when the double acid extractable P concentration was raised to 30 mg P kg^{-1}. Double acid extractable P before treatment ranged from 4 to 10 mg P kg^{-1}. A 24% increase in soybean yield with P fertilization was observed by Jones et al. (1977) on a soil with only 15 kg P ha^{-1} of double acid extractable P. White clover dry weight increased from 500 to 900 kg ha^{-1} with 200 kg P ha^{-1} on a soil originally containing 2 to 8 mg P kg^{-1} of Olsen extractable P in New Zealand (Jackman and Mouat, 1972). Haynes and Ludecke (1981) also reported a 700% increase in white clover yields using 350 kg P ha^{-1}, while DeRuiter (1981) observed a 3-fold increase in subterranean clover yields with 50 kg P ha^{-1} on soils known to be P deficient in New Zealand.

Phosphorus concentrations in plant tissue can vary among legume species. Mays et al. (1980) observed that the critical P concentration of some tropical legumes ranged from 0.12 to 0.30%. The critical P concentration in white clover was approximately 0.30% in experiments conducted by Haynes and Ludecke (1981) and Rayment (1970). Martin and Matocha (1973) recommended that the critical P concentration of both white and red clover be established as 0.25%. Tissue P concentrations for white clover and lotus (Lotus pedunculatus) increased from 0.25% to approximately 0.50% when fertilizer P was increased from 0 to 350 kg ha^{-1} on a P deficient soil in New Zealand (Haynes and Ludecke, 1981). Red clover P concentration increased from 0.30 to 0.71% P as fertilizer P was applied to an Egmont sandy loam in increasing rates from 50 to 2000 mg kg^{-1} (Hart, 1981). Fisher and Campbell (1972) observed that a P concentration greater than 0.12% in a tropical townsville stylo (Stylosanthes humilis) legume was associated with increased yields. A P concentration of 0.17% in alfalfa was deficient according to Lutz (1973) and increasing fertilizer P from 0 to 100 kg ha^{-1} increased percent P in alfalfa from 0.17 to 0.34%.

Available P stimulates nodule bacteria in the soil rhizosphere. Phosphorus has a direct effect on nodule size, formation, and number. Effective symbiosis is also increased by adequate P (Griffith, 1978). Jones et al. (1977) observed that applications of P increased soybean nodulation and pod formation. Percent total N increased in white clover from 3.01 to 3.29% when fertilizer P was increased from 50 to 350 kg ha^{-1} (Haynes and Ludecke, 1981). Percent N in red clover, white clover, alfalfa, townsville stylo, and lotus has also been increased with fertilizer P (Lutz, 1973; Hart et al., 1981; and Shaw et al., 1966). It has also been shown that percent N in clover does not increase with applied P when plant available P is not initially deficient (McLachlan and Norman, 1961).

Soil Sulfur

Organic sulfur, inorganic sulfide and sulfate are three major forms of soil S. It is estimated that the organic fraction comprises approximately 95% of the total which is unavailable for plant uptake (Follett et al., 1981; Tabatabai and Bremner, 1972). Inorganic SO_2 is the S product of bacterial mineralization of organic matter. Sulfide can be oxidized by Thiobacillus microorganisms in well aerated soils to produce SO_4^{2-} which is available for plant uptake but SO_4^{2-} can also be reduced in waterlogged soils. Microorganisms utilize the majority of SO_4^{2-} with the remainder being available for uptake by higher plants (Alexander, 1981).

According to Harward and Reisenauer (1966) SO_4^{2-} may account for less than 10% of total soil S in most surface soils. Sulfate can be retained in acid soils through specific anion adsorption by Fe and Al oxides. The strength of retention is lower than that of PO_4^{3-} and MoO_4^{2-} because the pK 1 and pK 2 of H_2SO_4 is approximately 1.7 and 1.9, respectively (Barrow, 1978). In strongly acid soils, however, SO_4^{2-} is more highly adsorbed than $H_2PO_4^{2-}$ (Barrow, 1970). But, in moderately acid soils, P fertilization decreases the amount of adsorbed SO_4^{2-} (Barrow, 1969). Jordan and Bardsley (1958), and Harward and Reisenauer (1966) reported that leached SO_4^{2-} generally accumulates in lower soil horizons with adsorption by Fe and Al oxides where it is less affected by the more strongly adsorbed anions. Mengel and Kirkby (1978) suggest that fertilizer S should thus be applied in the spring for early forage growth due to SO_4^{2-} losses by leaching.

Sulfur deficiencies have been increasing throughout the world (Platou and Irish, 1982). Less S as an impurity in high analysis fertilizers and pesticides has contributed to the reduction of soil S. Increased crop yields have increased the need for S. Restrictions on SO_2 emissions from smoke stacks in industrial areas, and less use of farmyard manure has also reduced soil SO_4^{2-} levels (Beaton et al., 1971; Elkins and Ensminger, 1971; Platou and Irish, 1982) According to Westermann (1974) critical SO_4^{2-} levels in soils for plant growth were below 3.0 to 4.0 mg kg^{-1} (water, $LiCl_2$, or KH_2PO_4 extraction methods). Walker and Doornenbal (1972) reported that podzolic and chernozemic soils with less than 2 mg kg^{-1} of SO_4^{2-} were deficient using a 0.1 M $CaCl_2$ extractant. Organic or total soil S measurements have not proven to be satisfactory indexes of S availability according to Westermann (1975).

Legumes and Sulfur

Plants generally require about as much S as they do P for adequate growth (Tisdale, 1977). Sulfur has been shown to be a constituent of certain amino acids (cysteine, methionine, and cystine) and some vitamins (Rhykerd and Overdahl, 1972). Additions of S to plants deficient in S can increase vitamin A, chlorophyll, and protein contents. Also, protein quality has improved, along with a reduction in N:S ratio, and non-protein nitrogen content of grasses with S applications to S deficient soils. On S deficient soils, protein formation is retarded and upper plant leaves become light yellow resulting in an overall stunting of growth and delayed maturity in many instances. Plant leaves may also become long and slender, abnormal branching can occur, and stands can become thin (Rhykerd and Overdahl, 1972).

Sulfur additions have increased crop yields in many studies on S deficient soils. During and Cooper (1973) observed a white clover yield increase of 390 kg ha^{-1} using 45 kg S ha^{-1} on a sandy loam soil in New Zealand originally containing 14 mg kg^{-1} KH_2PO_4 + $CaCl_2$ extractable SO_4^{2-}. However, Scott et al., (1983) received only a 4% increase in white clover yields when 40 kg S ha^{-1} was applied to a loam soil originally containing 6.0 mg kg^{-1} available SO_4^{2-} (KH_2PO_4 extractant). Subterranean clover yield increased 3.6 times when a S deficient Cole loam was treated with 56 kg S ha^{-1} in California (Jones, 1962). In a later experiment conducted by Jones (1964), subterranean clover yield increased 440% on a gravelly loam soil two years after an initial application of 90 kg S ha^{-1}.

Pumphrey and Moore (1956b) observed that alfalfa yield increases taken from the first cutting ranged from none to 350% using 56 kg S ha^{-1} on soils typical of Oregon. Yields increased on 20 of the 28 soils used in the experiment. Alfalfa yields were also increased using 67 kg S ha^{-1} on many soils within the mountain valleys of Idaho. These soils originally contained 3 mg kg^{-1} or less available SO_4^{2-} as determined by the water, $LiCl_2$, or KH_2PO_4 extraction methods (Westermann, 1974, 1975).

Sufficiency levels of plant S are different for individual legume species. Martin and Matocha (1973) indicated that red clover was S deficient if total plant S concentration was less than 0.20%, but they did not differentiate maturity stages. Walker and Bentley (1961) observed that red clover harvested between 10% bloom and full bloom containing 0.11% total S was S deficient. An application of 22.4 kg S ha^{-1} increased red clover S contents to 0.19% but there were no further yield responses to greater S additions in this particular study. Greenhouse-grown white clover plants were S deficient with 0.10% total S, while levels of 0.10 to 0.16% total S were found in S deficient field-grown plants (Jordan and Bardsley, 1958). An application of 36 kg S ha^{-1} increased S contents of field grown plants to 0.21% and alleviated the S deficiency in these experiments. Greenhouse studies with soybeans growing on a Dothan clay subsoil showed that an application of 8 mg S kg^{-1} increased total plant S from 0.15 to 0.18% when harvested three weeks after germination (Elkins and Ensminger, 1971). Westermann (1975) reported maximum alfalfa yields on many S deficient soils in Idaho when total S contents ranged between 0.15 and 0.21% for plants harvested at early bloom. Moreover, Seim et al. (1969) observed that total S levels in alfalfa greater than 0.30% were associated with maximum yields on a Dorset sandy loam; 0.12 and 0.15% total S contents at 10% bloom were considered critical concentrations.

Sulfur fertilization has increased total N in plants growing on S deficient soils. Total N in red clover harvested between 10% and full bloom increased from 2.46 to 3.20% with a S application of 22.4 kg ha^{-1} (Walker and Bentley, 1961). Pumphrey and Moore (1965a) working on S deficient soils in Oregon reported a 2% increase in total plant N of alfalfa harvested at 25% bloom when S was applied at 44 kg ha^{-1}. Alfalfa harvested at 10% bloom exhibited an increase in total N from 2.80 to 3.90% using either 38 or 76 kg S ha^{-1} on a Delhi sand in California (Rendig, 1956).

Sulfur is similar to N in its effect on percent protein, and therefore changes in S contents can influence forage feeding values (Rhykerd and Overdahl, 1972). Jones et al. (1982) reported that when lambs were fed alfalfa hay containing 0.25% S, better gains were observed than when fed alfalfa with 0.14% S. Ruminant animals utilize forage more efficiently when N:S ratios range between 15 to 20 (Coleman, 1966).

Total N:S ratios have been used to determine S deficiencies in plants (Westermann, 1975). Depending on the crop, a N:S ratio range of 14 to 16 has been associated with optimum plant growth if plant N is adequate (Tisdale, 1977). However, Westermann (1975) observed that an alfalfa N:S ratio of 12 was associated with maximum yields in his studies, while Sheard (1978) stated that a N:S ratio less than 16 was adequate for white clover.

Soil Molybdenum

Molybdenum is the least abundant micronutrient in soil. Total soil Mo generally ranges between 0.2 and 10 $\mu g\ g^{-1}$ (Follet et al., 1981). Total soil Mo is accountable as: (1) Mo in solution; (2) Mo adsorbed by soil colloids; (3) Mo within crystal latices; and (4) Mo in organic matter. Available Mo includes both the adsorbed and water soluble fractions, with adsorbed Mo representing the largest amount. Unavailable Mo comprises 95% of the total soil Mo (Gupta and Lipsett, 1981; Reisenauer et al., 1962). Barrow (1978) and Bohn et al., (1979), stated that MoO_4^{2-}, like PO_4^{3-} and SO_4^{2-}, can be converted from a readily available form to a less available form by specific anion adsorption by Fe and Al hydrous oxides in acid soils. The pK 1 for H_2MoO_4 and the pK 2 of $HMoO_4^-$ are both approximately 4. Therefore, as soil pH increases the dominant Mo anion exists as soluble MoO_4^{2-} (Barrow, 1978). Davies (1956) and Coleman et al., (1956), concluded that liming acid soils increases MoO_4^{2-} availability. However, if total soil Mo level is low, lime alone will not increase MoO_4^{-2} availability (Gupta and Lipsett, 1981).

Total soil Mo depends to a large extent on the parent material of a particular soil. Soils developed from sandstones, which have undergone extreme weathering and leaching, may be low in total soil Mo (Gupta and Lipsett, 1981). Australian soils derived from lateritic remnants, granite, quartzites, shale, and slate tend to have low total Mo levels according to Davies (1956). Granite, shale, limestone, and argillaceous shist parent material can weather to soils high in total Mo (Gupta and Lipsett, 1981). Soils derived from calcareous parent material can be low in total Mo, however, if the soil was formed during early acid leaching (Rubins, 1956). According to Kubota (1977), United States soils generally decrease in total Mo from west to east. Basic soils of low rainfall areas in the west average 6.0 $\mu g\ g^{-1}$ total Mo, whereas eastern acidic soils average 0.5 $\mu g\ g^{-1}$ total Mo.

Low levels of available soil Mo are often found in acid leached soils in humid regions (Kubota, 1977; Rubins, 1956). Seventy-five soil series typical of West Virginia were analyzed by Stone and Jencks (1963) for available Mo using an Aspergillus niger procedure. Available Mo averaged 0.14 $\mu g\ g^{-1}$ for all soils with a below average Mo concentration of 0.07 $\mu g\ g^{-1}$ in soils derived from sandstone and shale. An above average Mo value of 0.17 $\mu g\ g^{-1}$ was associated with soils formed from limestone. Karimian and Cox (1978) tested 32 soils from the Atlantic Coastal Plain and Piedmont regions for available Mo using an anion exchange resin procedure. All soils were low in available Mo and the Mo levels were highly correlated with soil pH. Soils having a low pH contained lower levels of

Mo due to increased anion adsorbing capacity. For example, a Pungo (Typic Medisaprist) soil having a pH of 3.4 contained on available Mo level of 0.6 μg kg^{-1}, while a Whitestore (Vertic Hapludalf) soil with a pH of 5.8 had an available Mo content of 11.1 μg kg^{-1}. Overall, the available Mo levels for these 32 soils normally ranged between 4.0 and 7.0 μg kg^{-1} with soil pH varying from 4.9 to 5.8. These values obtained by Karimian and Cox were considerably lower than the available Mo levels reported by Stone and Jencks (1963). According to Bhella and Dawson (1972), anion exchange resins produced more accurate available Mo levels than the Aspergillus niger procedure. Bache and Ireland (1980) reported that ion exchange resins closely simulated the desorbing effect of plant roots, and their experiments support the anion exchange resin procedure for measuring available Mo.

Legumes and Molybdenum

Legume response to Mo fertilization has been shown to be due to increases in symbiotic N_2-fixation (Anderson, 1956a). Legume root nodules contain the nitrogenase enzyme that requires Mo for the reduction of atmospheric N_2 to ammonia (Price et al., 1972). Molybdenum is utilized within the electron trasfer system in the nitrogenase enzyme as an electron carrier for N_2-fixation (Gupta and Lipsett, 1981). Anderson and Spencer (1950) observed that symbiotic N_2-fixation was inhibited when plant available Mo was deficient. Evans (1956) reported that N_2-fixation increased in red clover grown in a sand culture with Mo fertilization. Molybdenum deficient legumes may exhibit stunted growth, pale green or yellow leaves, and chlorotic interveinal tissue (Anderson, 1956a; Anderson 1956b; Gupta and Lipsett, 1981; Reisenauer, 1956). Nodules of Mo deficient legumes can be grey or brown in color, and although nodule population may be high, they are usually small and ineffective (Anderson, 1956a). Adequate Mo has been shown to increase nodule size, and decrease nodule population (Anderson and Spencer, 1950). Mortvedt (1981) also observed large nodules when Mo was supplied at 5 mg pot^{-1} to red, white, crimson, and subterranean clover grown on a Decatur silty clay (Rhodic Paleudult) in the greenhouse. Bloomfield (1954) reported that large, pink nodules were necessary for effective N_2 reduction.

Since Mo enhances the N_2-fixation process, total plant N would be expected to increase with Mo fertilization if available soil Mo is low. Reisenauer (1956) observed that total N in alfalfa leaves increased from 2.77 to 3.03% when Mo fertilization increased from 0 to 900 g ha^{-1}, respectively. Hagstrom and Berger (1963) reported that total N in soybeans increased frm 2.10 to 2.61% from an application of 874 g Mo ha^{-1}. Subterranean clover N increased to 3.21% from 2.00% with 109 g Mo ha^{-1} (Anderson and Spencer, 1950). Mortvedt (1981) showed that N contents in red and white clover increased 1.07 and 1.32-fold, respectively, with fertilizer Mo applied at 3.7 kg Mo ha^{-1}.

Lime or Mo applications are sometimes necessary to correct Mo deficiencies on acid soils. Molybdenum fertilization according to Petrie and Jackson (1982) can reduce or eliminate the need for lime. Moreover, Gupta and Lipsett (1981) stated that if total soil Mo is low, lime alone could not increase MoO_4^{2-} availability. Anderson (1956b) however, reported that legume yield increases to applied Mo occur only after correcting soil P and S deficiencies. Once the P and S requirements were met, Gupta and Lipsett (1981) observed that yield responses to Mo would begin at an early growth stage. Studies by de Mooy (1970) showed that soybean yield increased when Mo was added at a rate of 230 g per 67 kg seed planted in two acid Iowa soils. Soybean yield did not increase on two alkaline soils when Mo was applied at the same rate. Alfalfa yields increased on 9 of 18 acid to neutral soils in Indiana when Mo was applied at 2.24 kg ha^{-1} (Foy and Barber, 1959). Reisenauer (1956) reported that alfalfa yield increased from 2,374 to 3,113 kg ha^{-1} with fertilizer Mo application of up to 1.8 kg ha^{-1} on an acid Washington Couse silt loam. Red clover yields increased by 65% on an acid Wisconsin soil when sodium molybdate was applied at 2.24 kg ha^{-1} (Hagstrom and Berger, 1963) while Gupta et al. (1978) observed a 10% increase in red clover yield with a rate of 10 kg Mo ha^{-1} on an acid peat soil in Canada. No further increase in yield occurred from higher rates of Mo. Hawes et al (1976) reported that red clover yield on a Maury silt loam (pH 6.4) in Kentucky increased 20% with only 0.22 kg Mo ha^{-1}. Increasing fertilizer Mo to 2.64 kg ha^{-1} did not result in increased yields.

Total plant Mo concentrations less than 0.10 μg g^{-1} are normally considered Mo deficient (Anderson, 1956a; Jones, 1972). However, critical Mo concentrations in plant tissue differs between legume species. Hawes et al. (1976) observed that red clover Mo contents increased from 0.26 to 3.16 μg g^{-1} (flowering stage) when fertilizer Mo was increased from 0 to 2.64 kg ha^{-1}. Red clover Mo at early bloom increased from 0.10 tO 2.34 μg g^{-1} when fertilizer Mo was increased from 0 to 0.87 kg ha^{-1}, respectively, on an acid Wisconsin soil (Hagstrom and Berger, 1963). Gupta (1970) reported that red clover plants at bud stage were Mo deficient when tissue concentrations were less than 0.20 μg g^{-1}. In the same experiment, red clover deficiencies were corrected when plants contained 0.46 μg Mo g^{-1}. White clover Mo uptake increased 28-fold when Mo was applied at 3.7 kg ha^{-1} to an acid Decatur silty clay according to Mortvedt (1981). According to Reisenauer (1956), 0.5 μg Mo g^{-1} in alfalfa tissue was adequate for maximum yields. This agrees with James et al, (1968) whose research showed that alfalfa yields were not increased when plant Mo concentrations exceeded 0.3 to 0.5 μg g^{-1}. However, Gupta (1970) suggested that the optimum Mo content in alfalfa at 10% bloom was between 0.12 and 1.29 μg g^{-1}. In contrast to the deficiencies, plants may contain several hundred parts per million Mo without showing obvious damage (Gupta and Lipsett, 1981).

Legume Fertilzation with Phosphorus, Sulfur and Molybdenum

Increases in legumes growth have been observed with fertilizer combinations of P + S, P + Mo, S + Mo, and P + S + Mo. Douglas and Risk (1981) reported that red, white, and alsike (T. hybridum) clovers increased in vigor with increasing additions of P (22.5, 45, and 90 kg ha^{-1}) in combination with 56 kg S ha^{-1} and 140 g Mo ha^{-1}. Clover vigor was assessed with percent ground cover measurements and visual observations. Clover plant vigor was 2 to 3 times greater with P + S or P + S + Mo fertilization than with only P or P + Mo applications. Jones and Ruckman (1973) observed a subterranean clover-grass pasture yield increase of 2000 kg ha^{-1} when P was applied at 112 kg ha on a Pinole loam originally containing 5.1 mg P kg^{-1} ($NaHCO_3$ extraction) and 8.8 mg kg^{-1} available SO_4^{2-}. Sulfur applied alone at 112 kg ha^{-1} did not increase yield. Molybdenum fertilizaton at 282 g ha^{-1} without added P or S increased yields 500 kg ha^{-1}. However, yield increased 4500 kg ha^{-1} to a maximum of 6500 kg ha^{-1} with applications of either P + S or P + S + Mo. McLachlin (1955) treated seven soils of sedimentary origin with 336 kg P ha^{-1}, 112 kg S ha^{-1}, and 62 g Mo ha^{-1}. He observed that subterranean clover yields increased 350% with applied P, an additional 113% using P + S applications, and 35% further with P + S + Mo fertilization. Walker et al. (1955) working in New Zealand also reported similar trends using P + S, and Mo fertilization on red, alsike, and subterranean clover mixtures. Shulka and Pathak (1973) observed that berseem (T. alexandrinum), a warm temperate legume native to the Near East, was increased in yields the greatest using a fertilizer combination of 50 kg P ha^{-1}, and 0.5 kg Mo ha^{-1} on selected acid soils from the district of Naintal, U. P. However, Blackmore et al. (1978) observed that white clover yields on six soils in northern New Zealand increased more with 45 kg P ha^{-1} and 56 kg S ha^{-1}. White clover yield was increased on one soil with an application of 0.14 kg Mo ha^{-1}. Drlica and Jackson (1979) also showed that subterranean clover yield increased by an average of 22% with a combined application of 30 kg P ha^{-1} and 44 kg S ha^{-1} on an Oakland and Nonpareil soil series in Oregon. However, P fertilization without applied S did not increase yields. Bray-1 extractable P was approximately 10 mg kg^{-1} before P was applied to these two soils.

Legume Mo concentration has increased from both Mo and P fertilization (Shulka and Pathak, 1973). Singh and Kumar (1979) supported the fact that P fertilization can increase Mo uptake by forming a readily available phosphomolybdate anion. Sulfur fertilization, however, can decrease legume Mo concentrations. Reasons for the antagonistic effect could be that SO_4^{2-} and MoO_4^{2-} have the same charge and are approximately the same size. Therefore, SO_4^{2-} and MoO_4^{2-} can compete for absorbing sites on plant roots (Kumar and Singh, 1980; Gupta and Munro, 1969; Stout et al., 1951). Indirect evidence of this antagonism is shown in work by Jones and Ruckman (1973) who observed a subterranean clover Mo concentration of 6.5 mg kg^{-1} when fertilizer Mo was applied to a soil at 282 g ha^{-1}. When S was applied at 112 kg ha^{-1}, even with Mo, plant Mo concentration decreased to less than 1.0 mg kg^{-1}. Reisenauer (1963), fertilized field peas (*Pisum sativum*) with 1.12 and 2.24 kg Mo ha^{-1} and 0, 22.4, 67.2, and 89.6 kg S ha^{-1}. He observed a 100% reduction in plant Mo with increasing S fertilization. Singh and Kumar (1979) reported similar trends in decreased Mo concentrations of soybeans with increasing S fertilization. Kumar and Singh (1980) however, reported that the antagonistic effect between SO_4^{2-} and MoO_4^{2-} for uptake is overcome when available soil P is adequate.

MATERIALS AND METHODS

Experiment 1. Legume Seedling Response to P and Mo Fertilization in the Greenhouse.

Seedling growth of red clover, white clover, and birdsfoot trefoil was evaluated in an acid (pH 4.9), low fertility Gilpin silt loam (Table 1). The soils were was treated with four rates of P (0, 22, 67, and 201 mg P kg^{-1} as $CaH_2PO_4 \cdot H_2O$) and four rates of Mo (0, 22, 67, and 201 µg Mo ha^{-1} as $(NH_4)_6Mo_7O_{24} \cdot 4\ H_2O$) in a factorial combination. A completely randomized design with four replications was used for each species in the greenhouse.

Plastic bags were filled with 915 g of soil (800 g air dry) which had been passed through a 2 mm mesh screen. All soils received 10 ml of 68 mM K_2SO_4 which supplied 67 mg K kg^{-1} and 27 mg S kg^{-1}. Phosphorus was applied to the soils using 10 ml of 57 mM, 172 mM, and 518 mM Ca $H_2PO_4 \cdot H_2O$ to produce the 22, 67, and 201 mg P kg^{-1} treatments. Molybdenum was applied using 10 ml of 0.0027 mM, 0.008 mM, and 0.024 mM $(NH_4)_6MO_7O_4 \cdot 4\ H_2O$ to obtain the specified Mo rates. Soils were thoroughly mixed following treatment applications and allowed to equilibrate.

Each pot received 20 inoculated legume seeds planted at a depth of 5 mm on 17 March, 1983. The legumes were watered with deionized water to a total pot weight of 1067 g (90% water holding capacity) and randomly rotated on a table daily after seedling emergence, (21 March 1983). Red clover was thinned to six plants per pot on 29

Table 1. Chemical properties before fertilization of soils utilized in the greenhouse and field experiments.

Classification	Gilpin, fine loamy, mixed, mesic, Typic Hapludult.		
Texture	Silt loam		
	Greenhouse	Field	
pH	4.9	5.0	
Ca	3.1	3.0	cmol (+) kg^{-1}
Mg	0.28	0.22	cmol (+) kg^{-1}
K	0.26	0.26	cmol (+) kg^{-1}
Bray-1 Extractable P	4.0	12.5	mg kg^{-1}
Resin Extractable Mo	12.0	16.0	µg kg^{-1}

March 1983 when the first true leaf developed. White clover and birdsfoot trefoil were not thinned. These treatments therefore contained 10 to 20 plants per pot. Red clover height measurements and trifoliate leaf counts were recorded from four plants per pot 1 week after the first true leaf developed, 16 days after emergence. Subsequent measurements were made weekly for 8 weeks. White clover and birdsfoot trefoil height was recorded from three plants per pot once every 2 weeks for 8 weeks beginning 23 days after emergence.

All legumes were harvested 26 May 1983. Red and white clover trifoliate leaf area was measured by randomly cutting six trifoliate leaves per pot and placing them through a LI-3000 leaf area meter. Leaf area was not determined for birdsfoot trefoil. Total dry weight was measured after oven drying at 50°C for 48 hours. Plant samples were prepared for analysis by grinding in a Cyclone Sample Mill with stainless steel blades. Soil samples were prepared for analysis by air drying and grinding to pass a 2 mm mesh sieve.

Plant Tissue Analysis

Plant tissue samples were digested for P, Ca, Mg, and K analysis as described by Koch and McMeekin (1924). Seven milliliters of concentrated H_2SO_4 were added to a 0.25 g oven-dry plant sample in a digestion tube. This was allowed to digest for 12 hours after which four milliliters of H_2O_2 were added to each sample, and the samples were then heated at 380°C until the solutions turned clear. The samples were diluted to a total volume of 25 ml with deionized water.

Phosphorus in the digests was determined by using the molybdate-vanadate procedure described by Olsen and Sommers (1982). Absorbance was read on a Bausch and Lomb spectrophotometer 21 at 400 nm.

Concentrations of Ca^{2+}, Mg^{2+}, and K^{+} in the digests were determined by diluting a 0.1 ml aliquot with 4.9 ml of a 10% lanthium-oxide solution. Atomic absorption analysis was utilized for Ca^{2+} and Mg^{2+} determinations, and flame emission analysis was employed for K^{+}.

Total plant S was determined on a Leco S Determinator SC13 by the standard procedure associated with the instrument (Leco Corp. 1981). Total plant N was analyzed on a Leco C, H, N-600 Analyser as described by Leco Corp. (1984). An approximately 0.14 g plant tissue sample was used in the determination.

A 1.0 g plant tissue sample for Mo analysis was dry ashed at 400°C in a muffle furnace for 24 hours. The ashed samples were dissolved in 5.0 ml concentrated HCl and 15 ml deionized H_2O with gentle heating on a hot plate. The dissolved samples were filtered through Whatman #42 filter paper into 50 ml volumetric flasks and the dissolution vessels were double-rinsed with 10 ml of deionized H_2O. Molybdenum in the digests was determined with a modified thiocyanate procedure (Carel and Wimberly, 1982). Sodium tartrate (50 mg), 2.0 ml of ferrous ammonium sulfate, and 2.0 ml of 10% potassium thiocyanate were added to each sample in the order listed. The samples were mixed by swirling and allowed to stand 15 minutes. Each sample recieved 2.0 ml of a 10% $SnCl_2$ solution, were mixed and allowed to stand 15 minutes after which 4.0 ml of isoamyl alcohol was added. The samples were shaken for 2 minutes and allowed to stand 20 minutes for separation. The separate organic layer was pipetted into test tubes; 100 mg of anhydrous sodium sulfate was added to each sample followed by centrifuging at 1000 rpm for 10 minutes. Absorbance was monitored at 465 nm on a spectrophotometer. A blank and a National Bureau of Standards plant tissue standard were processed with each group of samples.

Soil Analysis

Soil pH was determined with a pH meter and glass electrode using a 1:1 soil to water slurry after a 20 minute equilibration. Plant available P was extracted by using the Bray-1 method (Olsen and Sommers, 1982). Total soil S was analyzed using the Leco S Determinator SC12 (Leco Corp. 1981).

Extractable soil Mo was determined by adding 25 g of soil and 100 ml of deionized H_2O to a 250 ml Erlenmeyer flask. A 60 mesh polyester net bag containing 20 meq. of a Cl^- form anion exchange resin (Fisher Certified Rexyn 210) was added to the flasks and shaken for 24 hours at 200 rpm (Wright and Hossner, 1984). The resin bags were removed from the flasks, rinsed with deionized H_2O to remove the soil, placed back into the empty 250 ml Erlenmeyer flasks, and 40.0 ml of 0.5 M NaCl was added to each sample. The flasks were shaken for 2 hours at 200 rpm, the resin bags removed, and the solution filtered through Whatman #42 filter paper. The resin bags were washed with 10 ml of 0.5 M NaCl and 10 ml of deionized water to be certain that the adsorbed Mo was released from the resin. The wash solutions were filtered into the initial filtrate and the entire filtrate was evaporated to dryness on a hotplate. The residues were heated in a muffle furnace at 500°C for 12 hours and then dissolved in 5 ml concentrated HCl and 15 ml of deionized H_2O with gentle heating on a hotplate. Molybdenum in the digest was determined by the modified thiocyanate procedure described previously by Carel and Wimberley (1982).

Exchangeable Ca^{2+}, Mg^{2+}, and K^+ were extracted using a 1 M ammonium acetate (pH 7.0) extracting solution as described by Thomas (1982). Calcium and Mg^{2+} in solution were analyzed by atomic absorption, and K^+ by flame emission.

Experiment 2. Legume Seedling Response to P, S, and Mo Fertilization in the 1983 Field Experiment.

Seedling growth of red clover, birdsfoot trefoil, and flatpea in response to P, S, and Mo fertilization was evaluated in an acid (pH 5.0), low fertility Gilpin silt loam (Table 1) in southern West Virginia. Each legume species was treated with four rates of P (0, 50, 150, and 450 kg P ha^{-1} as triple superphosphate), two rates of S (0 and 60 kg S ha^{-1} as gypsum), and two rates of Mo (0 and 874 g Mo ha^{-1} as sodium molybdate) in a factorial combination. A split block design with four replications was used resulting in 16 treatments for each legume and replication. The main plot (legume specie) dimension was 17.1 x 9.7 m and the sub-plots were 2.4 x 4.3 m in size. No comparisons were made between legume species.

Glyphosate was sprayed at 9.35 l ha^{-1} to kill the existing vegetation before seeding and insects were controlled by spraying with Furidan at 9.35 l ha^{-1}. Potassium chloride was applied to the entire experimental area at a rate of 112 kg K ha^{-1}. Triple superphosphate was applied at 268, 804, and 2414 g per sub-plot to obtain the 50, 150, and 450 kg P ha^{-1} treatments, respectively. Gypsum was added at 416 g per sub-plot for the 60 kg S ha^{-1} treatment. Sodium molybdate was sprayed on the appropriate sub-plots at a rate of 2.24 kg ha^{-1} using a tractor mounted hydraulic sprayer. This rate provided 874 kg Mo ha^{-1}, or 9.1 mg Mo per sub-plot.

On 9 May 1983 a Tye no-till seeder was used to sod-seed red clover (cv. Redman), birdsfoot trefoil (Empire), and flatpea (Lathco) at 9.2, 17.7, and 19.3 kg ha^{-1}, respectively. There were 10 drill rows per sub-plot. From one drill row within each sub-plot a 1 m section was staked off to make the growth observations. Height measurements of red clover and birdsfoot trefoil began 1 July 1983, 3 weeks after seeding, and measurements were made once a week for 8 weeks. Flatpea was slow germinating and weed competition made it impossible to make weekly measurements. Legumes were harvested 18 August 1983. Red clover and birdsfoot trefoil plants were counted and harvested from the 1 m section. Flatpea was harvested by cutting 5 to 9 plants per drill row within each sub-plot. Plant samples were oven dried at 50°C for 48 hours and total dry matter yields were recorded. Ten soil cores were removed from the red clover and birdsfoot trefoil sub-plots to a depth of 10 cm, air dried, and ground to pass a 2 mm mesh screen. Plant and soil analyses was performed using the same methods as described in the greenhouse experiment.

Experiment 3. Legume Seedling Response to P, S, and Mo Fertilization in the 1984 Field Experiment.

A second field experiment was initiated 24 May 1984 using red clover, birdsfoot trefoil and flatpea. All seeding, fertilizer, weed, and insect control treatments were the same as the previous year. However, flatpea seeds were scarified with H_2SO_4 to remove the seed coat and thus increase seed germination. On 2 July 1984 plant heights were recorded, 5.5 weeks after seeding. Red clover and birdsfoot trefoil growth measurements were made from a 1 m drill row section within each sub-plot. Flatpea growth was determined from one drill row within the entire length of the sub-plot. Height was recorded weekly for eight weeks and then the plants were harvested within the designated sections on 20 August 1984. The plant samples were oven dried at 50°C for 48 hours to determine total dry matter yields. Ten soil cores were removed from all sub-plots to a depth of 10 cm, air dried, and ground to pass a 2 mm mesh sieve. Plant and soil analysis was performed using the same techniques as described in the greenhouse experiment.

Statistical Analysis

Experiment 1. Legume Seedling Response to P and Mo Fertilization in the Greenhouse.

Legume growth parameters, plant elemental concentrations, and plant available P responses to P fertilization were evaluated independent of Mo fertilization. Data from the three P fertilization rates (22, 67, and 201 mg kg^{-1}) were averaged and compared against the 0 mg P kg^{-1} treatment using analysis of variance procedures and F tests. The measured responses were then tested to determine if an increase due to the added P treatments could be described by a linear or quadratic equation (Table 2) (Lentner and Bishop, personal commmunications). When a specific P fertilization rate was compared to another rate in order to reveal a percent increase, Mo treatments were averaged within each P rates. Figures and data tables (e.g. Figure 1; Appendix, Table 1) display each individual measurement.

The statistical significance of legume responses to Mo fertilization were evaluated within each P treatment. Data from the three Mo fertilization rates (22, 67, and 201 μg lg^{-1}) were averaged and compared against the 0 μg Mo kg^{-1} treatment. Measured responses were then tested to determine if an increase due to added Mo could be described by a linear or quadratic equation (Table 3) (Lentner and Bishop, personal communications).

Experiment 2 and 3. Legume Seedling Response to P, S, and Mo Fertiliztion in a Field Experiment in 1983 and 1984.

Measurements included legume growth parameters, plant elemental concentrations, and plant available nutrients. These responses to P fertilzation were evaluated independent of S and Mo fertilization. Data from the three P fertilization rates (50, 150, and 450 kg ha^{-1}) were averaged and compared against the 0 kg P ha^{-1} treatment. The measured responses were then analyzed to determine if an increase due to applied P could be described by a linear or quadratic equation (Lentner and Bishop, personal communications). When a specific P fertilzation rate was compared to another, the S and Mo treatments were averaged within the individual P rates.

The measured responses to S and Mo fertilization were evaluated independent of applied P. Data from sub-plots receiving S were averaged independent of fertilizer P and Mo, and compared against the 0 kg S ha^{-1} treatment. Data from the sub-plots receiving Mo were averaged independent of P and S fertilization, and compared against the 0 g Mo ha^{-1} rate (Lentner and Bishop, personal communications).

RESULTS AND DISCUSSION

Experiment 1. Legume Seedling Response to Phosphorus and Molybdenum Fertilization in the Greenhouse.

Phosphorus Fertilization

Height. Red clover height was increased with applied P at the beginning of the first measurement period (week one), 16 days after emergence (Table 2, Appendix Table 1). At week two, the individual P treatments (22, 67, 201 mg kg^{-1}) increased red clover height 52, 122, and 131%, respectively beyond the 0 mg P kg^{-1} rate. During weeks two through eight, P fertilized plants were always taller than the control. Between weeks two and six there was almost no growth in the non-fertilized P treatment. Red clover height doubled between weeks six and eight in the control, however, growth was still small in comparison to the P fertilized treatments. By week eight, the 22, 67 and 201 mg P kg^{-1} treatments increased height over the 0 mg P kg^{-1} rate 182, 204, and 237%, respectively (Figure 1, Appendix Plate 1).

White clover and birdsfoot trefoil height increased with fertilizer P at the beginning of the second measurement period (week two), 23 days after emergence (Table 3; Appendix Table 2). White clover height at week two increased by 15, 51, and 96% beyond the control treatment using 22, 67, and 201 mg P kg^{-1}, respectively. Birdsfoot trefoil height also increased during this same time period by 18 and 46% over the 0 mg P kg^{-1} treatment with either 22 and 67, or 201 mg

Table 2. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on red clover height and trifoliate leaf count during the first eight weeks of growth in the greenhouse.

		Height								Trifoliate count							
		Week								Week							
Source	df†	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8
Main Effects																	
P	3	**	**	**	**	**	**	**	**	**	**	**	**	**	**	**	**
0 P vs. 1,2,3 P‡	1	**	**	**	**	**	**	**	**	**	**	**	**	**	**	**	**
Linear 1,2,3 P	1	*	**	**	**	**	**	**	**	**	**	**	**	**	**	**	**
Quadratic 1,2,3 P	1	**	**	**	**	**	*	*	NS§	**	**	**	**	**	**	**	**
Mo	3	NS	NS	**	**	**	**	**	**	NS	NS	NS	NS	**	**	**	**
0 Mo vs. 1,2,3 Mo‡	1	NS	*	**	**	**	**	**	**	NS	NS	NS	NS	**	**	**	**
Linear 1,2,3 Mo	1	NS	NS	NS	*	**	**	**	**	NS	NS	NS	NS	**	**	**	**
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	**	**	**	NS	NS	NS	NS	NS	NS	*	NS	**
Interaction																	
P X Mo	9	NS	NS	NS	NS	**	**	**	**	NS	NS	NS	NS	*	**	**	**

*, ** Significant at the 0.05 and 0.01 levels, respectively.

† df = Degrees of freedom.

‡ 0,1,2,3 P and 0,1,2,3 Mo represents 0, 22, 67 and 201 mg P kg^{-1} and 0, 22, 67 and 201 ug Mo kg^{-1}, respectively.

§ NS = Not significant.

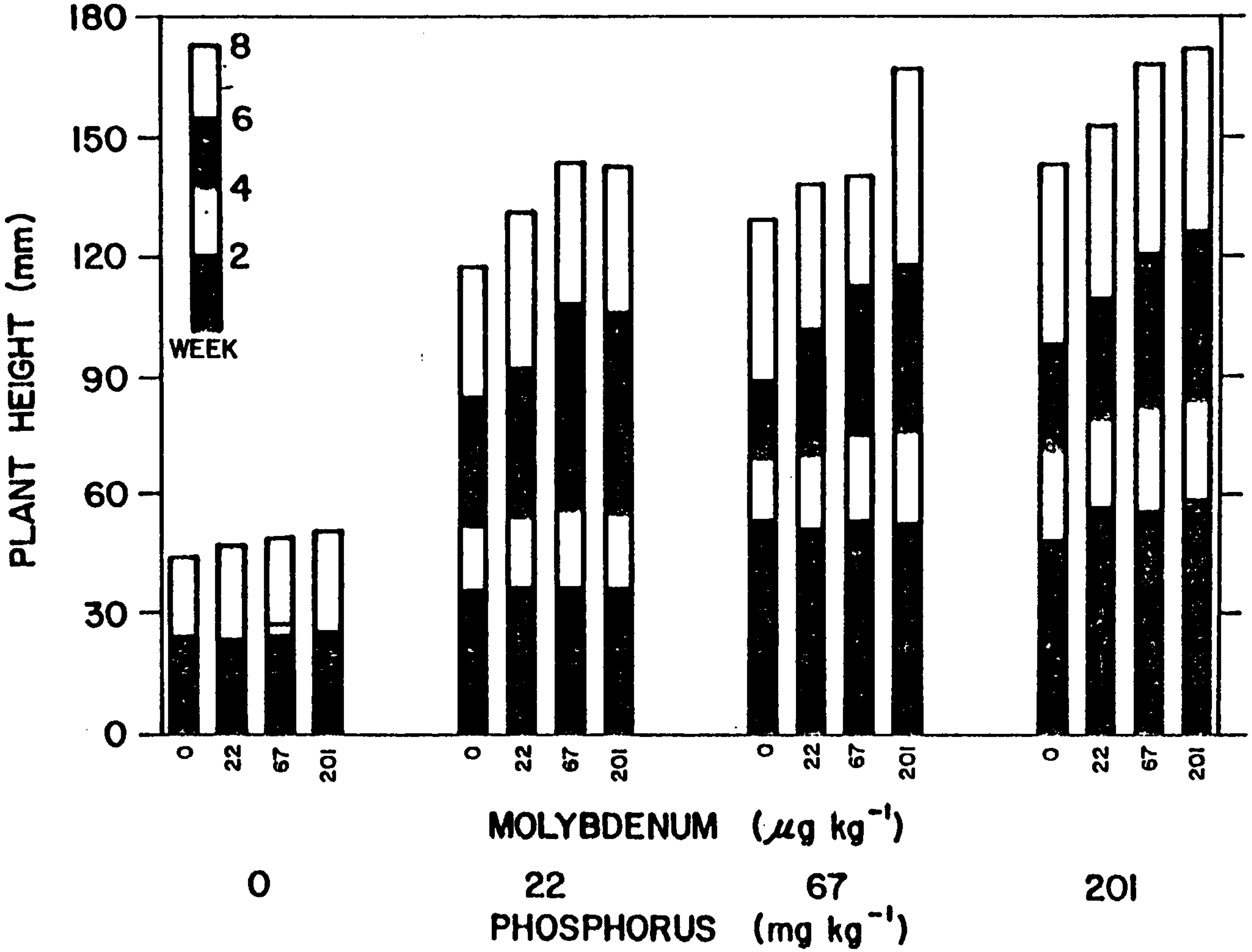

Figure 1. Red clover height as influenced by P and Mo fertilization during eight weeks of growth in the greenhouse.

Table 3. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on white clover and birdsfoot trefoil height during the first eight weeks of growth in the greenhouse.

		White clover week				Birdsfoot trefoil week			
Source	df†	2	4	6	8	2	4	6	8
Main Effects									
P	3	**	**	**	**	**	**	**	**
0 P vs. 1,2,3 P‡	1	**	**	**	**	**	**	**	**
Linear 1,2,3 P	1	**	**	**	**	**	**	**	**
Quadratic 1,2,3 P	1	**	**	**	**	**	**	**	**
Mo	3	NS§	NS	NS	NS	NS	NS	NS	**
0 Mo vs. 1,2,3 Mo‡	1	NS	NS	NS	NS	NS	NS	*	**
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	**
Interaction									
P X Mo	9	NS	NS	NS	NS	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.

† df = Degrees of freedom.

‡ 0,1,2,3 P and 0,1,2,3, Mo respresents 0, 22, 67, and 201 mg P kg^{-1} and 0, 22, 67 and 201 μg Mo kg^{-1}, respectively.

§ NS = Not significant.

P kg^{-1}, respectively. White clover and birdsfoot trefoil growth was poor within the control treatment through week eight. Height of white clover only increased approximately 5 mm without applied P between weeks two and eight while birdsfoot trefoil height increased an average of 11 mm in the 0 mg P kg^{-1} during this same time period. Measurements at week eight showed that for the 22, 67, and 201 mg P kg^{-1} treatments, white clover height was 340, 434, and 528% greater than the control, respectively. Birdsfoot trefoil height was 258, 386, and 400% greater than the pots not receiving P, respectively (Figures 2 and 3; Appendix Plates 2 and 3). Moreover, Munns (1965a) reported that in P deficient alfalfa, growth was poor, leaves were blue-green, and petioles were reddish brown, all of which was observed within the 0 mg P kg^{-1} treatment for each legume in these experiments.

Legume seedling vigor as determined by plant height was significantly increased beyond the control with each increment of fertilizer P. The initial 22 mg P kg^{-1} treatment produced the largest growth increase relative to additional P fertilization within each legume species. Shoope et al. (1961) reported that white clover height increased with increasing P fertilization up to 312 kg P on a sandy loam in the southern Piedmont section of Virginia. The fact that legume height increased with added P within the first and second week of measurement supports the importance of fertilizer P as a starter fertilizer for plant establishment as supported by Wallingford (1978b).

<u>Trifoliate Leaf Count</u>. Phosphorus fertilization increased red

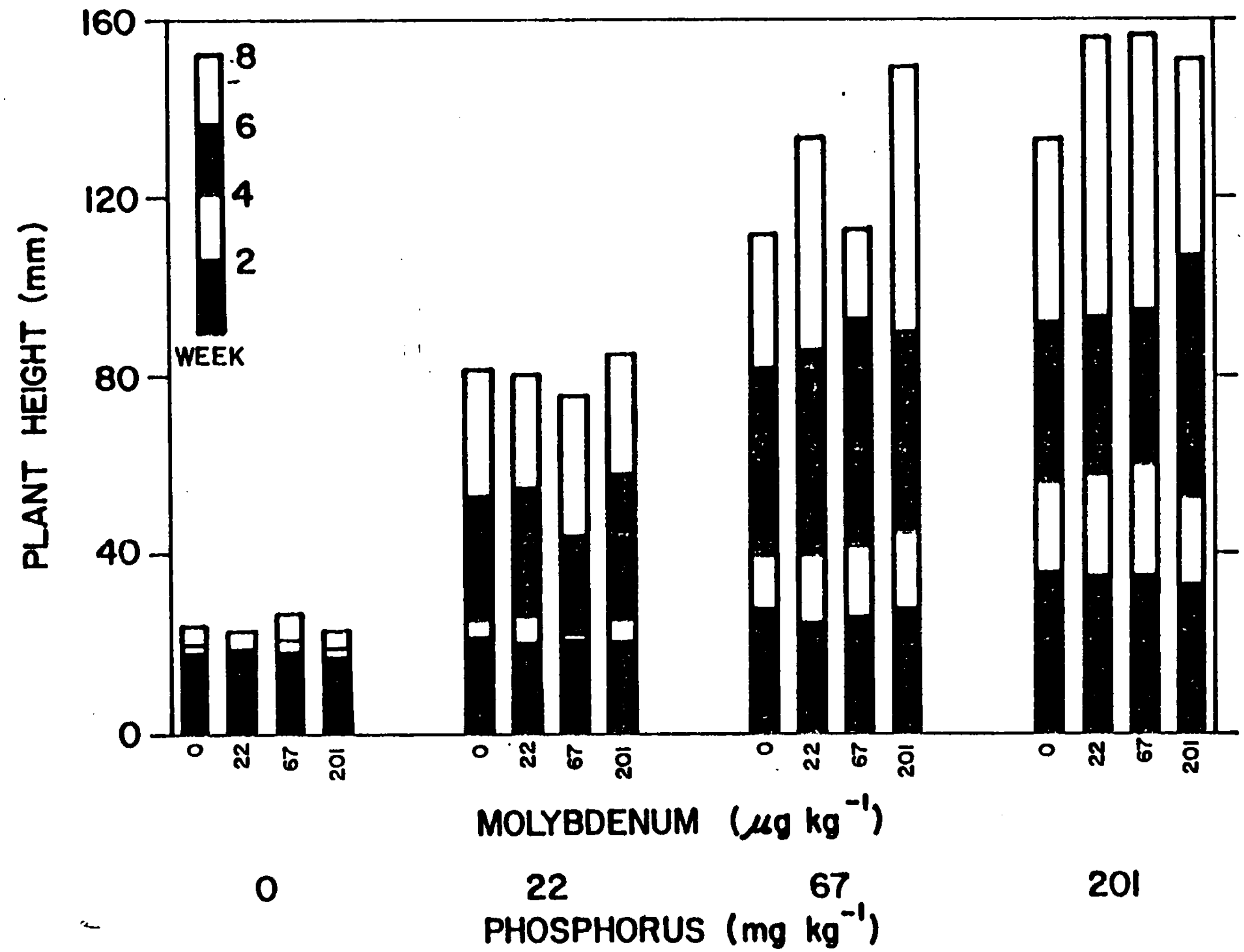

Figure 2. White clover height as influenced by and Mo fertilization during eight weeks of growth in the greenhouse.

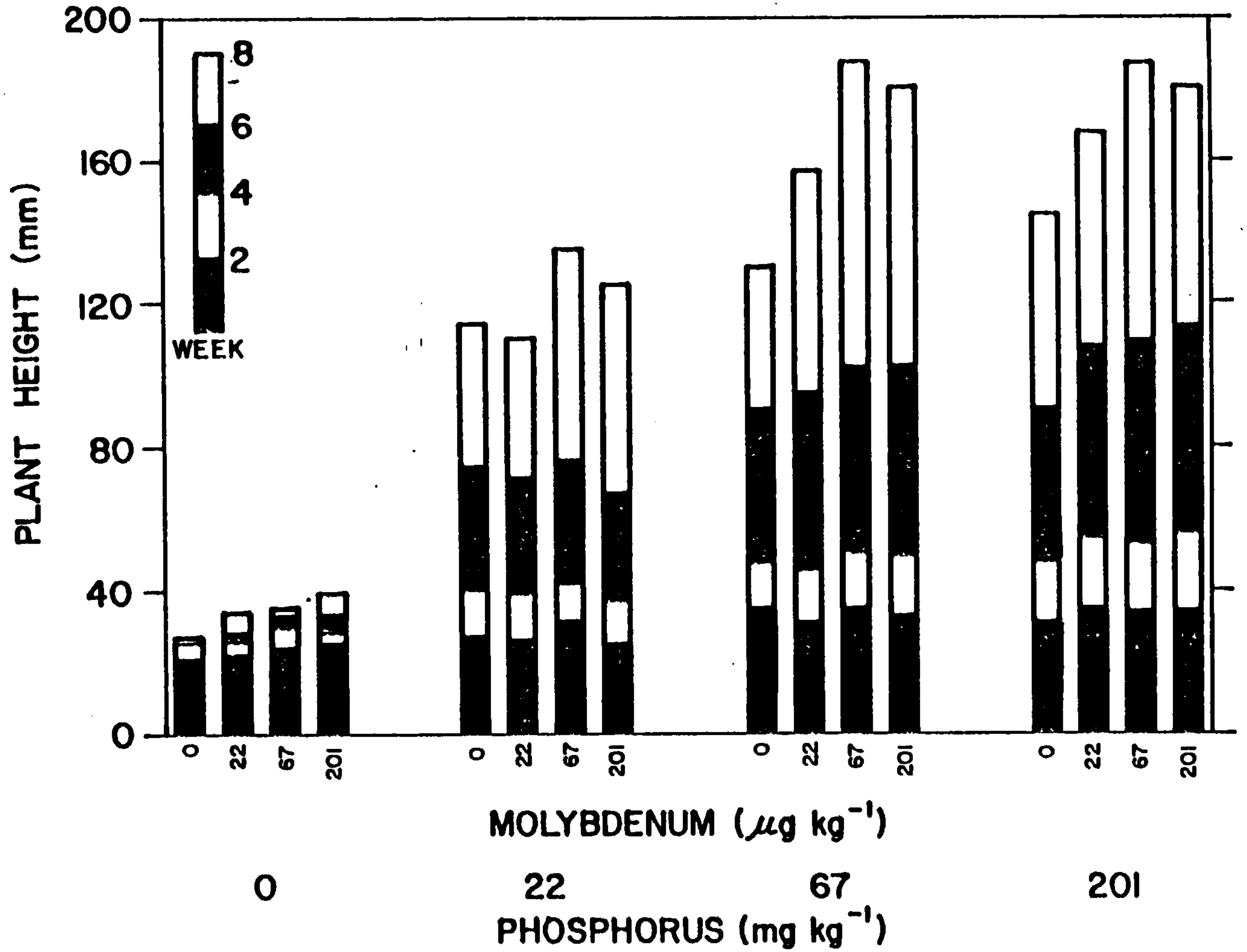

Figure 3. Birdsfoot trefoil height as influenced by P and Mo fertilization during eight weeks of growth in the greenhouse.

clover trifoliate leaf count during the first measurement week (Table 2; Appendix Table 1). During the second week, trifoliate leaf count increased 90% beyond the control treatment using either 67 or 201 mg P kg^{-1}. Leaf count increased each week within the 0 mg P kg^{-1} treatment, however, the rate of development was much slower than the rate in the fertilized pots. At week eight, the 22, 67, and 201 mg P kg^{-1} treatments increased leaf count 200, 290, and 385%, respectively, over the 0 mg P kg^{-1} rate (Figure 4). During this same time period, leaf count was 185% greater in the 201 mg P kg^{-1} treatment than in the 22 mg P kg^{-1} application, but the largest increase with respect to fertilizer rate occured with the initial 22 mg P kg^{-1} treatment.

Fertilizer P applications increased seedling vigor in terms of higher leaf counts. This implies an increased plant canopy. Greater plant canopy coverage of the soil surface increases the competitive ability of the legume relative to undersirable species. Since plant nutrients are more concentrated in leaves as compared to stems, the feeding value per unit area is increased with increasing leaf count. It is also reasonable to expect increases in dry matter yields as a result of higher leaf count values. Moreover, Qualls and Cooper (1968) stated that seedling vigor as measured in this experiment by increased leaf counts is directly correlated with seedling emergence and ultimately with forage yield.

Dry Weight. Red clover, white clover, and birdsfoot trefoil increased in total dry weight with each increment of fertilizer P (Tables 4 and 5). Red clover dry weight averaged 0.14, 1.96, 3.19,

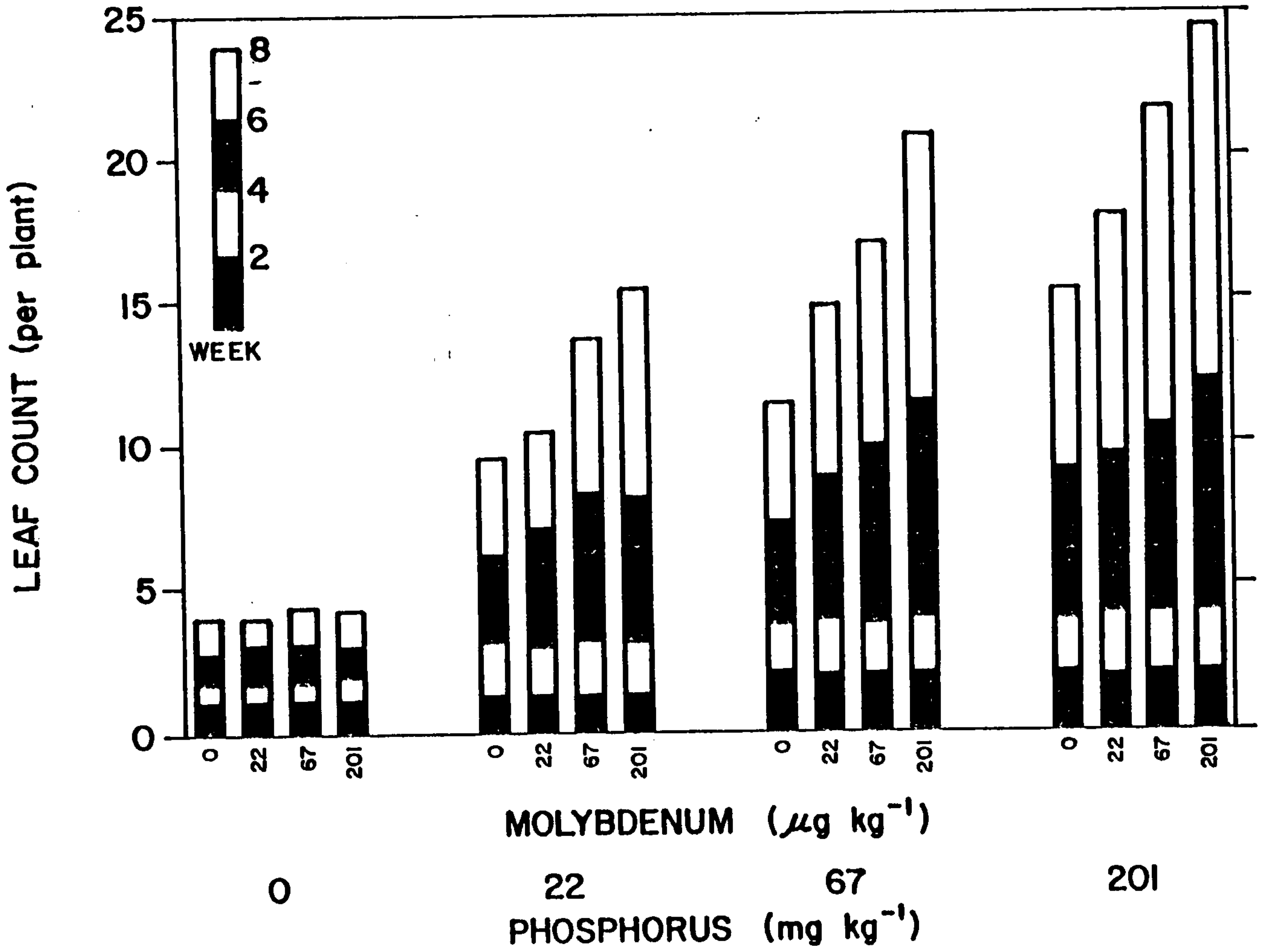

Figure 4. Red clover trifoliate leaf count as influenced by P and Mo fertilization during eight weeks of growth in the greenhouse.

Table 4. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on red clover dry weight, trifoliate leaf area, and tissue concentrations of P, Mo, and N after eight weeks of growth in the greenhouse.

Source	df†	Dry wt.	Trifoliate Area	P‡	df	Mo	df	N
Main Effects								
P	3	**	**	**	1	*	2	**
0 P vs. 1,2,3 P§	1	**	**	**		–¶		–
Linear 1,2,3 P	1	**	**	**		–	1	**
Quadratic 1,2,3 P	1	**	NS#	NS		–	1	NS
Mo	3	**	**	**	3	**	3	**
0 Mo vs. 1,2,3 Mo§	1	**	**	**	1	**	1	**
Linear 1,2,3 Mo	1	**	**	**	1	**	1	**
Quadratic 1,2,3 Mo	1	*	NS	**	1	NS	1	**
Interaction								
P X Mo	9	**	**	**	3	NS	6	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.

† df = Degrees of freedom.

‡ Statistical analyses performed on P uptake (mg pot^{-1}) only.

§ 0,1,2,3 P and 0,1,2,3 Mo represents 0, 22, 67 and 201 mg P kg^{-1} and 0, 22, 67, and 201 µg Mo kg^{-1}, respectively.

¶ Insufficient data for statistical analysis.

NS = Not significant.

Table 5. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on white clover dry weight and trifoliate leaf area, and birdsfoot trefoil dry weight after eight weeks of growth in the greenhouse.

		White Clover		Birdsfoot Trefoil
Source	df†	Dry wt.	Trifoliate Area	Dry wt.
Main Effects				
P	3	**	**	**
0 P vs. 1,2,3 P‡	1	**	**	**
Linear 1,2,3 P	1	**	**	**
Quadratic 1,2,3 P	1	**	**	**
Mo	3	**	**	NS§
0Mo vs. 1,2,3 MO‡	1	**	**	**
Linear 1,2,3 Mo	1	**	**	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS
Interaction				
P X Mo	9	NS	**	NS

*, ** Significant at the 0.05, and 0.01 levels, respectively.

† df = Degrees of freedom.

‡ 0,1,2,3 P and 0,1,2,3 Mo represents 0, 22, 67, and 201 mg P kg^{-1}, and 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.

§ NS = Not significant.

and 4.57 gms pot^{-1} with the 0, 22, 67, and 201 mg P kg^{-1} treatments, respectively. White clover and birdsfoot trefoil dry weights for the same P rates averaged 0.22, 1.01, 2.53, and 4.73, and 0.09, 0.73, 1.80, and 2.53 gms pot^{-1}. Red clover dry weight increased the most with the initial application of P (22 mg kg^{-1}). White clover and birdsfoot trefoil dry weights however, increased the most with respect to fertilizer treatment with 67 mg P kg^{-1} (Figures 5a, 6a, and 7). These increases in dry weight are in agreement with McLean and Ssali (1977) who reported a 400% increase in alfalfa yields from a 37 kg P ha^{-1} application on a moderately acid loam.

Trifoliate Leaf Area. Red clover trifoliate leaf area increased 556, 569, and 744% beyond the control with 22, 67, and 201 mg P kg^{-1}, respectively (Figure 5b). White clover trifoliate leaf area increased 420, 1100, and 1500% over the 0 mg P kg^{-1} treatment with the same P fertilizer rates (Figure 6b). The largest increase in leaf area relative to P fertilization occured at the initial 22 mg P kg^{-1} treatment for red clover; white clover leaf area showed the largest increase from the 67 mg P kg^{-1} application (Tables 4 and 5; Figures 5b and 6b; Appendix Tables 3 and 4). Phosphorus fertilization clearly improved legume seedling vigor as shown by these trifoliate leaf area measurements.

Plant Tissue Analysis. Phosphorus fertilizer applications of 22, 67, and 201 mg P kg^{-1} increased P uptake in red clover 25, 45, and 100-fold, respectively, in comparison to the non-fertilized soil (Tables 4 and 6). The same P fertilizer rates increased white clover

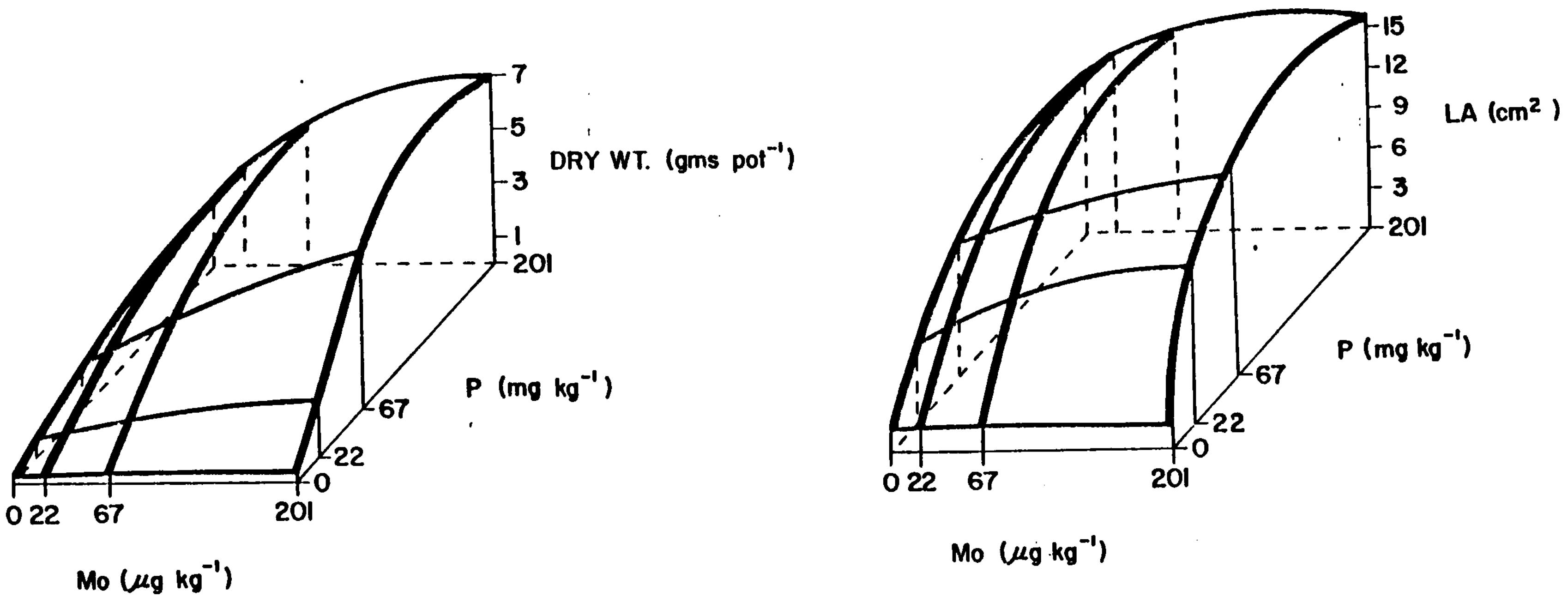

Figure 5. Red clover dry weight (a) and red clover trifoliate leaf area (b) as influenced by P and Mo fertilization after eight weeks of growth in the greenhouse.

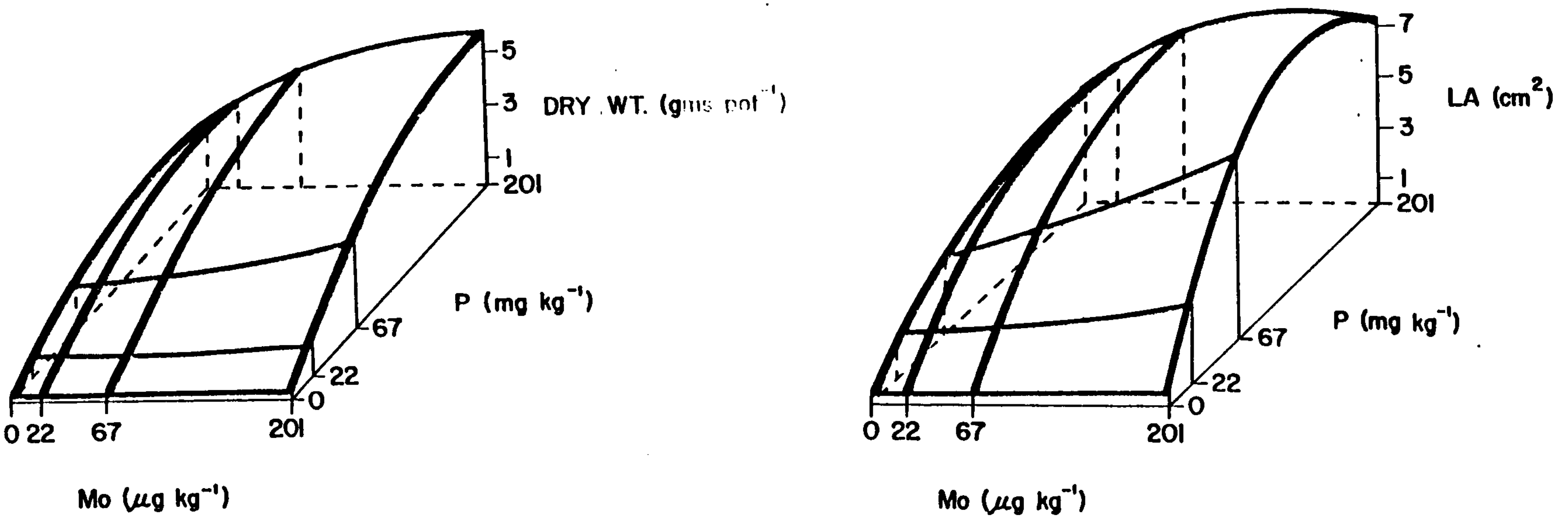

Figure 6. White clover dry weight (a) and white clover trifoliate leaf area (b) as influenced by P and Mo fertilization after eight weeks of growth in the greenhouse.

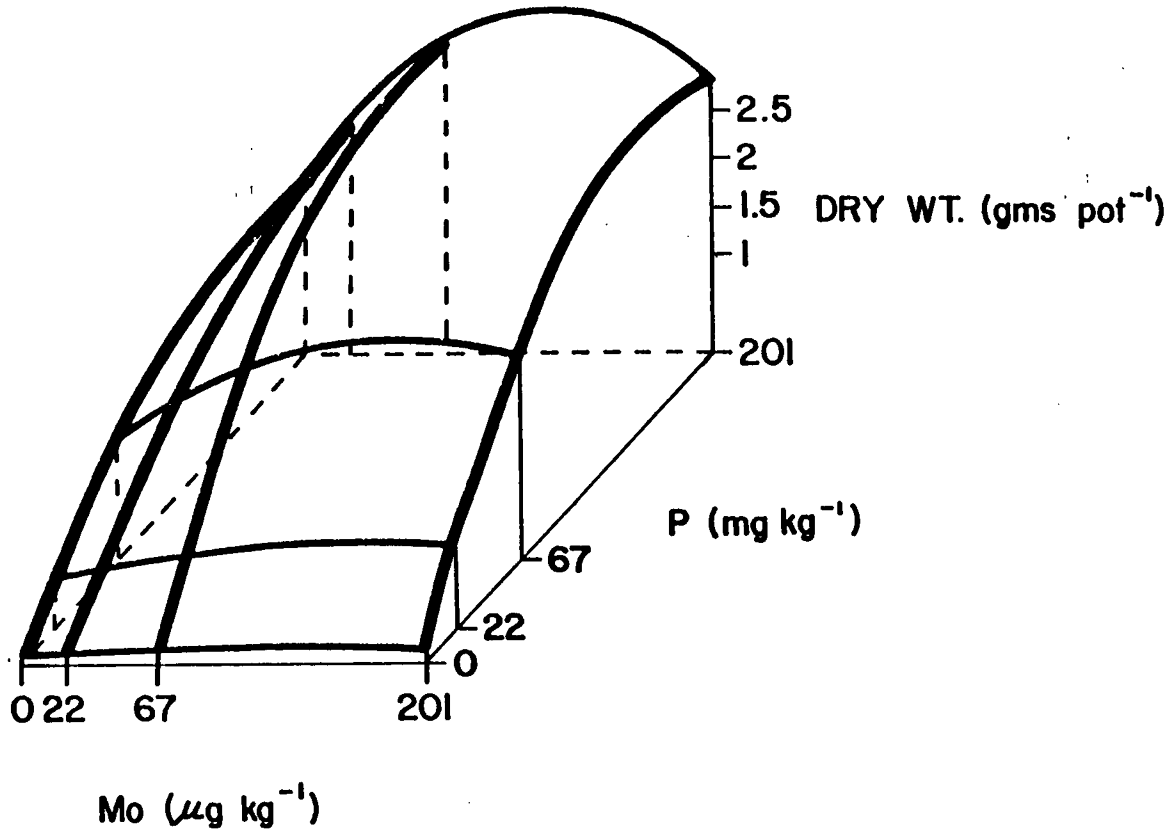

Figure 7. Birdsfoot trefoil dry weight as influenced by P and Mo fertilization after eight weeks of growth in the greenhouse.

Table 6. Phosphorus uptake and tissue concentrations of P, Mo, and N for red clover, as affected by P and Mo fertilization after eight weeks of growth in the greenhouse.

Treatment		P†		Mo	N
P	Mo				
mg kg^{-1}	μg kg^{-1}	mg pot^{-1}	mg kg^{-1}	mg kg^{-1}	g kg^{-1}
0	0	0.15	1003	-‡	-
	22	0.11	800	-	-
	67	0.13	901	-	-
	201	0.15	1000	-	-
22	0	2.44	2249	-	18.3
	22	3.31	2141	-	20.6
	67	3.53	1620	-	25.2
	201	4.01	1317	-	28.3
67	0	4.31	3102	0.10	19.0
	22	5.29	2134	0.12	22.0
	67	6.23	1923	0.19	25.3
	201	8.44	1495	0.28	27.6
201	0	8.52	3443	0.13	24.0
	22	9.93	2768	0.23	26.2
	67	15.48	2952	0.25	30.0
	201	18.31	2614	0.33	31.0

† Plant sample replications from treatments 1 through 4 were combined within each Mo rate to provide adequate tissue for analysis.

‡ Insufficient sample.

and birdsfoot trefoil P uptake 7, 19, and 67 times, and 11, 30 and 71 fold, respectively, over the 0 mg P kg^{-1} treatment (Tables 7 and 8). Phosphorus concentration in plant tissue increased in all legume species with P fertilization (Tables 7 and 8). At pre-bud stage, the P concentration in all legumes was equal to or less than 1300 mg P kg^{-1} without applied P. According to Martin and Matocha (1973) the critical P concentration for red and white clover was equivalent to 0.25% (2500 mg P kg^{-1}) but they did not report stage of maturity. Based on a 2500 mg P kg^{-1} critical level, an adequate P concentration in red clover and white clover in this experiment was not obtained until 67 mg P kg^{-1} and 201 mg P kg^{-1} had been applied, respectively. Birdsfoot trefoil P concentration increased above 0.30% when fertilized with 201 mg P kg^{-1} (Tables 7 and 8). Therefore, it is apparent that these legumes may require different available soil P levels to provide adequate P concentrations in the tissues. Many of the measured legume growth parameters exhibited the greatest increase relative to the control with an initial 22 mg P kg^{-1} treatment. However, the tissue P concentrations associated with this fertilizer rate were lower than the critical P concentration as stated by Martin and Matocha (1973) (Tables 7 and 8). A possible explanation is that stage of plant maturity differed between this experiment and that of Martin and Matocha (1973), or the level of available soil P was higher in the results presented by Martin and Matocha.

Phosphorus fertilization increased the concentration of Mo in red

Table 7. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on white clover P uptake, Mo, and N tissue concentrations, and birdsfoot trefoil P uptake and tissue N after eight weeks of growth in the greenhouse.

Source	White clover						Birdsfoot Trefoil			
	df†	P	df	Mo	df	N	df	P	df	N
Main Effects										
P	3	**	1	NS‡	2	**	3	**	2	**
OP vs. 1,2,3 P§	1	**		–¶		–	1	**		–
Linear 1,2,3 P	1	**		–	1	**	1	**	1	**
Quadratic 1,2,3 P	1	NS		–	1	NS	1	NS	1	NS
Mo	3	NS	3	*	3	**	3	NS	3	**
OMo vs. 1,2,3 Mo§	1	NS	1	NS	1	**	1	NS	1	**
Linear 1,2,3 Mo	1	NS	1	*	1	NS	1	NS	1	**
Quadratic 1,2,3 Mo	1	NS	1	NS	1	NS	1	NS	1	**
Interpretation										
P X Mo	9	NS	3	NS	6	NS	9	NS	6	NS

*, ** Significant at the 0.05, and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P and 0,1,2,3 Mo represents 0, 22, 67, and 201 mg P kg^{-1} and 0, 22, 67 and 201 μg Mo kg^{-1}, respectively.
¶ Insufficient data for statistical analysis.

Table 8. Phosphorus uptake and tissue concentrations of P, Mo, and N for white clover and birdsfoot trefoil as affected by P and Mo fertilization after eight weeks of growth in the greenhouse.

Treatment		White clover				Birdsfoot trefoil†		
P	Mo	-----P------		Mo	N	------P-----		N
mg kg^{-1}	μg kg^{-1}	mg pot^{-1}	--mg kg^{-1}--		mg pot^{-1}	mg kg^{-1}	--g kg^{-1}--	
0	0	0.18	800	-§	-	0.06	900	-
	22	0.19	1001	-	-	0.11	1103	-
	67	0.18	749	-	-	0.14	1300	-
	201	0.22	902	-	-	0.13	1105	-
22	0	1.24	1182	-	18.1	1.16	1705	25.2
	22	1.21	1374	-	22.2	1.25	1670	27.0
	67	1.30	1274	-	22.1	1.45	1811	31.7
	201	1.57	1413	-	18.9	1.11	1642	29.5
67	0	3.43	1828	0.14	21.0	2.96	2216	27.9
	22	3.60	1611	0.15	23.1	2.98	1835	28.4
	67	3.50	1465	0.14	25.7	3.63	1741	30.2
	201	4.46	1248	0.23	25.8	3.63	1670	32.5
201	0	11.23	2995	0.09	24.2	5.44	3615	27.8
	22	13.48	2852	0.13	29.1	9.48	3270	29.7
	67	12.54	2672	0.14	32.3	8.58	2908	33.5
	201	14.74	2566	0.16	32.1	7.85	2825	34.8

† Insufficient samples for Mo analysis.
‡ Plant sample replications from treatments 1 through 4 were combined within each Mo rate to provide adequate tissue for analysis.
§ Insufficient sample.

clover, and total N in all legumes. Total red clover Mo content increased 1.36 times when applied P was increased from 67 to 201 mg kg^{-1} (Tables 4 and 7). Singh and Kumar (1979) stated that Mo uptake could be increased with applied P by forming a readily available phosphomolybdate anion and the data in Tables 4 and 7 appear to support their work. Total N in red clover increased 1.02 and 1.21 fold beyond the 22 mg P kg^{-1} treatment when fertilizer P was increased to 67 and 201 mg kg^{-1}, respectively (Tables 4 and 7). White clover and birdsfoot trefoil total N increased 1.2 and 1.4 times, and 1.04 and 1.11 fold, respectively over the 22 mg P kg^{-1} rate when 67 and 201 mg P kg^{-1} was applied (Tables 6 and 8). Griffith (1978) observed that available P stimulated nodule bacteria and therefore often increased effective symbiosis. Similar results were obtained by Haynes and Ludecke (1981) with work in which they reported that total N in white clover increased from 3.01 to 3.29% when fertilizer P was increased from 50 to 350 kg ha^{-1}. Phosphorus fertilization did not affect tissue concentrations of Ca, Mg, and K in red clover (Appendix Table 5).

Available Soil Phosphorus. Bray-1 extractable P of soils in which all legumes were grown increased by an average of 3, 10, and 40 times with each increase in applied P (Tables 9 and 10; Appendix Table 6). Plant available P within the non-fertilized pots averaged 4 mg kg^{-1} and this resulted in extremely poor legume growth relative to the fertilized treatments (Appendix Plates 1, 2, 3). After fertilizing with 22 mg P kg^{-1}, available P increased to 12 mg kg^{-1} and this

Table 9. Statistical significance levels, based on F tests, for the effects of P and Mo fertilization on Bray-1 extractable P and resin extractable Mo from the red clover greenhouse experiment, and Bray-1 extractable P from the white clover and birds-foot trefoil greenhouse experiments.

Source	df†	Red Clover P	Red Clover Mo	White Clover P	Birdsfoot Trefoil P
Main Effects					
P	3	**	NS‡	**	**
0 P vs. 1,2,3 P§	1	**	NS	**	**
Linear 1,2,3 P	1	**	NS	**	**
Quadratic 1,2,3 P	1	**	NS	**	
Mo	3	NS	**	*	NS
0 Mo vs. 1,2,3 Mo§	1	NS	**	*	NS
Linear 1,2,3 Mo	1	NS	**	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS
Interaction					
P X Mo	9	NS	NS	*	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.

† df = Degrees of freedom.

‡ NS = Not significant.

§ 0,1,2,3 P and 0,1,2,3 Mo represent 0, 22, 67, and 201 mg P kg^{-1} and 0, 22, 67, and 201 µg Mo kg^{-1}, respectively.

Table 10. Bray-1 extractable P and resin extractable Mo as affected by P and Mo fertilization of a Gilpin soil utilized in the greenhouse experiments.

Applied P	Bray-1 Extractable P	Applied Mo	Resin Extractable Mo
mg kg^{-1}	mg kg^{-1}	μg kg^{-1}	μg kg^{-1}
0	4	0	12
22	12	22	15
67	39	67	21
201	158	201	29

was normally associated with the largest increase in plant growth per increment of applied P. McLean and Ssali (1977) reported that alfalfa yields in a greenhouse were drastically increased when 37 kg P ha^{-1} was applied to a soil originally containing 2.7 mg kg^{-1} Bray-1 extractable P. They did not observe a yield increase beyond a Bray-1 soil extractable P level of 12.5 mg kg^{-1}. This disagrees with the present study because additional increases in plant growth were observed at Bray-1 extractable P levels of 39 and 158 mg kg^{-1}. However, these additional increases in plant growth were normally smaller than the increase observed from the initial 22 mg P kg^{-1} rate beyond the control.

Molybdenum Fertilization

Height. Red clover height was increased with applied Mo during weeks one through five within the highest P treatment (Table 11, Figure 1). This was supported by Gupta and Lipsett (1981) who observed that Mo affected yield responses at an early plant growth stage. During weeks six through eight red clover height increased with Mo fertilization within the 22, 67, and 201 mg P kg^{-1} treatments. Molybdenum applications of 201 μg kg^{-1} increased red clover height at week eight beyond the non-fertilized Mo treatments by 21, 29, and 20% within the 22, 67, and 201 mg P kg^{-1} rates, respectively (Table 11; Figure 1; Appendix Table 7).

White clover and birdsfoot trefoil height increased with Mo fertilization beginning at the sixth week of measurement within the

Table 11. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on red clover height and trifoliate leaf count during the first eight weeks of growth in the greenhouse.

		Height				Trifoliate Count			
		-----P ($mg\ kg^{-1}$)-----				---P ($mg\ kg^{-1}$)-----			
Source	df†	0	22	67	201	0	22	67	201
Wk. 2 Mo	3	NS‡	NS	NS	*	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo§	1	NS	NS	NS	**	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Wk. 4 Mo	3	NS	NS	NS	**	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	**	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Wk. 6 Mo	3	NS	**	**	**	NS	*	**	**
0 Mo vs. 1,2,3 Mo	1	NS	**	**	**	NS	**	**	**
Linear 1,2,3 Mo	1	NS	**	**	**	NS	NS	**	**
Quadratic 1,2,3 Mo	1	NS	**	*	NS	NS	NS	NS	NS
Wk. 8 Mo	3	NS	**	**	**	NS	**	**	**
0 Mo vs. 1,2,3 Mo	1	NS	**	**	**	NS	**	**	**
Linear 1,2,3 Mo	1	NS	NS	**	**	NS	**	**	**
Quadratic 1,2,3 Mo	1	NS	*	NS	*	NS	*	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67 and 201 $\mu g\ Mo\ kg^{-1}$, respectively.

Table 12. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on white clover and birdsfoot trefoil height during the first eight weeks of growth in the greenhouse.

Source	df†	White Clover Height P (mg kg^{-1}) 0	22	67	201	Birdsfoot Trefoil Height P (mg kg^{-1}) 0	22	67	201
Wk. 2 Mo	3	NS‡	NS	NS	NS	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo§	1	NS	NS	NS	NS	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	*	NS	NS
Wk. 4 Mo	3	NS	NS	NS	NS	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	*	NS	NS	NS	NS	NS	NS	NS
Wk. 6 Mo	3	NS	NS	NS	*	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	**	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Wk. 8 Mo	3	NS	NS	NS	*	NS	NS	**	*
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	**	NS	NS	**	**
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	**	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo respresents 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.

201 mg P kg^{-1} treatment. Molybdenum applied at 201 μg kg^{-1} increased white clover height during week eight beyond the 0 μg Mo kg^{-1} treatment by 13% within the 201 mg P kg^{-1} rate. Birdsfoot trefoil height increased at week eight over the non-fertilized Mo treatment by 45 and 29% with 67 μg Mo kg^{-1} within the 67 and 201 mg P kg^{-1} treatments, respectively (Table 12; Figures 2 and 3).

Seedling vigor, as measured by plant height, increased with Mo fertilization within each of the applied P treatments. Many times when Mo was applied at 201 μg kg^{-1} within the 22 or 67 mg P kg^{-1} rates, legume height was greater than the next highest P treatment without Mo fertilization. Molybdenum fertilization did not increase legume height within the 0 mg P kg^{-1} treatment (Figures 1, 2, and 3; Appendix Plates 1a, 2a, and 3a). Only after the P requirement was satisfied did Mo increase height. This is in agreement with Anderson's (1956b) results.

Trifoliate Leaf Count. Molybdenum fertilization increased red clover leaf count during weeks five through eight within each of the applied P treatments (Table 11; Figure 4; Appendix Table 7). During week eight Mo applied at 201 μg kg^{-1} increased leaf count beyond the non-fertilized Mo treatment by 62, 82, and 60% within the 22, 67, and 201 mg P kg^{-1} treatments, respectively. Molybdenum fertilization therefore enhanced seedling vigor by increasing trifoliate leaf count within all the fertilizer P treatments. When Mo was applied at 67 or 201 μg kg^{-1} within the 22 and 67 mg P kg^{-1} treatments, trifoliate leaf count was greater than the next highest P rates without Mo fertilization (Figure 4). Molybdenum fertilization did not increase leaf count within the non-fertilized P treatment. By visual inspection, the legumes not receiving Mo within the applied P treatments had more chlorotic leaves than those with fertilizer Mo (Appendix Plates 1, 2, and 3). Gupta and Lipsett (1981) also observed this effect and attributed the response to a reduction of N_2-fixation within the plants.

Dry Weight. Molybdenum applied at 201 µg kg^{-1} increased red clover dry weight beyond the 0 µg Mo kg^{-1} treatment by an average of 220% within the 22, 67, and 201 mg P kg^{-1} applications (Table 13, Figure 5a) but white clover dry weight only increased within the 67 mg P kg^{-1} treatment. However, a 90% increase in white clover dry weight was observed for the 201 µg Mo kg^{-1} application within the 67 mg P kg^{-1} treatment (Table 14; Figure 6a). Though not statistically significant, birdsfoot trefoil dry weight increased also with 201 µg Mo kg^{-1} by 95% within the 201 mg P kg^{-1} treatment (Table 14; Figure 7).

The increase in red clover dry weight with applied Mo is a reflection of increased seedling vigor. These observed increases in dry weight are also in accordance with similar research by Reisenauer (1956), and Hagstrom and Berger (1963). However, Mo fertilization did not increase legume dry weight within the non-fertilized P treatment and therefore demonstrated the need for adequate available P. White clover and birdsfoot trefoil dry weight were not increased with applied Mo in many of the added P rates also, but this may indicate a

Table 13. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment on red clover dry weight, trifoliate leaf area, and tissue concentrations of P, Mo and N after eight weeks of growth in the greenhouse.

Source	df†	P (mg kg^{-1}) 0	22	67	201
Dry weight					
Mo	3	NS‡	**	**	**
0 Mo vs. 1,2,3 Mo§	1	NS	**	**	**
Linear 1,2,3 Mo	1	NS	**	**	**
Quadratic 1,2,3 Mo	1	NS	NS	NS	*
Trifoliate Area					
Mo	3	NS	**	**	**
0 Mo vs. 1,2,3 Mo	1	NS	**	**	**
Linear 1,2,3 Mo	1	NS	**	**	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS
P¶					
Mo	3	NS	NS	**	**
0 Mo vs. 1,2,3 Mo	1	NS	*	**	**
Linear 1,2,3 Mo	1	*	NS	**	**
Quadratic 1,2,3 Mo	1	NS	NS	NS	**
Mo					
Mo	3	-#	-	*	**
0 Mo vs. 1,2,3 Mo	1	-	-	*	**
Linear 1,2,3 Mo	1	-	-	*	**
Quadratic 1,2,3 Mo	1	-	-	NS	NS
N					
Mo	3	-	**	**	**
0 Mo vs. 1,2,3 Mo	1	-	**	**	**
Linear 1,2,3 Mo	1	-	**	**	**
Quadratic 1,2,3 MO	1	-	*	*	*

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.
¶ Statistical analysis performed on P uptake (mg pot^{-1}).
# Insufficient data for statistical analysis.

Table 14. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on white clover dry weight and trifoliate leaf area, and birdsfoot trefoil dry weight after eight weeks of growth in the greenhouse.

Source	df†	P (mg kg^{-1}) 0	22	67	201
White clover					
Dry weight					
Mo	3	NS‡	NS	**	NS
0 Mo vs. 1,2,3 Mo§	1	NS	NS	*	NS
Linear 1,2,3 Mo	1	NS	NS	**	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS
Trifoliate Area					
Mo	3	NS	NS	**	**
0 Mo vs. 1,2,3 Mo	1	NS	NS	**	**
Linear 1,2,3 Mo	1	NS	NS	**	**
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS
Birdsfoot Trefoil					
Dry Weight					
Mo	3	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS
Quadrate 1,2,3 Mo	1	NS	NS	NS	NS

*, ** Significant at the 0.05, and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.

differential Mo requirement relative to red clover, or a need for more precise measurement techniques since white clover and birdsfoot trefoil seedlings were not counted in the pots.

Trifoliate Leaf Area. The 201 μg Mo kg^{-1} application increased trifoliate leaf area by 55, 53, and 28% beyond the 0 μg Mo kg^{-1} treatment within the 22, 67, and 201 mg P kg^{-1} rates, respectively (Table 13; Figure 5b). Molybdenum applied at 201 μg kg^{-1} increased white clover leaf area relative to the non-fertilized Mo treatment by 67 and 69% within the 67 and 201 mg P kg^{-1} rates, respectively (Table 14; Figure 6b). Trifoliate leaf area did not increase with Mo fertilization without correcting the P deficiency. Thus, within the added P treatments, an increase of leaf area with Mo fertilization can be interpreted as an enhancement of seedling vigor.

Plant Tissue Analysis. Molybdenum fertilization at 201 μg kg^{-1} enhanced red clover P uptake 1.64, 1.98, and 2.15 times beyond the 0 μg Mo kg^{-1} treatment within the 22, 67, and 201 mg P kg^{-1} rates (Tables 7 and 13). The 201 μg Mo kg^{-1} treatment increased P uptake in white clover 1.3 fold over the non-fertilized Mo rate within the 201 mg P kg^{-1} treatment. Also, P uptake increased in birdsfoot trefoil 1.4 fold beyond the control when 201 μg Mo ha^{-1} was applied within the highest P treatment (Tables 8 and 15). Plant P concentrations decreased with Mo fertilization within the applied P treatments. However, this can be attributed to increased growth of the legumes which resulted in a "dilution effect" (Tables 7 and 8).

Molybdenum concentration in legumes was noted earlier to have

Table 15. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on white clover P uptake, Mo and N concentrations, and birdsfoot trefoil P uptake and tissue N after eight weeks of growth in the greenhouse.

		P (mg kg^{-1})			
Source	df†	0	22	67	201
White Clover					
P¶					
Mo	3	NS‡	NS	NS	**
0 Mo vs. 1,2,3 Mo§	1	NS	NS	NS	**
Linear 1,2,3 Mo	1	NS	NS	NS	**
Quadratic 1,2,3 Mo	1	NS	NS	NS	**
Mo					
Mo	3	-#	-	NS	NS
0 Mo vs. 1,2,3 Mo	1	-	-	NS	NS
Linear 1,2,3 Mo	1	-	-	*	NS
Quadratic 1,2,3 Mo	1	-	-	NS	NS
N					
Mo	3	-	NS	*	**
0 Mo vs. 1,2,3 Mo	1	-	NS	*	**
Linear 1,2,3 Mo	1	-	NS	NS	NS
Quadratic 1,2,3 Mo	1	-	NS	NS	NS
Birdsfoots trefoil					
P¶					
Mo	3	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS
N					
Mo	3	-	**	NS	**
0 Mo vs. 1,2,3 Mo	1	-	**	NS	**
Linear 1,2,3 Mo	1	-	NS	NS	**
Quadratic 1,2,3 MO	1	-	**	NS	*

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.
¶ Statistical analysis performed on P uptake.
# Insufficient data for statistical analysis.

increased with P fertilization by probably forming a readily available phosphomolybdate anion. The reverse was also observed, that is, Mo fertilization enhanced P uptake. A possible explanation may be, that Mo fertilized legumes grew more extensive root systems that were able to absorb larger quantities of available soil P.

Molybdenum fertilization increased red clover Mo concentration 2.8 and 2.5 times above the non-fertilized Mo rate within the 67 and 201 mg P kg^{-1} treatment, respectively (Tables 7 ad 13). Red clover without applied Mo averaged less than 0.13 mg Mo kg^{-1} in above ground tissue. According to Gupta (1970), red clover, when harvested at bud stage, was Mo deficient when tissue concentrations were less than 0.20 mg kg^{-1}. Red clover Mo concentration did not increase above 0.20 mg kg^{-1} within the 67 mg P kg^{-1} treatment until the application of 201 µg Mo kg^{-1}. However, within the highest P treatment, red clover Mo concentration increased beyond 0.20 mg Mo kg^{-1} with just the first rate of fertilizer Mo (Table 7). This may suggest that higher rates of Mo need to be applied when using lower P fertilization rates. Since P applications of 22 and 67 mg kg^{-1} resulted in the largest plant growth increases, 201 µg Mo kg^{-1} would thus be an appropriate Mo fertilization rate. The highest plant Mo concentrations found in these experiments are not considered plant or animal toxic according to Gupta and Lipsett (1981).

Molybdenum fertilization enhanced red clover tissue total N 1.55, 1.47, and 1.30 times beyond the 0 µg Mo kg^{-1} rate when fertilized at 201 µg Mo kg^{-1} within the 22, 67 and 201 mg P kg^{-1}

treatments, respectively (Tables 7 and 13). White clover tissue N increased with application of 201 μg Mo kg^{-1} by 1.23 and 1.33 fold above the non-fertilized Mo rate within the 67 and 201 mg P kg^{-1} treatments, respectively. Similar increases were recorded in birdsfoot trefoil (Tables 8 and 15). These increases in total plant N are probably a result of Mo enhancing N_2-fixation (Anderson, 1956a) and are similar to reports by Reisenauer (1956), and Mortvedt (1981). Since total plant N increased with Mo fertilization, the initial available soil Mo level could be considered low.

Available Soil Molybdenum. Resin extractable Mo averaged 12 μg kg^{-1} without applied Mo within the greenhouse soils (Table 10). This available Mo level can be regarded as deficient for optimum legume establishment since the plant growth parameters and total plant N increased with applied Mo. Karimian and Cox (1978) reported that deficient available Mo levels ranged between 0.6 and 11.1 μg kg^{-1} from 32 acid Coastal Plain and Piedmont soils using the ion exchange resin procedure. Available Mo levels equivalent to 29 μg kg^{-1} in this experiment were associated with enhanced plant growth, and higher plant N concentrations (Tables 10 and 16).

Table 16. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on resin extractable soil Mo in the red clover greenhouse experiment.

		P (mg kg^{-1})			
Source	df†	0	22	67	201
<u>Resin Extractable Mo</u>					
Mo	3	NS‡	*	NS	*
0 Mo vs. 1.2,3 Mo §	1	NS	NS	*	NS
Linear 1,2,3 Mo	1	*	*	NS	*
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67, and 201 μg Mo kg^{-1}, respectively.

Experiment 2. Legume Seedling Response to Phosphorus, Sulfur, and Molybdenum Fertilization in the 1983 Field Experiment.

Phosphorus Fertilization

<u>Height</u>. Red clover height during the fourth week of measurement increased beyond the non-fertilized P treatment 22% with the 150 kg P ha^{-1} application (Table 17; Figure 8; Appendix Table 8). Higher rates of P did not increase height. During weeks five through eight red clover was defoliated by rodents and this confounded the height data. Birdsfoot trefoil height was increased above the 0 kg P ha^{-1} treatment 10% during the seventh and eighth week of measurement with P applied at 150 kg ha^{-1}. When P fertilization was increased to 450 kg ha^{-1} trefoil height only increased approximately 13% beyond the 150 kg P ha^{-1} treatment during this same time period (Table 17; Figure 9; Appendix Table 8).

Legume seedling vigor, as measured by plant height, was not enhanced by P fertilization within the first 3 weeks of measurement. Limited rainfall (4.9 cm) six weeks after seeding (third week of measurement) may have been the major factor limiting legume height. Gist and Mott (1957) observed that alfalfa, red clover, and birdsfoot trefoil seedlings were greatly reduced in growth as soil moisture decreased. The plots received 15.6 cm of rain between the third and seventh measurement weeks (Appendix Table 9). As a result, red clover responded to applied P at week four and birdsfoot trefoil at week seven and eight. All legume sub-plots which received 450 kg P ha^{-1} were low in percent stand compared to the lower P treatments.

Table 17. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on red clover and birdsfoot trefoil height during the first eight weeks of growth in the 1983 field experiment.

		Red clover								Birdsfoot trefoil							
		Week								Week							
Source	df†	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8
Main Effects																	
P	3	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
O P vs. 1,2,3 P§	1	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Linear 1,2,3 P	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	*
Quadratic 1,2,3 P	1	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S	1	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS
Mo	1	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	**	*	*	NS	**	**
Interactions																	
P x S	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
P x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	**	*	NS	NS	NS
P x S x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

*, ** Significant at 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not Significant.
§ 0,1,2,3, P represents 0, 50, 150, and 450 kg P ha^{-1}, respectively.

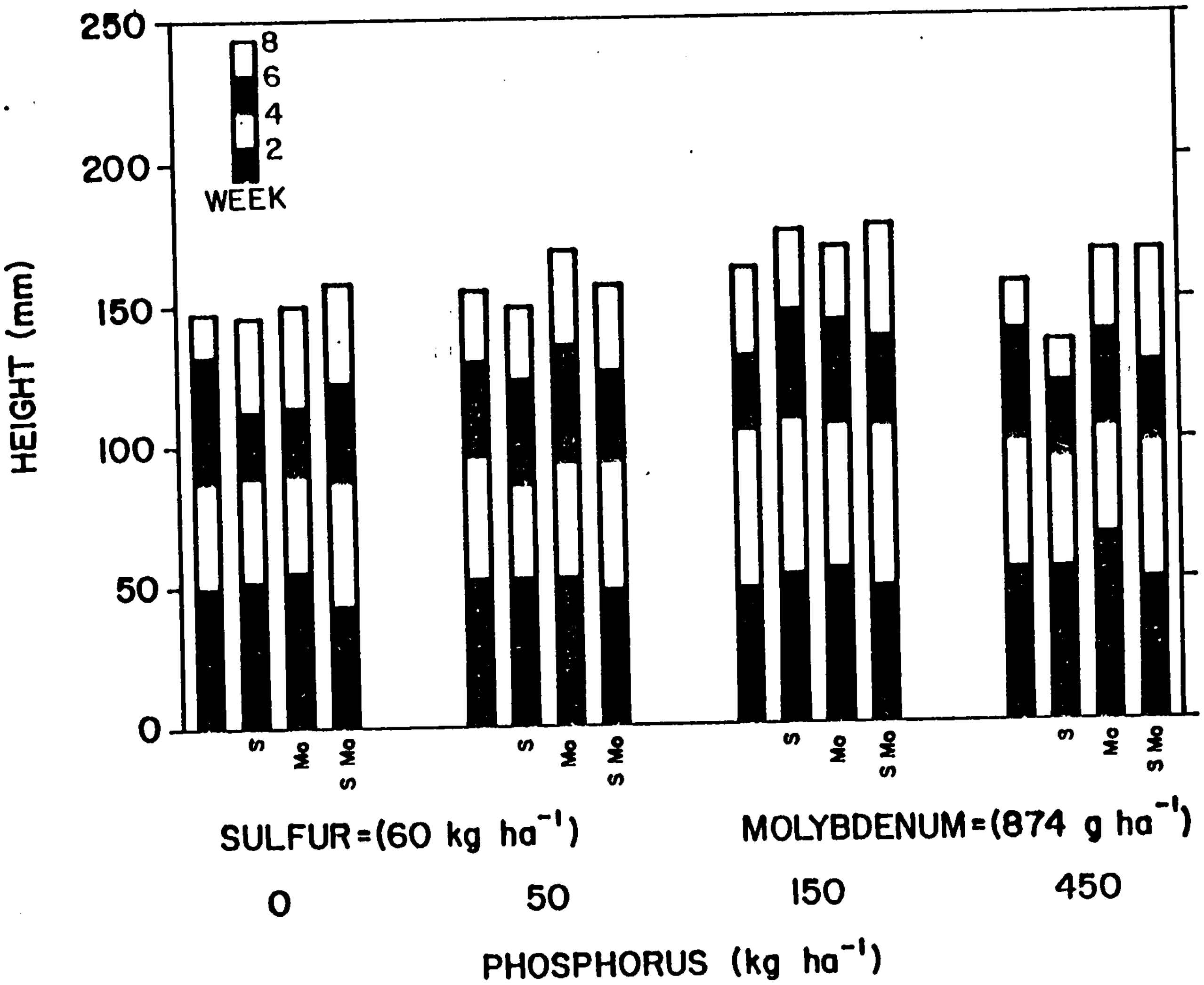

Figure 8. Red clover height as influenced by P, S, and Mo fertilization during eight weeks of growth on a Gilpin silt loam in 1983.

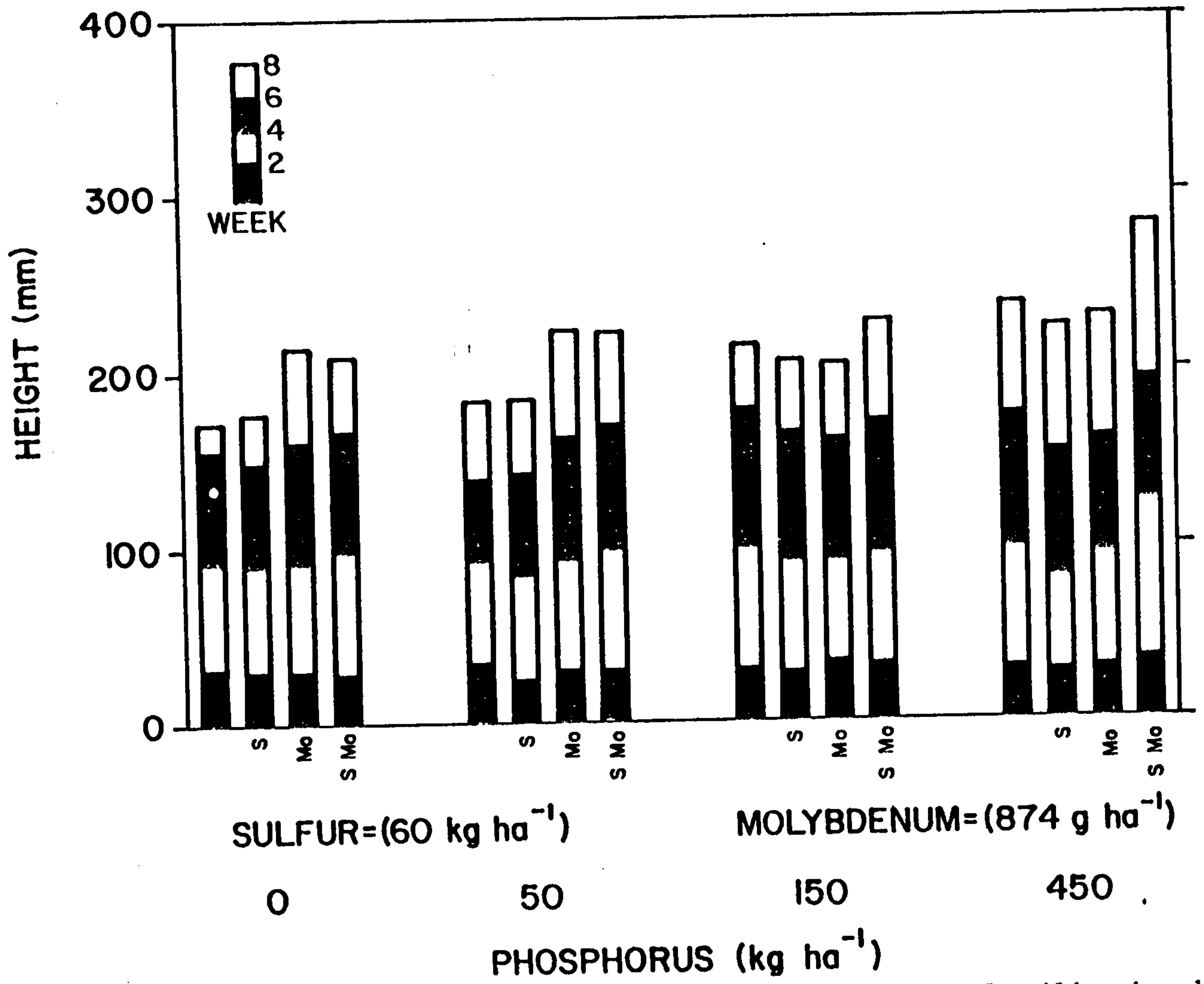

Figure 9. Birdsfoot trefoil height as influenced by P, S, and Mo fertilization during eight weeks of growth on a Gilpin silt loam in 1983.

This was attributed to the salt effect from the high rate of P and low rainfall.

Dry Matter Yield Birdsfoot trefoil yield was increased 36% beyond the non-fertilized P treatment using 50 kg P ha^{-1}. An additional 64% increase in yield was observed when applied P was increased from 50 to 450 kg P ha^{-1} (Table 18; Figure 10). However, the number of plants per sub-plot within the highest P treatment was low compared to the 50 kg P ha^{-1} treatment because of the salt effect mentioned previously. Red clover and flatpea yields were not significantly increased with P fertilization. This was attributed to rodent damage of red clover, and the slow germinating seeds and seedling growth of flatpea (Figures 11 and 12).

At harvest, birdsfoot trefoil yield was therefore enhanced as a result of an increase in seedling vigor. This was supported by Qualls and Cooper (1968) since total yield was correlated with increased seedling vigor. The 36% increase in yield using 50 kg P ha^{-1} was similar to the results of Lutz (1973) who observed a 20% increase in alfalfa with the same rate of applied P on a Fredrick silt loam originally containing 8 mg P kg^{-1} double acid extractable P.

Plant Tissue Analysis. Phosphorus applied at 50, 150, and 450 kg P ha^{-1} increased red clover and birdsfoot trefoil P concentrations an average of 1.26, 1.36, and 1.67 times, respectively relative to the control (Tables 19 and 20). Flatpea P concentration was only increased 1.09, 1.24, and 1.30 fold above the non-fertilized P treatment using the same applied P rates. These increases in plant P were

Table 18. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on red clover, birdsfoot trefoil, and flatpea yield after eight weeks of growth on a Gilpin silt loam in 1983.

Source	df†	Red Clover	Birdsfoot Trefoil	Flatpea
Main Effects				
P	3	NS‡	*	NS
0 P vs. 1,2,3 P§	1	NS	NS	NS
Linear 1,2,3 P	1	NS	*	NS
Quadratic 1,2,3 P	1	NS	NS	NS
S	1	NS	NS	NS
Mo	1	NS	NS	NS
Interactions				
P x S	3	NS	NS	NS
P x Mo	3	NS	NS	NS
S x Mo	1	NS	NS	NS
P x S x Mo	3	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degree of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150 and 450 kg P ha^{-1}.

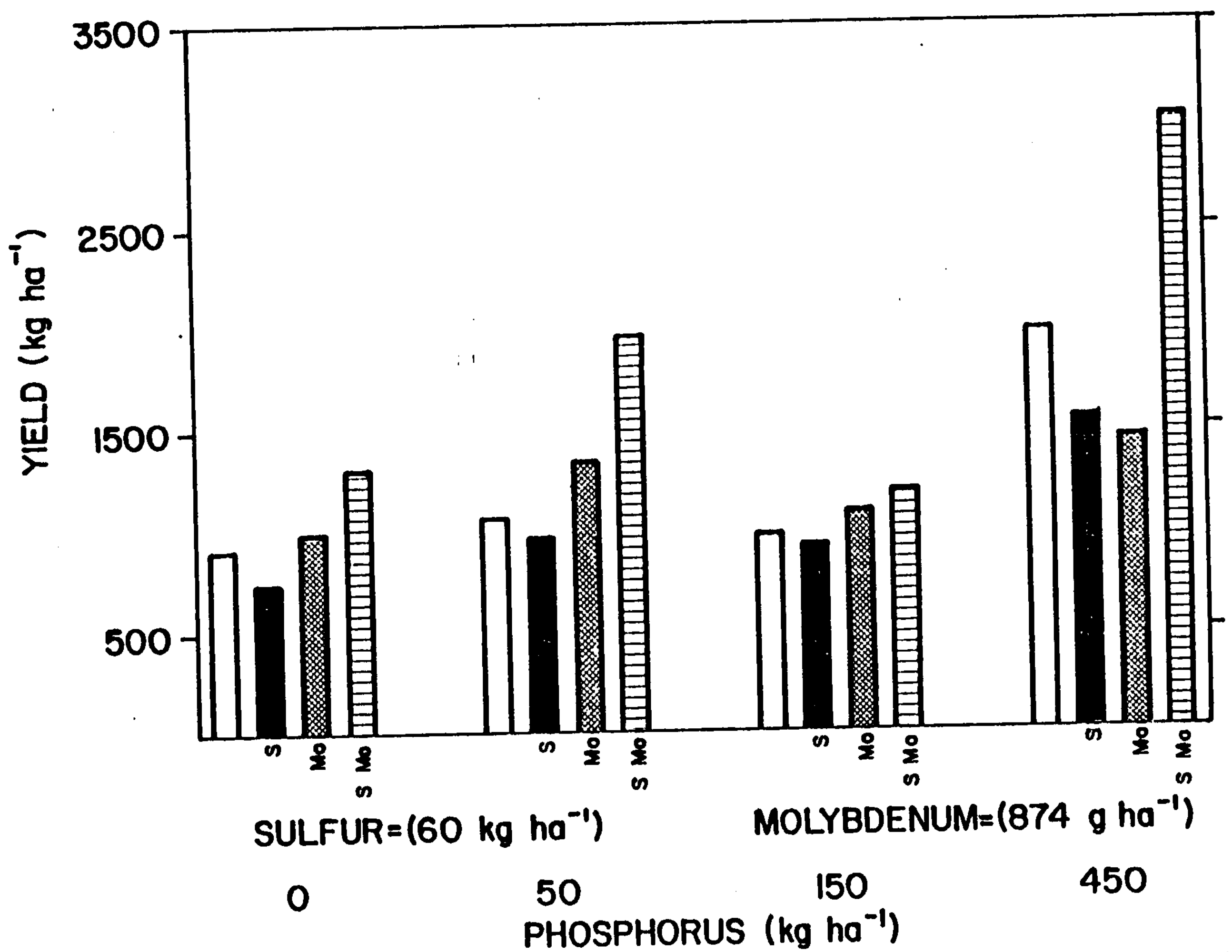

Figure 10. Birdsfoot trefoil dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1983.

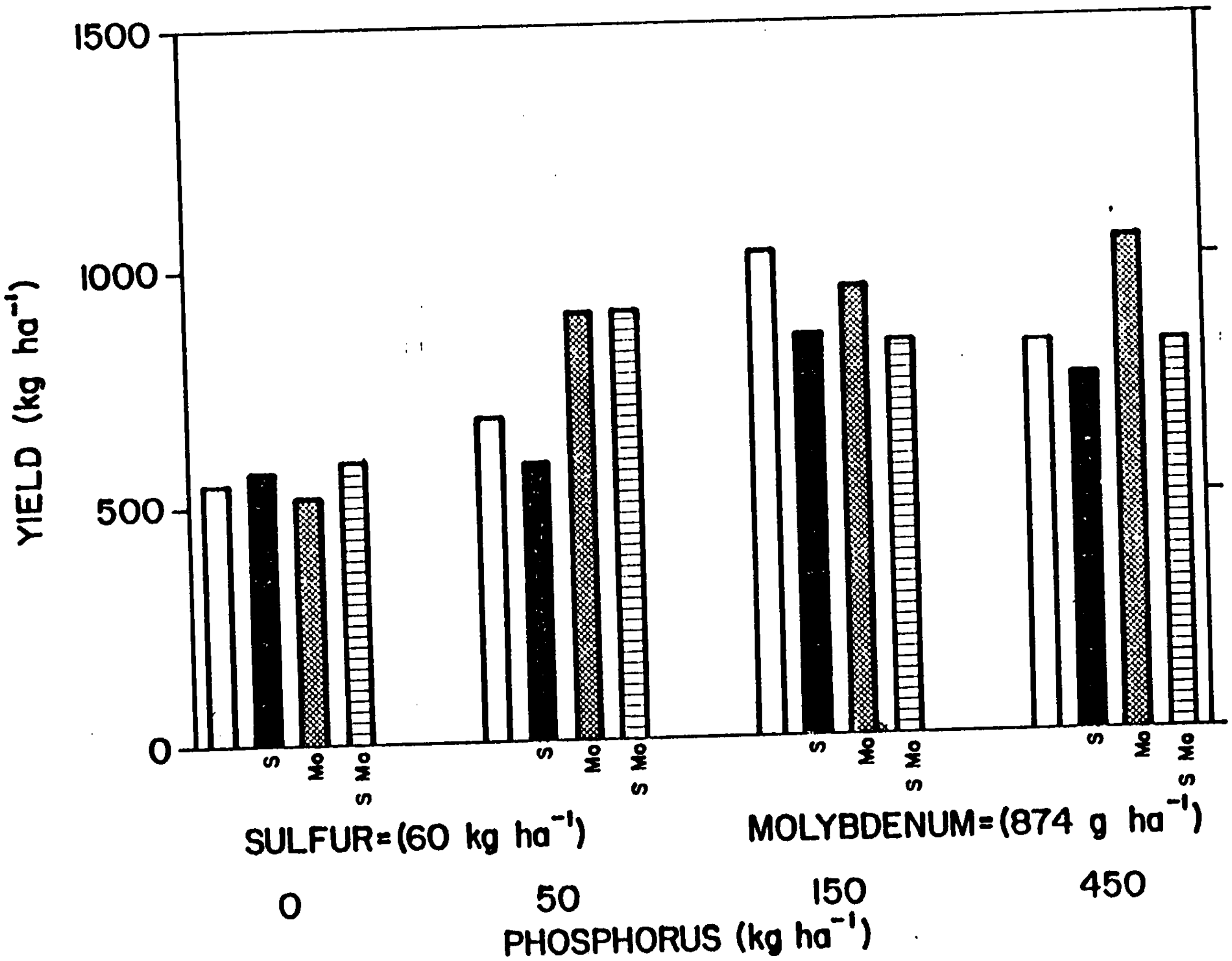

Figure 11. Red clover dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1983.

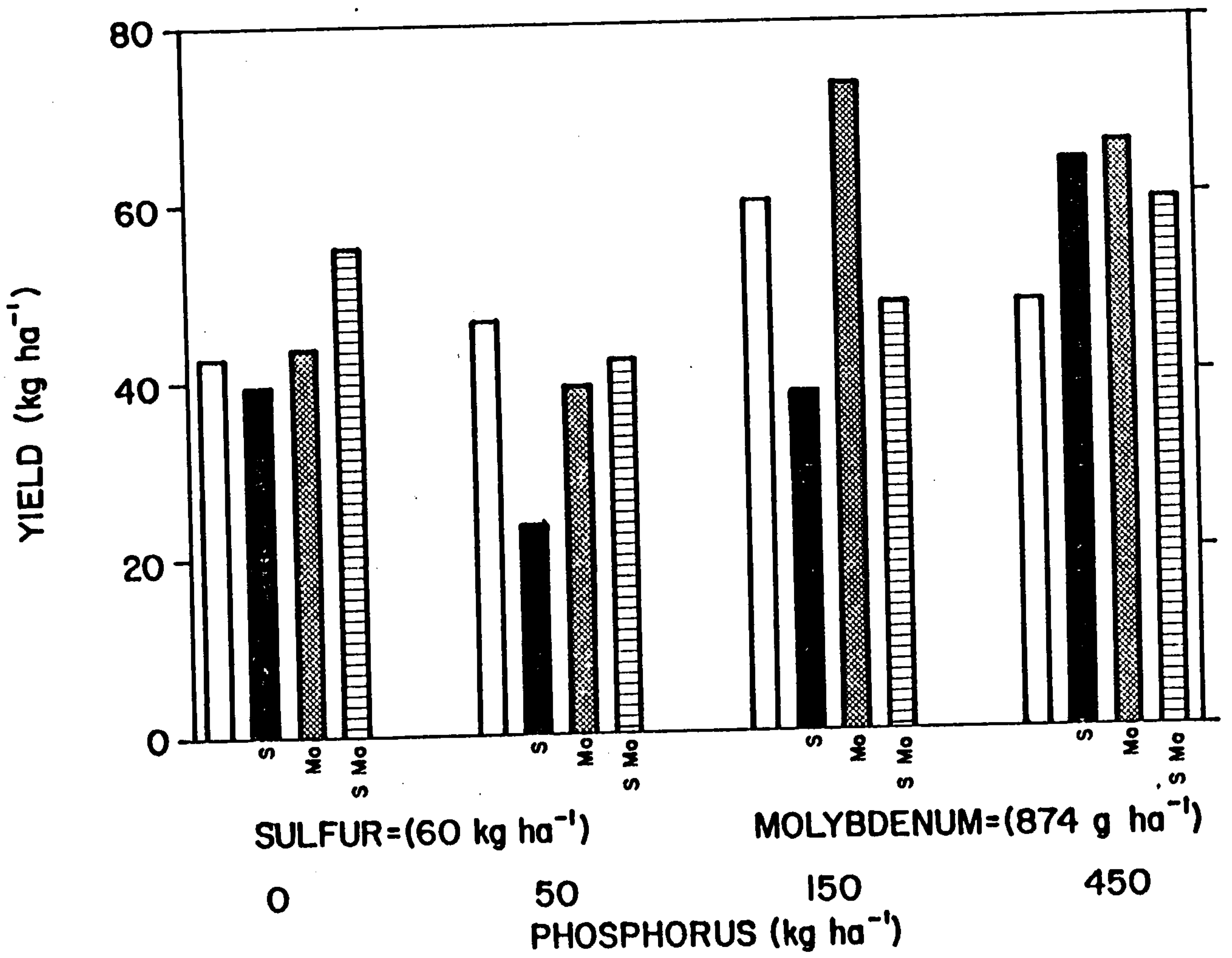

Figure 12. Flatpea dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1983.

Table 19. Statistical significance levels based on F tests, for the effects of P, S, and Mo fertilization on red clover, birdsfoot trefoil, and flatpea tissue concentrations of P, S, Mo, and N after eight weeks of growth on a Gilpin silt loam in 1983.

Source	df†	Red clover				Birdsfoot trefoil				Flatpea			
		P	S	Mo	N	P	S	Mo	N	P	S	Mo	N
Main Effects													
P	3	**	*	NS	NS	**	*	NS	*	*	NS	NS	NS
0 P vs. 1,2,3 P§	1	**	NS	NS	NS	**	NS	NS	*	**	NS	NS	NS
Linear 1,2,3 P	1	**	**	NS	NS	**	NS	NS	*	*	NS	NS	NS
Qaudratic 1,2,3 P	1	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS
S	1	NS	**	NS	NS	NS	**	NS	NS	NS	*	NS	NS
Mo	1	NS	NS	**	*	**	**	**	**	NS	NS	**	**
Interactions													
P x S	3	*	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS
P x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS
P x S x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150, and 450 kg P ha^{-1}, respectively.

Table 20. Red clover, birdsfoot trefoil, and flatpea tissue concentrations of P, S, Mo, and N as affected by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1983.

Treatment			Red Clover				Birdsfoot Trefoil				Flatpea			
P	S	Mo	P	S	Mo	N	P	S	Mo	N	P	S	Mo	N
kg ha^{-1}	kg ha^{-1}	g ha^{-1}	------mg kg^{-1}-----			g kg^{-1}	-----mg kg^{-1}------			g kg^{-1}	----mg kg^{-1}-------			g kg^{-1}
0	0	0	1975	1802	0.05	26.3	2150	2262	0.09	25.5	2550	2302	0.15	36.1
	60	0	2050	2045	0.07	29.6	2250	2850	0.09	17.8	2950	2425	0.15	39.2
	0	874	1900	1915	3.67	31.0	2100	2282	4.97	31.7	2625	2225	2.13	41.5
	60	874	1900	1914	1.90	29.5	2200	2707	5.03	31.6	2500	2275	1.26	40.9
50	0	0	2475	1805	0.10	29.1	2800	2217	0.09	26.9	2700	1977	0.09	35.1
	60	0	2424	2025	0.13	28.5	3125	2957	0.05	26.5	2900	2662	0.28	36.0
	0	874	2525	1860	2.91	30.3	2600	2135	3.06	33.2	2850	2087	0.79	41.1
	60	874	2300	1987	2.13	30.2	2625	2360	2.90	33.2	3100	2317	1.68	44.3
150	0	0	2825	1867	0.11	28.2	2875	2147	0.52	25.9	3150	2247	0.17	34.7
	60	0	2725	1920	0.19	28.0	3225	2552	0.10	31.9	3250	2190	0.11	39.6
	0	874	2600	1732	2.53	28.9	2775	1917	1.99	32.2	3300	2207	1.35	41.1
	60	874	2750	1975	1.14	30.8	2775	2362	3.74	32.3	3475	2272	1.65	43.4
450	0	0	3225	2002	0.17	29.7	3725	2505	0.10	31.1	3525	2147	0.13	37.8
	60	0	3575	2280	0.18	27.8	3726	2917	0.27	29.0	3400	2207	0.18	39.1
	0	874	3125	2050	3.25	30.1	3425	2402	1.45	34.0	3650	2097	1.09	40.4
	60	874	3625	2127	1.22	30.2	3175	2440	1.50	32.0	3275	2307	0.85	36.8

of the same magnitude as those obtained by Lutz (1973). Critical P concentrations can differ between legume species. Red clover P tissue level was approximately 1950 mg kg^{-1} without applied P. Birdsfoot trefoil and flatpea P concentration averaged 2150 and 2650 mg kg^{-1}, respectively, within the control. These P concentrations were equivalent to, and greater than the P concentrations (1500 to 1800 mg P kg^{-1}) obtained in the greenhouse experiment after the application of 22 mg P kg^{-1} (Tables 7 and 8). However, the P concentration in birdsfoot trefoil without applied P could be regarded as deficient since yield was increased with P fertilization. The initial P levels in red clover and flatpea may also be low, but due to red clover leaf defoliation and the slow growth of flatpea, insufficient growth differences were observed in order to make this assessment.

Phosphorus fertilization had opposing effects on red clover and birdsfoot trefoil total S concentrations (Table 20). Red clover increased in total S 1.03 times while birdsfoot trefoil total S was reduced 1.05 fold with applied P, but this may have been a dilution affect due to increasing dry matter with the trefoil. Since the legume species responded differently, it is difficult to make any general assessment of the effect of P fertilization on total plant S (Tables 19 and 20).

Red clover and birdsfoot trefoil total Mo concentrations increased approximately 2.5 fold when fertilizer P increased from 0 to 150 kg ha^{-1} (without applied Mo) possibly by forming a readily available phosphomolybdate anion as proposed by Singh and Kumar (1979). Another possible explanation is that increased PO_4^{3-} in the soil solution may exchange with unavailable MoO_4^{2-} from Fe and Al hydrous oxides thereby increasing plant available Mo.

Total plant N was increased from approximately 29.1 to 31.5 g kg^{-1} in birdsfoot trefoil when P was increased from 0 to 450 kg ha^{-1} (Tables 19 and 20). Effective symbiosis as reported by Griffith (1978) was most likely increased with P fertilization. Haynes and Ludecke (1981) also observed an increase in white clover total N with P fertilization.

Available Soil Phosphorus. Bray-1 extractable P from the soils within the red clover and birdsfoot trefoil plots increased beyond the control approximately 3, 7, and 18 times with each increment of fertilizer P (Tables 21 and 22). Plant available P averaged 12 mg kg^{-1} in the sub-plots not receiving P applications. This was approximately the level of extractable P observed in the greenhouse experiment after the initial application of 22 mg P kg^{-1} (Table 10). However, since birdsfoot trefoil yield increased 36% in the field with applied P this initial available P level could be regarded as low. Similar results were also observed in earlier work by Lutz (1973).

Sulfur Fertilization

Height. Sulfur fertilization did not significantly increase red clover or birdsfoot trefoil height during the eight weeks of growth (Table 17; Figures 8 and 9). As a result, legume seedling vigor with

Table 21. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on Bray-1 extractable P and total S in the Gilpin silt loam soil utilized in the 1983 field experiment.

		Red clover plots		Birdsfoot trefoil plots	
Source	df†	P	S	P	S
Main Effects					
P	3	**	NS‡	**	NS
0 P vs. 1,2,3 P§	1	**	NS	**	NS
Linear 1,2,3 P	1	**	NS	**	NS
Quadratic 1,2,3 P	1	NS	NS	NS	NS
S	1	NS	*	NS	**
Mo	1	NS	NS	NS	NS
Interactions					
P x S	3	NS	NS	**	NS
P x Mo	3	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS
P x S x Mo	3	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150, and 450 kg P ha^{-1}, respectively.

Table 22. Soil pH, Bray-1 extractable P, and total soil S as influenced by P, S, and Mo fertilization of a Gilpin silt loam in the 1983 field experiment.†

Treatment			Red Clover		Birdsfoot Trefoil	
P	S	Mo	P	S	P	S
kg ha^{-1}	kg ha^{-1}	g ha^{-1}	mg kg^{-1}			
0	0	0	12	250	10	251
	60	0	10	271	14	313
	0	874	10	258	12	256
	60	874	11	309	14	309
50	0	0	41	278	35	278
	60	0	23	294	54	328
	0	874	25	261	37	296
	60	874	27	303	35	315
150	0	0	68	258	86	290
	60	0	76	285	104	317
	0	874	68	293	72	281
	60	874	71	285	93	318
450	0	0	184	268	218	288
	60	0	169	271	220	323
	0	874	232	259	263	304
	60	874	180	284	208	312

† Soil pH ranged from 4.9 to 5.1.

respect to height was not increased with applied S. Low rainfall during the first six weeks after seeding probably inhibited an early height response to applied S (Appendix Table 9).

Dry Matter Yield. Sulfur fertilization increased birdsfoot trefoil yield 19%, however, because of variability among replications, the legume yield increase was not statistically significant with applied S (Table 18; Figures 10, 11, and 12; Appendix Table 8). This percent increase in yield was lower than the increases obtained by McLachlin and DeMarco (1973), and Pumphrey and Moore (1965b) when S was applied at approximately the same rate to subterranean clover and alfalfa on S deficient soils in Australia and California.

Plant Tissue Analysis. Sulfur fertilization did not significantly affect plant tissue concentrations of P, Mo, or N in red clover, birdsfoot trefoil, or flatpea (Tables 19 and 20). Applied S did increase total plant S within each legume specie. Red clover tissue S concentrations increased from approximately 1900 to 2000 mg S kg^{-1}; birdsfoot trefoil tissue S concentrations increased from 2200 to 2650 mg S kg^{-1}, and flatpea increased from 2150 to 2350 mg S kg^{-1} when S was applied at 60 kg ha^{-1} relative to 0 kg S ha^{-1}. Plant species can vary in tissue concentrations of S, however, it is suspected that birdsfoot trefoil and flatpea total S concentrations without applied S were not S deficient. The S concentration observed in red clover was not S deficient in referance to studies conducted by Walker and Bentley (1961), and also because yield increases were not observed from S applications (Appendix Table 11).

Sulfur fertilization did not decrease the legume N:S ratios. Red clover total N:S ratio varied between 14 to 16, with or without applied S, which according to Tisdale (1977) can be associated with maximum yields. Ruminant animals can also utilize these legumes efficiently with respect to their N:S ratios according to Coleman (1966).

Total Soil Sulfur. Total soil S increased from approximately 272 to 312 mg kg^{-1} when fertilizer S increased from 0 to 60 kg ha^{-1} (Tables 21 and 22). It is suspected however, that available SO_4^{2-} is not deficient on this soil because of the negligible growth response and adequate plant tissue S concentrations with or without S fertilization. Coal operated electrical power plants in the Ohio valley and western neighboring states release atmospheric SO_2 that may provide sufficient soil S levels for plant uptake. Beaton et al. (1971) reported that only coarse-textured and neutral (pH 7) soils in West Virginia would be low in available S.

Molybdenum Fertilization

Height. Birdsfoot trefoil height was increased approximately 10% beyond the non-fertilized Mo plots using 874 g Mo ha^{-1} during weeks 3 through 8 (Table 17; Figure 9). Molybdenum fertilization did not increase height within the first 2 weeks of measurement. Low rainfall during this period could have been the limiting factor since Mo has been noted to affect legumes at an early growth stage (Gupta and Lipsett, 1981). Molybdenum fertilization increased height within the

0 kg P ha^{-1} treatment which contrasted to the 0 mg P kg^{-1} rate within the greenhouse experiment. According to Anderson (1956b) P or S deficiencies need to be corrected before legumes can respond to applied Mo, and therefore, this evidence implies that plant available P in the control plots was adequate.

Dry Matter Yield. Birdsfoot trefoil yield increased beyond the 0 g Mo ha^{-1} treatment by 36% when Mo was applied at 874 g ha^{-1} (Table 19; Figure 10). Yield was also increased within the 0 kg P ha^{-1} treatment with fertilizer Mo. Red clover and flatpea yield did not respond to Mo fertilization (Figures 11 and 12). Leaf defoliation of red clover, and slow flatpea seed germination and seedling growth were the factors contributing to this lack of response.

Plant Tissue Analysis. Molybdenum fertilization decreased birdsfoot trefoil tissue concentration of P and S (Tables 19 and 20). Since trefoil yield was increased with applied Mo, the decrease in tissue P and S was therefore caused by a dilution effect. This decrease in tissue P concentration with increasing Mo fertilization was also observed during the greenhouse experiment (Tables 7 and 8). Phosphorus uptake when calculated from Table 20 and Appendix Table 10 increased with applied Mo. These results are supported by Shulka and Pathak (1973) who reported that Mo fertilization improved the root system and vigor of berseem and may lead to a higher uptake of P.

Molybdenum fertilzation increased red clover tissue Mo from 0.25 to 2.34 mg Mo kg^{-1}; concentrations in birdsfoot trefoil increased from 0.16 to 3.08 mg Mo kg^{-1}; and flatpea concentrations increased from 0.15 to 1.47 mg Mo kg^{-1} (Tables 19 and 20). Red clover Mo concentration was not in the sufficiency range of 0.46 to 1.08 mg kg^{-1} without Mo fertilization (Gupta, 1970). Since birdsfoot trefoil and flatpea Mo concentrations were less than red clover without Mo fertilization, these species could also be Mo deficient. Different plant species can vary with respect to adequate Mo levels, however insufficient information regarding other legume tissue levels of Mo can support these findings.

The highest plant tissue concentration of Mo was 5.03 mg kg^{-1} when fertilized with 874 g Mo ha^{-1}. These plant tissue concentrations of Mo as reported by Gupta and Lipsett (1981) were well below a level of 10 mg Mo kg^{-1} which could lead to molybdenosis in cattle. Molybdenosis is a disease caused from high levels of Mo in forage. High Mo concentrations induce Cu deficiencies in ruminants and can result in death. Thus, fertilizing this particular soil with 874 g Mo ha^{-1} should not result in legume tissue concentrations of Mo toxic for animal consumption.

Total plant N increased 1.06, 1.16, and 1.11 times in red clover, birdsfoot trefoil, and flatpea, respectively, when Mo was applied to the soil (Tables 19 and 20). These results are in agreement with Mortvedt (1981) who showed that red, white, subterranean, and crimson clover total N increased when Mo was applied at 1.67 mg kg^{-1} to an acid Decatur silty clay. These increases in total N were a result of enhanced N_2-fixation from Mo fertilization because Mo serves as an electron carrier within the nitrogenase enzyme in legume nodules to reduce N_2 to NH_3.

Experiment 3. Legume Seedling Response to Phosphorus, Sulfur, and Molybdenum Fertilization in the 1984 Field Experiment.

Phosphorus Fertilization

Height. Red clover height during the fifth, sixth, seventh, and eighth week of measurement increased beyond the control by 19, 22, 27 and 28%, respectively using 50 kg P ha^{-1}. The 150 kg P ha^{-1} treatment only increased red clover height an average of 3% above the 50 kg P ha^{-1} treatment during the same time period. Height within the 450 kg P ha^{-1} rate was no greater than the height within the 50 kg P ha^{-1} treatment (Table 23; Figure 13; Appendix Table 13). Phosphorus applied at 50 kg ha^{-1} increased birdsfoot trefoil height above the non-fertilized P rate by 24, 18, 17, 25, 22,and 22% during the third through eighth week of measurement, respectively. There was no increase in height above the 50 kg P ha^{-1} rate using 150 kg P ha^{-1}, and only a 10% increase in height over the 50 kg P ha^{-1} treatment using the highest P rate (Table 23; Figure 14). Flatpea height was enhance 21% beyond the 0 kg P ha^{-1} treatment with P applied at 150 kg P ha^{-1} at week eight (Table 23; Figure 15).

Legume seedling vigor, as measured by plant height, was not increased with P fertilization within the first two weeks of measurement for trefoil, and red clover height did not increase until the fourth week of measurement. Only 1.78 cm of rain fell during the 3

Table 23. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on red clover, birdsfoot trefoil and flatpea height during the first eight weeks of growth in the 1984 field experiment.

Source	df†	Red clover Week 1	2	3	4	5	6	7	8	Birdsfoot trefoil Week 1	2	3	4	5	6	7	8	Flatpea week 1	2	3	4	5	6	7	8
Main Effects																									
P	3	NS‡	NS	NS	NS	NS	*	*	**	NS	NS	*	NS	*	**	**	**	NS	NS	NS	NS	NS	NS	NS	*
0 P vs. 1,2,3 P§	1	NS	NS	NS	NS	*	**	**	**	NS	NS	**	*	*	**	**	**	NS	NS	NS	NS	NS	NS	NS	*
Linear 1,2,3 P	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	**
Qaudratic 1,2,3 P	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S	1	NS	NS	NS	NS	NS	*	**	*	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Mo	1	NS	NS	NS	NS	NS	NS	**	*	NS	NS	NS	**	**	*	**	**	NS	*	NS	*	*	NS	NS	NS
Interactions																									
P x S	3	NS	NS	NS	NS	NS	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
P x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	*	*	**	*	NS	NS	NS	NS	NS	NS	NS	NS
P x S x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	**	*	**	*	**	*	NS	*	NS	NS	NS	NS	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150, and 450 kg P ha^{-1}, respectively.

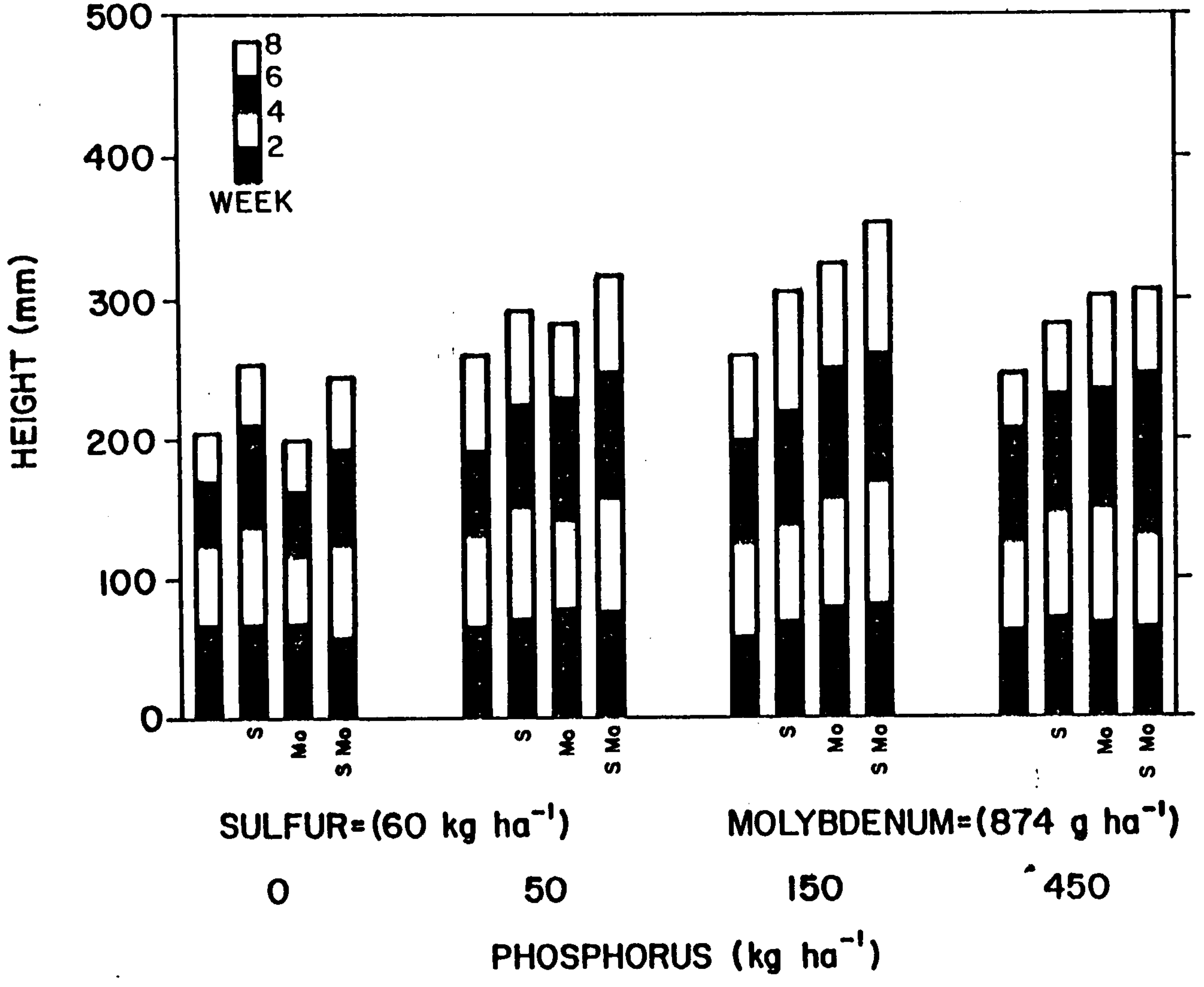

Figure 13. Red clover height as influenced by P, S, and Mo fertilization during eight weeks of growth on a Gilpin silt loam in 1984.

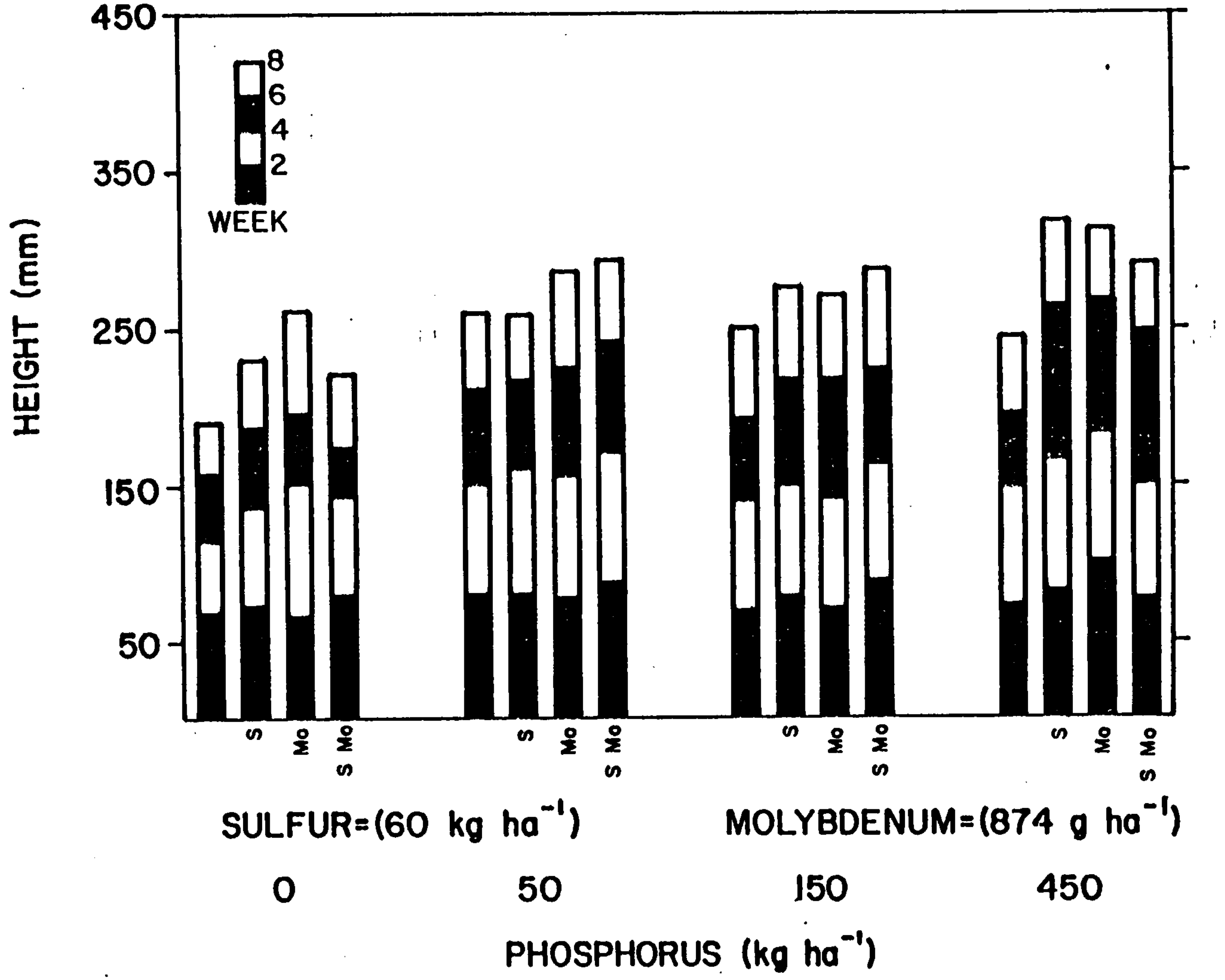

Figure 14. Birdsfoot trefoil height as influenced by P, S, and Mo fertilization during eight weeks of growth on a Gilpin silt loam in 1984.

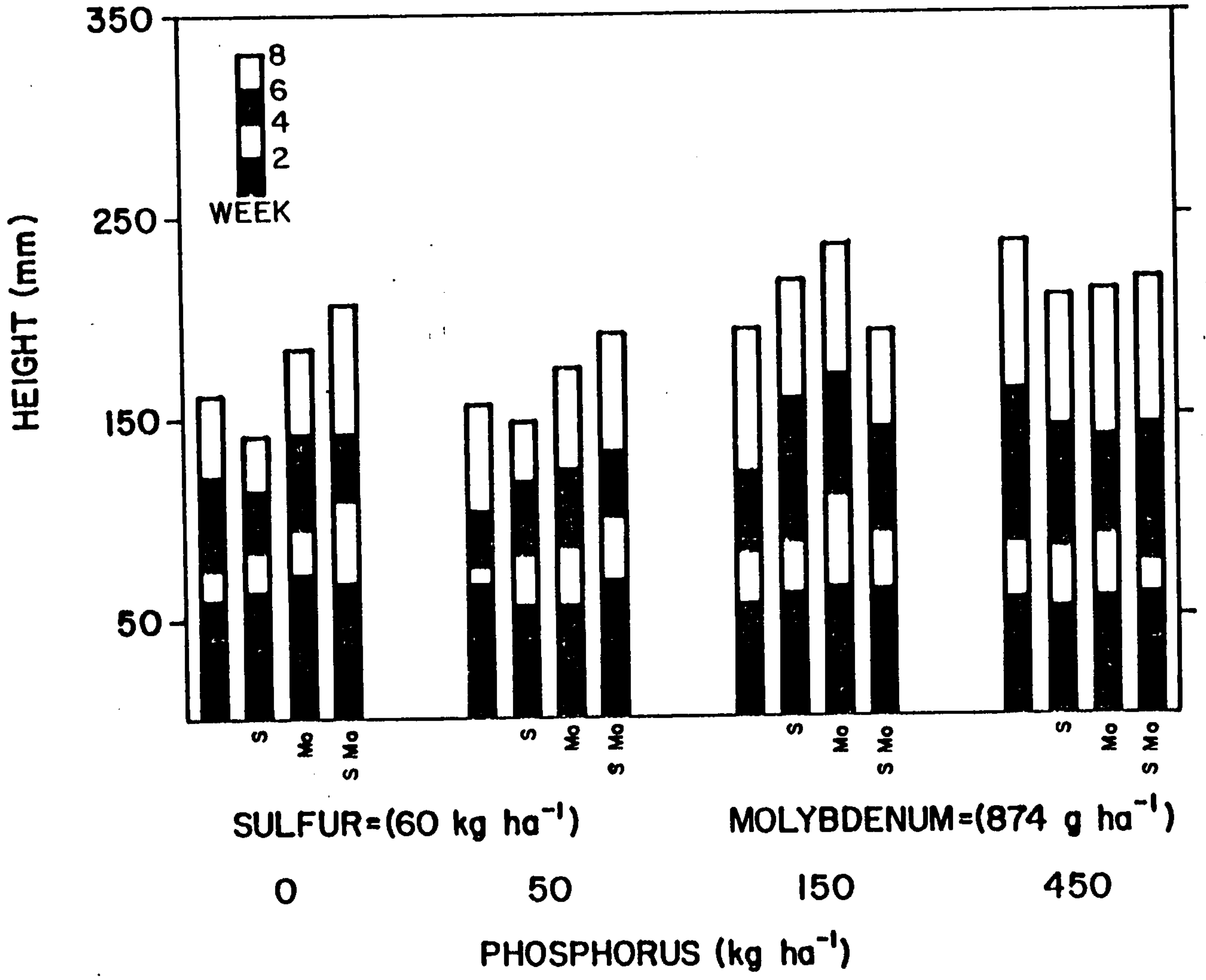

Figure 15. Flatpea height as influenced by P, S, and Mo fertilization during eight weeks of growth on a Gilpin silt loam in 1984.

weeks after seeding, and the seedlings remained essentially dormant until an additional 6.6 cm of rain fell 2.5 weeks later. As a result, the beginning height measurements were delayed 2.5 weeks past the time when the first field experiment heights were recorded in 1983 (Appendix Table 14). Adequate rain fell in July and the legumes began to increase in height in response to applied P. Flatpea height, near the end of the experiment, increased with fertilizer P as opposed to the previous year. Two factors which may have accounted for this were; (1) the seeds were scarafied with H_2SO_4 which increased seed germination and; (2) the legumes were 2.5 weeks older than seedlings measured the previous year. Percent legume stand in the highest P treatment was poor in comparison to the other P rates and was again attributed to excessive salt concentrations.

Dry Matter Yield. Phosphorus fertilization increased red clover yield 98% beyond the control when applied at 50 kg ha^{-1} (Table 24; Figure 16). Birdsfoot trefoil yield increased 51% above the non-fertilized P treatment with 50 kg P ha^{-1} (Figure 17; Appendix Table 15). Phosphorus applied at 150 kg ha^{-1} enhanced flatpea yields 40% over the 0 kg P ha^{-1} treatment (Table 14; Figure 18). Thus, seedling vigor as measured by dry matter yield was improved with P fertilizaton.

Plant Tissue Analysis. Phosphorus applied at 50, 150, and 450 kg P ha^{-1} increased each legume species P concentration approximately 1.22, 1.37 and 1.52 fold, respectively, beyond the control (Tables 25 and 26). The P tissue concentration in each legume specie was

Table 24. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on red clover, birdsfoot trefoil, and flatpea yield in the 1984 field experiment.

Source	df†	Red Clover	Birdsfoot Trefoil	Flatpea
Main Effects				
P	3	*	NS‡	*
0 P vs. 1,2,3 P§	1	*	NS	NS
Linear 1,2,3 P	1	NS	NS	NS
Quadratic 1,2,3 P	1	NS	NS	**
S	1	*	NS	NS
Mo	1	*	*	**
Interactions				
P x S	3	NS	NS	NS
P x Mo	3	NS	NS	NS
S x Mo	1	NS	*	NS
P x S x Mo	3	NS	**	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150 and 450 kg P ha^{-1}.

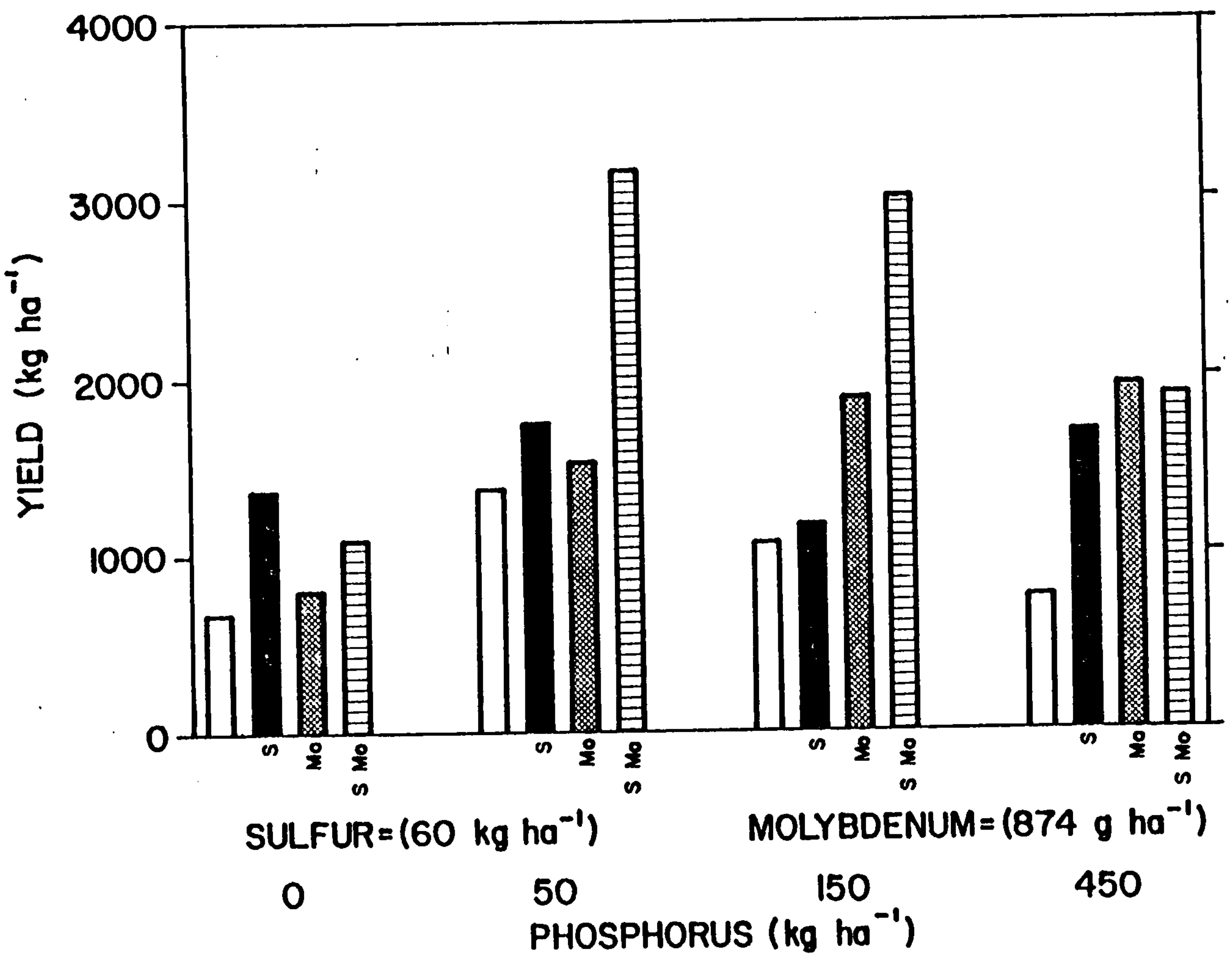

Figure 16. Red clover dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1984.

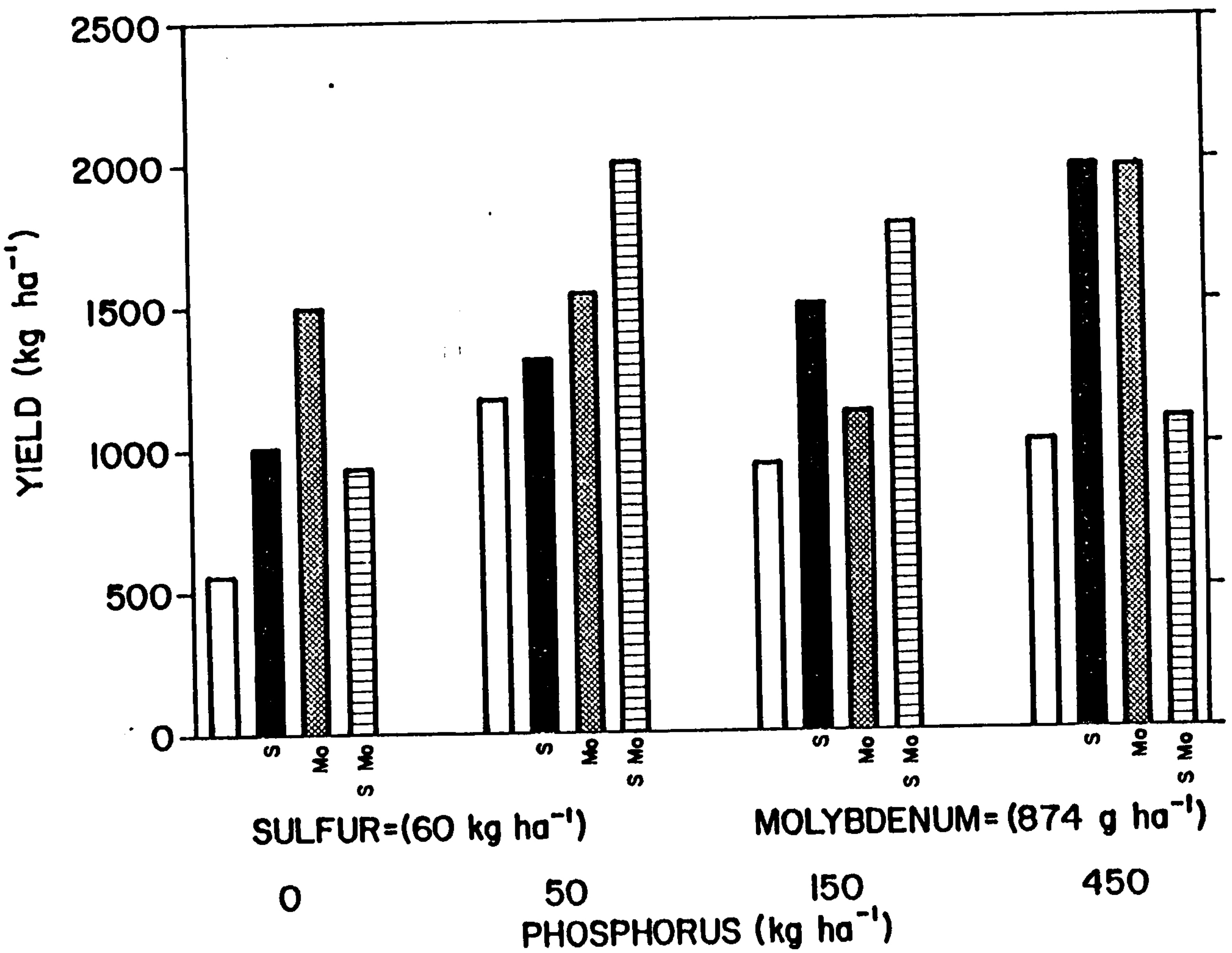

Figure 17. Birdsfoot trefoil dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1984.

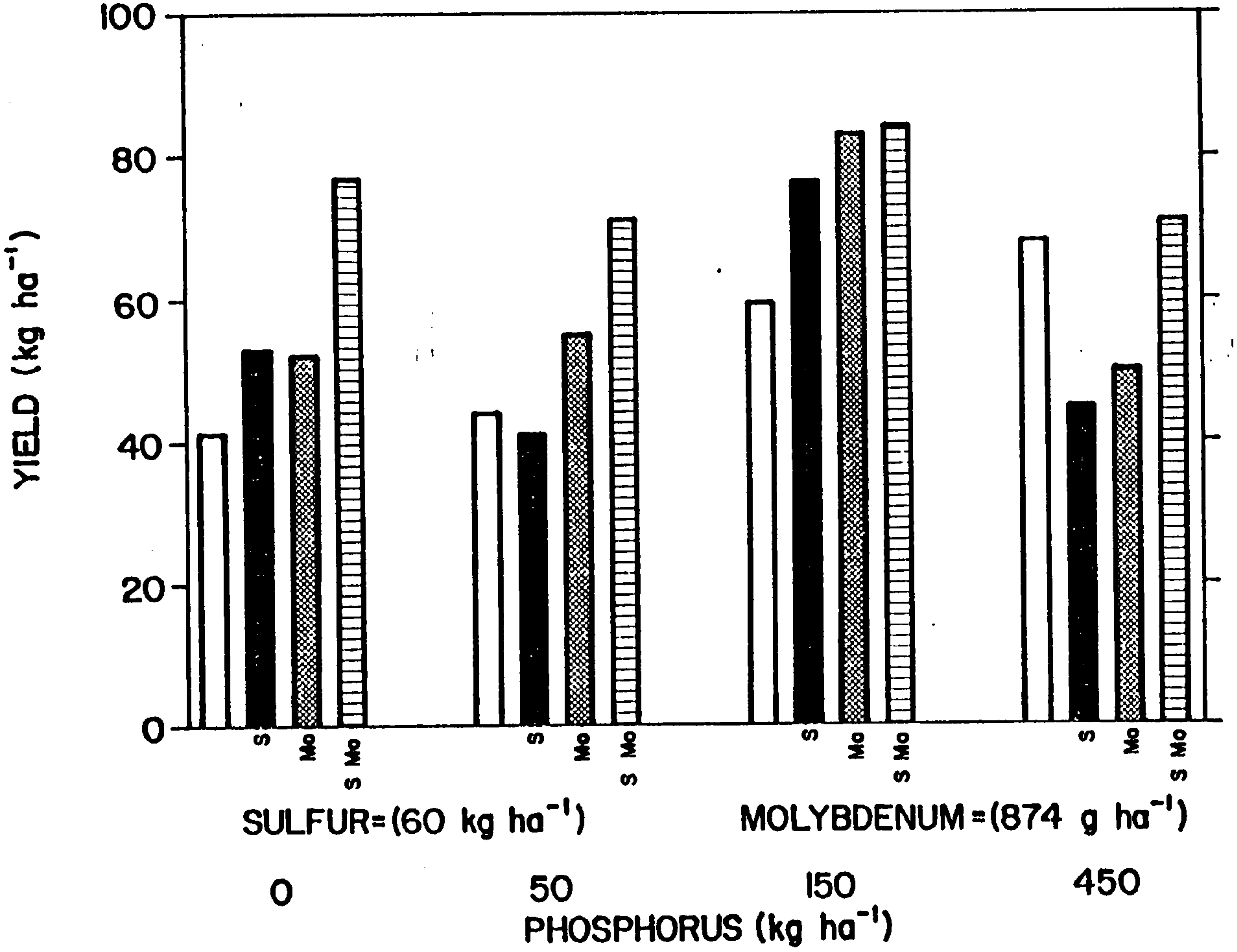

Figure 18. Flatpea dry matter yield as influenced by P, S, and Mo fertilization after eight weeks of growth on a Gilpin silt loam in 1984.

greater than 2000 mg kg^{-1} without applied P, and therefore may not be initially regarded as P deficient according to the work of Ozanne (1980). However, yield increases occurred in this experiment and the largest increase in yield normally occurred with 50 kg P ha^{-1}. This level of applied P was associated with plant P concentrations greater than 2500 mg kg^{-1} (Table 26).

Total plant N was increased in birdsfoot trefoil from approximately 28.2 to 31.0 g kg^{-1} when fertilizer P was increased from 0 to 450 kg ha^{-1} (Tables 25 and 26). Red clover and flatpea total N concentrations did not increase. Similar results by Griffith (1978), and Haynes and ludecke (1981) indicated that effective symbiosis increased as available P increased with P fertilization.

Available Soil Phosphorus. Bray-1 extractable P from the soils within the red clover, birdsfoot trefoil and flatpea plots increased beyond the 0 kg P ha^{-1} treatment by 2, 4, and 13 fold with each increasing addition of fertilizer P (Tables 27 and 28). Plant available P averaged 13.0 mg kg^{-1} within the non-fertilized sub-plots. This level of available P was approximately the same as the concentration of P in the greenhouse experiment after fertilizing with 22 mg P kg^{-1} (Table 6). After an addition of 50 kg P ha^{-1} the Bray-1 extractable P level increased to approximately 26 mg kg^{-1} and this was associated with the largest increase in growth.

Sulfur Fertilization

Height. Fertilizer S at 60 kg ha^{-1} increased red clover height beyond the control by 11, 11, and 13%, respectively during measurement

Table 25. Statistical significance levels based on F tests, for the effects of P, S, and Mo fertilization on red clover, birdsfoot trefoil, and flatpea tissue concentrations of P, S, Mo, and N in the 1984 field experiment.

Source	df†	Red Clover				Birdsfoot Treofil				Flatpea			
		P	S	Mo	N	P	S	Mo	N	P	S	Mo	N
Main Effects													
P	3	**	NS‡	NS	NS	**	NS	NS	NS	**	*	NS	NS
0 P vs. 1,2,3 P§	1	**	NS	NS	NS	**	NS	NS	*	**	**	NS	NS
Linear 1,2,3 P	1	**	NS	NS	NS	**	NS	NS	NS	*	*	NS	*
Quantitative 1,2,3 P	1	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS
S	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS
Mo	1	*	NS	**	NS	NS	NS	**	**	NS	*	**	*
Interactions													
P x S	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
P x Mo	3	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS
P x S x Mo	3	NS	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150, and 450 kg P ha^{-1}, respectively.

Table 26. Red clover, birdsfoot trefoil, and flatpea tissue concentrations of P, S, Mo, and N as affected by P, S, and Mo fertilization in the 1984 field experiment.

Treatment			Red Clover				Birdsfoot Trefoil				Flatpea			
P	S	Mo	P	S	Mo	N	P	S	Mo	N	P	S	Mo	N
kg ha-1		g ha^{-1}	mg kg-1			g kg-1	mg kg-1			g kg-1	mg kg-1			g kg-1
0	0	0	2075	1940	0.08	33.0	2025	2595	0.08	24.5	2375	2250	0.15	38.9
	60	0	2175	2130	0.13	33.3	2100	2802	0.13	28.4	2450	2477	0.26	40.4
	0	874	2200	2070	3.06	36.1	2050	2275	2.03	30.2	2575	2440	1.55	41.5
	60	874	2200	2097	4.24	34.8	1875	2862	2.85	29.8	2700	2465	1.58	46.2
50	0	0	2625	1887	0.09	32.2	2725	2245	0.13	28.3	2775	2207	0.19	39.7
	60	0	2525	2032	0.11	33.6	2600	2852	0.07	29.1	3050	2435	0.15	42.1
	0	874	2575	1945	4.08	35.0	2475	2065	3.02	30.8	3100	2400	0.97	43.4
	60	874	2575	1975	3.21	35.0	2600	2410	2.57	31.8	2850	2397	2.94	44.8
150	0	0	2825	2042	0.12	34.7	3050	2455	0.10	28.3	3225	2335	0.17	39.0
	60	0	2825	2037	0.13	35.4	3325	2512	0.13	28.7	3033	2290	0.18	43.2
	0	874	2875	1950	3.58	35.6	2950	2247	1.21	34.9	3275	2317	1.33	39.7
	60	874	2875	1965	3.68	33.2	3125	2595	1.93	35.9	3100	2447	1.59	40.5
450	0	0	2900	1900	0.10	32.5	3750	2417	0.12	27.7	3475	2232	0.15	37.1
	60	0	3075	2057	0.15	34.2	3275	2500	0.15	31.6	3375	2260	0.15	35.8
	0	874	3400	1965	2.63	34.0	3375	2420	1.72	33.3	3300	2232	1.32	38.6
	60	874	3350	2015	2.40	33.3	3800	2552	2.30	30.8	3250	2382	1.48	36.3

Table 27. Statistical significance levels, based on F tests, for the effects of P, S, and Mo fertilization on Bray-1 extractable P, total S, and resin extractable Mo in the soils utilized in the 1984 field experiment.

		Red clover			Birdsfoot trefoil		Flatpea	
Source	df†	P	S	Mo	P	S	P	S
Main Effects								
P	3	**	NS‡	NS	**	NS	**	NS
0 P vs. 1,2,3 P§	1	**	NS	NS	**	NS	**	NS
Linear 1,2,3 P	1	**	NS	NS	**	NS	**	NS
Quadratic 1,2,3 P	1	NS	NS	*	NS	NS	NS	NS
S	1	NS	**	NS	NS	**	NS	*
Mo	1	NS	NS	**	NS	*	NS	NS
Interactions								
P x S	3	NS	NS	NS	NS	NS	NS	NS
P x Mo	3	NS	NS	NS	NS	NS	NS	NS
S x Mo	1	NS	NS	NS	NS	NS	NS	NS
P x S x Mo	3	NS	NS	NS	NS	NS	*	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 P represents 0, 50, 150 and 450 kg P ha^{-1}.

Table 28. Soil pH, Bray-1 extractable P, and total soil S as influenced by P, S, and Mo fertilization in the 1984 field experiment.†

Treatment			Red Clover			Birdsfoot Trefoil		Flatpea	
P	S	Mo	P	S	Mo	P	S	P	S
--kg ha^{-1}--		g ha^{-1}	-mg kg^{-1}--		-μg kg^{-1}-	--mg kg^{-1}--		--mg kg^{-1}--	
0	0	0	15	217	12	15	206	19	229
	60	0	12	239	12	11	243	15	251
	0	874	16	225	85	7	223	16	232
	60	874	16	249	95	7	257	11	261
50	0	0	21	231	18	25	228	23	252
	60	0	18	265	14	23	224	26	266
	0	874	20	240	62	15	226	21	230
	60	874	25	260	76	22	238	26	258
150	0	0	45	233	18	49	211	62	253
	60	0	60	244	14	42	236	56	256
	0	874	50	229	96	37	238	61	264
	60	874	45	257	100	46	259	62	277
450	0	0	170	222	19	175	234	179	269
	60	0	158	248	21	165	249	139	273
	0	874	156	247	84	153	226	161	263
	60	874	153	244	92	170	264	202	257

† Soil pH ranged from 4.9 to 5.3

weeks six, seven, and eight (Table 23; Figure 13). Birdsfoot trefoil and flatpea height was unaffected by S fertilization (Figures 14 and 15). However, these three legume species did not show any S deficiency symptoms such as yellow, slender leaves, and stunted growth, as reported by Rhykerd and Overdahl (1972). Low rainfall during the first 5.5 weeks after seeding may have inhibited an early height increase to fertilizer S.

Dry Matter Yield. Sulfur fertilization increased red clover yield 56% above the non-fertilized S treatment. However, birdsfoot trefoil and flatpea yields were increased approximately 17% with S fertilization (Table 24; Figures 16, 17, and 18). Variability between species could account for such a large yield increase of red clover with applied S. Though these legumes did increase in yield using 60 kg S ha^{-1}, the response was much lower than that obtained by Meyer and Marcum (1980) who received over a 200% increase in alfalfa yields when 50 kg S ha^{-1} was applied to the S deficient soil in California.

Plant Tissue Analysis. Sulfur fertilization did not increase plant tissue concentrartions of P, Mo or N in any of the legume species (Tables 19 and 20). Applied S only increased total plant S in flatpea 1.04 times. Total S averaged over all species was 2300 mg kg^{-1} without applied S and 2400 mg kg^{-1} with S fertilization. Total S can vary between plant species, but if the critical concentration of 2000 mg S kg^{-1} reported by Walker and Bentley (1961) for red clover is utilized as a reference, then all of the legumes contained sufficient S with S fertilization.

Total N:S ratios within each legume species did not decrease when S was applied (Appendix Table 16). Accoding to Tisdale (1977) and Coleman (1966) the observed N:S ratios are adequate for maximum yields and the forage can be utilized efficiently by ruminant animals. In both field experiments initial soil S can be considered adequate for maximum forage production on the basis of the unaffected N:S ratio with applied S.

Total Soil Sulfur. Total soil S increased from approximately 234 to 253 mg kg^{-1} when fertilizer S was increased from 0 to 60 kg ha^{-1} (Tables 27 and 28). Available sulfate-S is not suspected to be deficient in this soil because of the adequate red clover tissue S concentration and the sufficient total N:S ratios of the legumes without applied S.

Molybdenum Fertilization

Height. Molybdenum fertilization increased red clover height during the seventh and eighth week of measurement by approximately 12.5% beyond the non-fertilized Mo treatment (Table 23; Figures 13, 14, and 15). Birdsfoot trefoil height increased with applied Mo by an average of 10% each week after the fourth measurement period. Flatpea height was also enhanced with Mo fertilization by 12% during weeks 2, 4, and 5. Plant height also increased with Mo fertilization within the 0 kg P ha^{-1} treatment. This supported the evidence that the initial available soil P level was sufficient since legumes generally do not respond to applied Mo if soil P is deficient (Anderson, 1956b).

Dry Matter Yield. Molybdenum fertilization increased red clover yield by 56% above the non-fertilized Mo treatment. Birdsfoot trefoil and flatpea yields were also increased by 26% with applied Mo (Table 24; Figures 16, 17, and 18). These results were in accordance with Hagstrom and Berger (1963) who observed a 65% increase in red clover yields with 874 g Mo ha^{-1}.

Plant Tissue Analysis. Red clover P concentration was increased from 2637 mg kg^{-1} to 2756 mg kg^{-1} when Mo was applied at 874 g Mo ha^{-1} (Tables 25 and 26). Fertilizer Mo can improve root systems and plant vigor thereby causing a higher uptake of P (Shulka and Pathak, 1973).

Total plant Mo was increased approximately 30 times (0.11 to 3.36 mg kg^{-1}) in red clover when Mo fertilization was employed. Birdsfoot trefoil increased in total Mo from 0.11 to 2.20 mg kg^{-1} when fertilizer Mo was increased from 0 to 874 g ha^{-1}. Flatpea total Mo was also increased from 0.17 to 1.59 mg kg^{-1} with applied Mo (Tables 25 and 26). Without applied Mo these tissue Mo concentrations are considered deficient (Gupta, 1970). The highest plant Mo concentration reached 4.24 mg kg^{-1} with an 874 g Mo ha^{-1} application. This should not produce toxic concentrations of Mo for livestock (Gupta and Lipsett, 1981).

Total plant N increased in birdsfoot trefoil from 28.3 to 32.1 g kg^{-1} when Mo was applied to the soil, while flatpea total N concentration was increased from 39.5 to 41.3 g kg^{-1} with fertilizer

Mo (Tables 25 and 26). Utilization of Mo within the nitrogenase enzyme thus enhanced the fixation of N by the legumes.

Available Soil Molybdenum. Resin extractable Mo was equivalent to 16 µg kg^{-1} from soils not receiving Mo fertilization. The availability of this element was therefore low in comparison to results obtained by Karimian and Cox (1978). Low concentrations of plant tissue Mo were also observed without Mo fertilization (Tables 20 and 26). When Mo was applied to the surface soil at 874 g ha^{-1}, available Mo increased from 16.0 to approximately 86 µg kg^{-1} (Tables 27 and 28). This level of extractable Mo was not high enough to cause animal toxic Mo concentrations in the legumes.

SUMMARY AND CONCLUSIONS

Northeastern United States soils utilized for forage production are often deficient in available P, S, and Mo. Establishing legumes in such soils can be difficult. The effects of P, S, and Mo fertilization on legume seedling vigor were studied in order to determine fertilizer needs for more efficient establishment of legumes in low fertility soils. Seedling vigor was measured by seedling height, trifoliate leaf count, dry weight, and trifoliate leaf area during the first ten weeks of growth. Greenhouse experiments were conducted with red clover, birdsfoot trefoil, and white clover, while red clover, birdsfoot trefoil, and flatpea were studied in the field. Plant nutrient concentrations of P, S, Mo, and N were measured to confirm fertilization effects on the seedling legumes.

Legume seedling vigor in the greenhouse as measured by seedling height, trifoliate leaf count, dry weight, and trifoliate leaf area increased with P fertilization on a low fertility (4 mg kg^{-1} Bray-1 extractable P) Gilpin silt loam. The initial 22 mg P kg^{-1} treatment produced the largest increase in seedling vigor in most instances. Further growth increases were observed with higher P rates but increases were generally not of the magnitude observed with the initial application. The growth parameter measurements were very low in the control pots, showing that without P fertilization other establishment practices would be of little benefit. Phosphorus fertilization increased legume height and trifoliate leaf count as early as the first or second week in the measurement period. Hence, adequate P availability is a prerequisite for legume establishment on this naturally infertile soil.

Resin-extractable Mo (available Mo) was 12 $\mu g\ kg^{-1}$ in the Gilpin soil utilized in the greenhouse. Soil pH averaged 4.9 and thus available Mo could probably be increased with liming. However, Mo fertilization at this low pH level increased the seedling vigor of red clover, white clover, and birdsfoot trefoil during the first eight weeks of growth, once adequate P was applied to the soil. The highest rate of Mo application (201 μg Mo kg^{-1}) generally resulted in the largest growth increase within any given P application.

Field experiments were conducted during 1983 and 1984 on a Gilpin silt loam with an initial Bray-1 extractble P level of 12.5 $\mu g\ kg^{-1}$. The 50 kg P ha^{-1} treatment generally produced the largest increases in seedling vigor as measured by dry weight and height. Height increases did not occur as quickly in the field as in the greenhouse, and thus at the end of the eight week measurement period absolute increases were less than those recorded in the greenhouse. This is reasonable because field growing conditions were not as optimal as those in the controlled environment of the greenhouse. Also, the field soil before fertilization contained a Bray-1 extractable P level approximately equal to the level obtained after the initial 22 mg P kg^{-1} addition in the greenhouse. Hence, lower responses to P applications would be expected, and were observed in these experiments.

Sulfur fertilization on the field experiments produced only one measurable increase in legume seedling vigor. Red clover height and dry matter yields increased with a 60 kg S ha^{-1} treatment during the 1984 experiment. Height response was first observed in the sixth week of the measurement period. Limited rainfall during the early part of the measurement period during both years may have reduced legume response to S.

The moderately acid (pH 5.2) Gilpin silt loam in the field had a resin-extractable Mo level of 16 µg kg^{-1}. Applications of 874 g Mo ha^{-1} generally increased legume seedling height and dry matter yields. Total N content in tissue was also enhanced by Mo fertilization and, thus, Mo probably increased the N_2-fixation capacity of the legumes. Therefore, the initial resin-extractable Mo level can be considered to be low for legume growth under these experimental conditions.

Phosphorus fertilization increased legume tissue concentration and uptake of P. Legume seedling vigor in the greenhouse was poor when plant tissue concentrations were below 1300 mg P kg^{-1}. However, after the initial application of 22 mg P kg^{-1}, plant P levels increased to between 1500 and 1800 mg kg^{-1}. This increase was normally associated with the greatest increase in seedling vigor. Legumes established in the field without applied P contained tissue levels of P greater than 1800 mg kg^{-1}. Most of the growth parameters in the field increased the most using 50 kg P ha^{-1}, and the plant P tissue levels associated with this rate were approximately 2500 mg kg^{-1}. Phosphorus fertilization enhanced plant Mo concentrations in the greenhouse by a possible formation of a readily available phosphomolybdate anion. Added P also increased total plant N in the legumes.

Sulfur fertilization increased legume total S levels. However, without S fertilization, red clover tissue S level was approximately 1900 mg kg^{-1}, and this was near the sufficiency level reported by Martin and Matocha (1973). Legume N:S ratios usually decrease with S fertilization if plant availabel S is initially low, but this was not observed in these experiments. Therefore, the results imply that plant available S for legume establisment was not originally low on this soil.

Molybdenum fertilization increased legume Mo concentrations in both the greenhouse and field experiments, and did not result in plant Mo tissue levels high enough to cause molybdenosis in cattle. Applied Mo increased the uptake and concentration of P within the legumes, probably as a result of a more extensive root system being developed by plants receiving Mo fertilization. Molybdenum fertilization also enhanced total plant N, which was to be expected if available soil Mo levels were initially low.

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APPENDIX

Appendix Table 1. Red clover height and trifoliate leaf count as influenced by fertilizer P, and Mo during the first eight weeks of growth in the greenhouse.

Treatment		Height								Trifoliate Count							
		Week								Week							
P	Mo	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8
mg kg^{-1}	ug kg^{-1}	mm								count							
0	0	20	23	25	24	23	24	32	44	0.6	1.0	1.1	1.7	2.1	2.7	3.1	3.9
	22	21	22	24	22	22	23	37	47	0.7	1.0	1.1	1.7	2.2	3.0	2.2	3.9
	67	23	24	26	23	25	26	35	48	0.7	1.0	1.1	1.7	2.1	2.0	3.5	4.3
	201	23	25	25	25	25	24	36	50	0.6	1.0	1.0	1.9	2.3	2.9	3.7	4.2
22	0	25	35	44	52	67	84	100	117	0.7	1.2	1.9	3.0	4.5	6.0	7.8	9.5
	22	26	36	45	54	70	91	111	130	0.8	1.2	2.0	2.9	4.8	7.0	9.0	10.4
	67	24	36	47	56	79	107	128	143	0.7	1.2	2.0	3.1	5.3	8.2	10.6	13.7
	201	26	36	47	55	77	105	127	142	0.7	1.2	2.0	3.1	5.0	8.1	10.6	15.4
67	0	30	53	61	69	75	88	109	129	1.0	2.0	2.9	3.7	5.6	7.2	9.0	11.4
	22	27	51	62	70	80	101	121	138	0.8	1.9	2.7	3.8	6.0	8.8	11.9	14.8
	67	29	53	65	75	93	112	127	140	0.9	1.9	2.7	3.7	6.5	9.9	12.7	17.1
	201	28	52	64	76	96	117	142	167	1.0	2.0	2.9	3.9	6.9	11.5	14.9	20.8
201	0	24	48	60	71	78	97	117	143	0.9	2.0	2.9	3.8	5.9	9.1	11.5	15.4
	22	30	56	69	79	88	109	130	153	0.9	1.9	2.8	4.1	6.6	9.6	14.0	18.0
	67	28	55	70	82	97	120	142	168	0.9	2.0	2.9	4.1	7.1	10.6	14.6	21.7
	201	30	58	76	84	106	126	146	172	1.0	2.0	3.0	4.1	8.0	12.2	17.7	24.6

a.

b.

c.

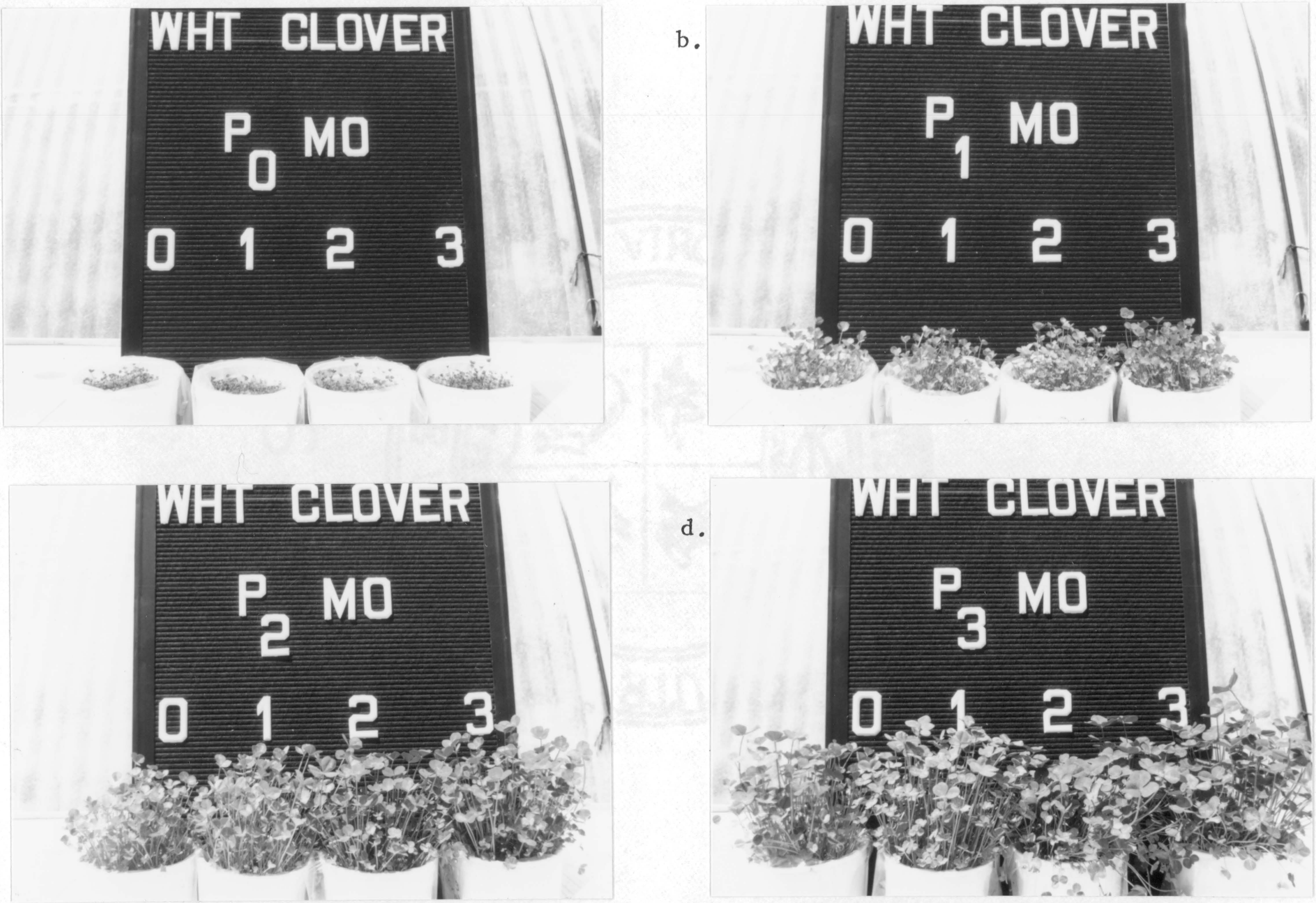
d.

Appendix Plate 1. Red clover height after eight weeks of growth in the greenhouse as influenced by (a) 0 mg P kg^{-1} and applied Mo (0, 22, 67, and 201 µg Mo kg^{-1}), (b) 22 mg P kg^{-1} and applied Mo, (c) 67 mg P kg^{-1} and applied Mo, and (d) 201 mg P kg^{-1} and applied Mo.

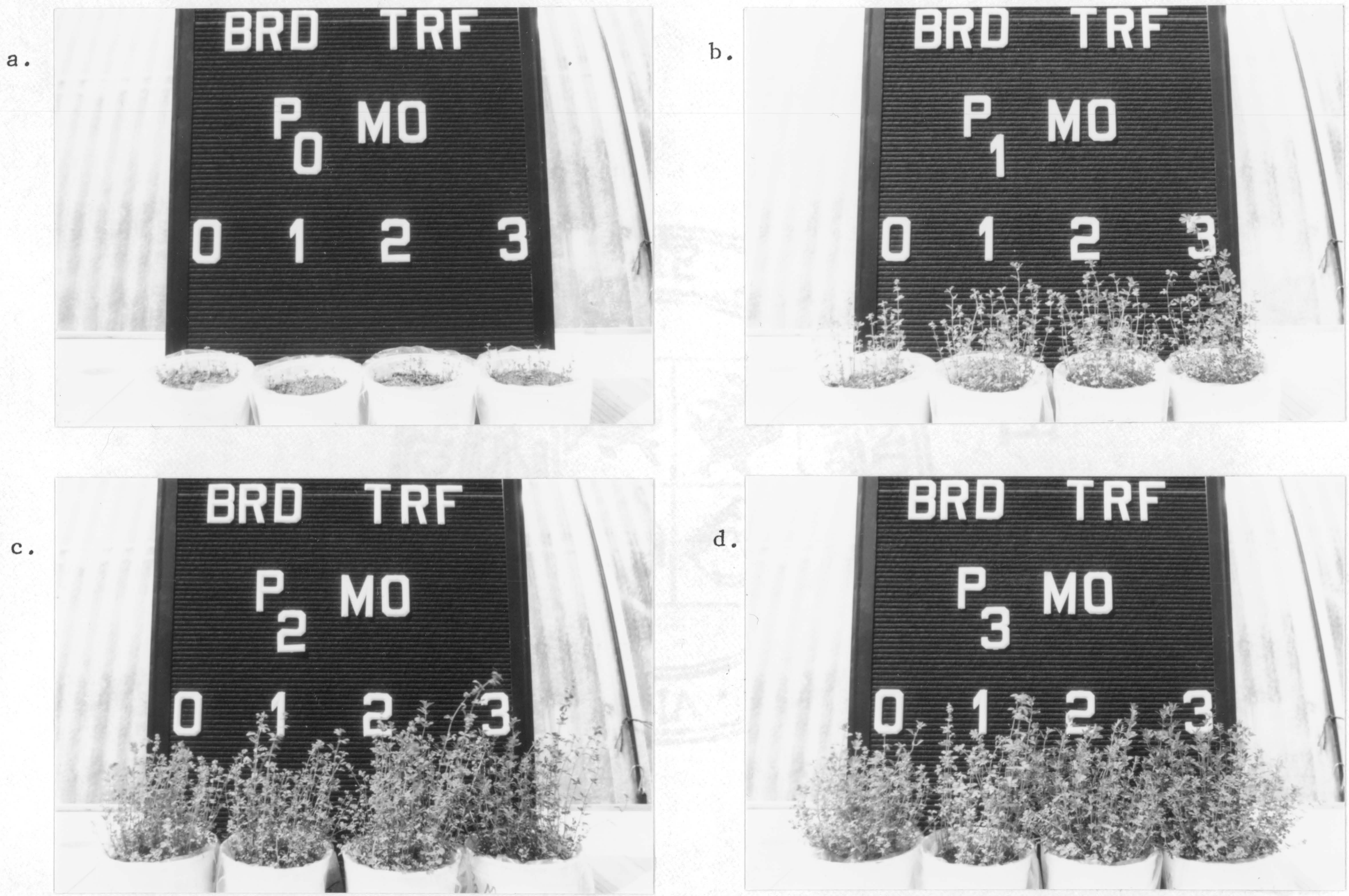
Appendix Table 2.. White clover and birdsfoot trefoil height as influenced by fertilizer P, and Mo during the first eight weeks of growth in the greenhouse.

Treatment		White Clover Height Week				Birdsfoot Trefoil Height Week			
P	Mo	2	4	6	8	2	4	6	8
mg kg^{-1}	μg kg^{-1}	---------mm---------				---------mm---------			
0	0	18	20	19	23	21	25	27	27
	22	18	18	17	22	22	26	28	34
	67	18	21	21	27	24	30	33	35
	201	17	19	19	23	25	28	33	39
22	0	21	25	53	82	27	40	74	114
	22	20	26	55	80	26	39	71	110
	67	21	22	44	76	31	42	76	135
	201	20	25	58	85	25	37	67	125
67	0	28	40	82	112	35	48	90	130
	22	25	40	86	133	31	46	95	157
	67	26	42	93	113	35	51	102	188
	201	28	45	90	149	33	50	102	181
201	0	36	56	92	133	31	48	90	145
	22	35	58	93	156	35	55	107	168
	67	35	60	95	157	34	53	109	187
	201	33	53	107	151	34	56	113	181

a. b. c. d.



Appendix Plate 2. White clover height after eight weeks of growth in the greenhouse as influenced by (a) 0 mg P kg^{-1} and applied Mo (0, 22, 67, and 201 µg Mo kg^{-1}), (b) 22 mg P kg^{-1} and applied Mo, (c) 67 mg P kg^{-1} and applied Mo, and (d) 201 mg P kg^{-1} and applied Mo.



Appendix Plate 3. Birdsfoot trefoil height after eight weeks of growth in the greenhouse as influenced by (a) 0 mg P kg^{-1} and applied Mo (0, 22, 67, and 201 μg Mo kg^{-1}), (b) 22 mg P kg^{-1} and applied Mo, (c) 67 mg P kg^{-1} and applied Mo, and (d) 201 mg P kg^{-1} and applied Mo.

Appendix Table 3. Red clover dry weight and trifoliate leaf area means as affected by P and Mo fertilization after eight weeks of growth in the greenhouse.

Treatment		Dry wt.	Trifoliate Area
P	Mo		
mg kg^{-1}	µg kg^{-1}	gms pot^{-1}	cm^2
0	0	0.14	1.48
	22	0.14	1.60
	67	0.15	1.72
	201	0.15	1.70
22	0	1.08	8.19
	22	1.55	9.65
	67	2.18	11.40
	201	3.04	12.85
67	0	1.39	8.72
	22	2.48	10.14
	67	3.24	10.80
	201	5.65	13.37
201	0	2.47	11.65
	22	3.59	13.28
	67	5.24	14.08
	201	7.00	14.95

Appendix Table 4. White clover dry weight and trifoliate leaf area, and birdsfoot trefoil dry weight as affected by P and Mo fertilization after eight weeks of growth in the greenhouse.

Treatment		White clover		Birdsfoot Trefoil
P	Mo	Dry Wt.	Trifoliate	Dry Wt.
mg kg^{-1}	µg kg^{-1}	gms pot^{-1}	cm^2	gms pot^{-1}
0	0	0.23	0.37	0.06
	22	0.19	0.34	0.10
	67	0.22	0.41	0.11
	201	0.24	0.37	0.11
22	0	1.05	1.78	0.68
	22	0.88	2.13	0.75
	67	1.02	1.73	0.80
	201	1.11	2.08	0.68
67	0	1.88	3.00	1.34
	22	2.24	4.18	1.62
	67	2.42	4.53	2.08
	201	3.58	5.00	2.18
201	0	3.75	4.38	1.51
	22	4.73	5.74	2.90
	67	4.69	6.65	2.95
	201	5.74	7.39	2.78

Appendix Table 5. Red clover tissue concentrations of Ca, Mg, and K as affected by P and Mo fertilization after eight weeks of growth in the greenhouse.

Treatment				
P	Mo	Ca	Mg	K
mg kg^{-1}	µg kg^{-1}	mg kg^{-1}		
0	0	7000	3200	10,000
	22	7500	3000	10,000
	67	8500	3000	10,000
	201	6000	3000	10,000
22	0	7400	3400	18,000
	22	9200	3400	18,000
	67	7200	3000	17,000
	201	8100	2400	14,000
67	0	7600	3300	21,000
	22	8900	3000	18,000
	67	7500	2800	17,000
	201	7200	2200	14,000
201	0	9600	2500	21,000
	22	9500	2100	19,000
	67	8600	2000	18,000
	201	9500	1700	16,000

Appendix Table 6. Soil pH, and extractable P, Mo, Ca, Mg, and K as affected by P and Mo fertilization of the soils utilized in the red clover, white clover, and birdsfoot trefoil greenhouse experiments.†

Treatment		Red Clover					White Clover	Birdsfoot Trefoil
P	Mo	P	Mo	Ca‡	Mg‡	K‡	P	P
mg kg^{-1}	μg kg^{-1}	mg kg^{-1}	μg kg^{-1}	----cmol(+)kg^{-1}----			mg kg^{-1}	mg kg^{-1}
0	0	4	10	3.3	0.33	0.52	4	5
	22	3	12	2.9	0.33	0.49	3	4
	67	3	20	2.9	0.33	0.49	3	4
	201	4	29	2.9	0.33	0.50	3	4
22	0	11	13	3.2	0.29	0.46	11	14
	22	13	14	2.8	0.29	0.43	12	14
	67	11	19	2.8	0.29	0.39	11	13
	201	13	30	2.8	0.29	0.30	11	14
67	0	44	10	3.3	0.29	0.45	36	41
	22	42	19	3.1	0.29	0.44	36	44
	67	38	28	2.9	0.25	0.40	36	41
	201	36	23	2.6	0.17	0.25	36	42
201	0	167	14	4.0	0.29	0.41	161	162
	22	159	16	3.4	0.25	0.41	153	159
	67	156	18	3.3	0.21	0.29	154	159
	201	160	32	3.2	0.21	0.33	148	154

† Soil pH was 4.9 for all treatments.
‡ Values recorded from 16 randomly choosen samples which represented each treatment.

Appendix Table 7. Statistical significance levels, based on F tests, for the effect of Mo fertilization, within each P treatment, on red clover height and trifoliate leaf count during the first eight weeks of growth in the greenhouse.

Source	df†	Height P (mg kg^{-1})				Trifoliate Count P (mg kg^{-1})			
		0	22	67	201	0	22	67	201
Wk. 1 Mo	3	NS‡	NS	NS	**	NS‡	NS	NS	NS
0 Mo vs. 1,2,3 Mo§	1	NS	NS	NS	**	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Wk. 3 Mo	3	NS	NS	NS	**	NS	NS	NS	NS
0 Mo vs. 1,2,3 Mo	1	NS	NS	NS	**	NS	NS	NS	NS
Linear 1,2,3 Mo	1	NS	NS	NS	*	NS	NS	NS	*
Quadratic 1,2,3 Mo	1	NS	NS	NS	NS	NS	NS	NS	NS
Wk. 5 Mo	3	NS	*	**	**	NS	NS	*	*
0 Mo vs. 1,2,3 Mo	1	NS	*	**	**	NS	*	*	*
Linear 1,2,3 Mo	1	NS	NS	**	**	NS	NS	*	*
Quadratic 1,2,3 Mo	1	NS	NS	**	NS	NS	NS	NS	NS
Wk 7 Mo	3	NS	**	**	**	*	**	**	**
0 Mo vs. 1,2,3 Mo	1	NS	**	**	**	*	**	**	**
Linear 1,2,3 Mo	1	NS	**	**	**	NS	NS	**	**
Quadratic 1,2,3 Mo	1	NS	**	NS	*	NS	NS	NS	NS

*, ** Significant at the 0.05 and 0.01 levels, respectively.
† df = Degrees of freedom.
‡ NS = Not significant.
§ 0,1,2,3 Mo represents 0, 22, 67, and 201 ug Mo kg^{-1}, respectively.

Appendix Table 8. Red clover and birdsfoot trefoil height as influenced by P, S, and Mo fertilization during the first eight weeks of growth in the 1983 field experiment.

Treatment			Red clover								Birdsfoot Trefoil							
			Week								Week							
P	S	Mo	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8
kg ha^{-1}	kg ha^{-1}	g ha^{-1}	mm								mm							
0	0	0	27	29	84	87	114	132	142	146	16	32	65	92	129	155	161	172
	60	0	28	51	76	89	105	112	135	145	13	30	70	90	117	148	179	176
	0	874	33	55	83	89	106	113	134	148	13	30	72	91	142	160	194	213
	60	874	22	43	77	86	100	121	141	156	14	28	74	98	144	166	182	208
50	0	0	29	52	74	96	104	130	149	155	17	34	68	93	124	139	162	183
	60	0	29	52	83	83	101	120	137	146	12	25	53	85	104	142	174	184
	0	874	28	52	82	94	116	135	144	169	14	29	65	93	132	162	193	223
	60	874	26	48	82	94	106	126	137	156	14	29	75	99	138	169	217	222
150	0	0	23	48	76	105	101	131	142	162	16	29	68	100	144	178	207	212
	60	0	29	52	78	109	125	147	157	175	15	28	66	92	131	165	189	204
	0	874	29	56	89	107	131	144	158	170	16	34	72	93	124	161	191	203
	60	874	26	48	90	106	103	137	154	177	17	32	74	97	150	171	200	226
450	0	0	30	54	88	100	117	139	154	156	14	30	71	99	134	174	208	237
	60	0	34	53	92	94	109	120	134	136	14	28	65	83	127	154	206	224
	0	874	37	66	86	105	115	139	151	167	14	29	65	96	133	161	208	231
	60	874	27	50	84	99	107	127	132	167	16	34	78	125	170	194	248	282

Appendix Table 9. Daily precipitation from April to September during the 1983 field experiment.

Day	Apr.	May	June	July	Aug.	Sept.
	Month					
	cm					
1	.†	.	0.08	.	0.05	.
2	0.63	.	.	.	.	.
3	*‡	1.17	1.98	0.13	.	.
4	0.18	0.08	0.18	1.54	.	.
5	0.30	.	.	0.05	2.54	.
6	.	.	.	.	.	.
7	0.13	.	.	.	.	.
8	.	0.76	.	.	.	.
9	2.19	.	.	.	.	.
10	0.58	.	.	.	.	.
11	.	.	.	.	4.01	.
12	.	.	.	.	.	.
13	.	.	.	.	.	.
14	0.05	0.05	0.23	.	.	.
15	2.03	0.71	.	.	.	.
16	*	2.06	0.43	.	.	.
17	*	.	0.46	0.86	.	.
18	*	.	0.05	0.05	0.10	.
19	*	1.50	0.18	0.33	.	.
20	*	.	0.68	2.18	.	0.05
21	0.10	1.50	.	.	0.05	1.19
22	.	0.43	0.05	0.63	0.10	.
23	3.53	0.02	.	4.72	0.13	.
24	1.98	.	.	0.23	0.05	.
25	.	.	.	0.02	.	.
26	.	.	.	.	.	.
27	.	.	.	.	1.19	.
28	.	0.41	1.12	.	3.40	.
29	.	1.60	0.08	.	0.02	.
30	.	0.02	.	.	.	2.46
31	M§	.	M	.	.	M
TOTAL	12.45	10.31	5.51	10.67	11.66	3.70

† No rainfall.
‡ Precipitation included in next day.
§ Missing data.

Appendix Table 10. Red clover, birdsfoot trefoil, and flatpea dry weight yield as influenced by P, S, and Mo fertilization in the 1983 field experiment.

Treatment			Red Clover	Birdsfoot trefoil	Flatpea
P	S	Mo		yields	
kg ha^{-1}	kg ha^{-1}	g ha^{-1}		kg ha^{-1}	
0	0	0	545	898	43
	60	0	572	733	39
	0	874	514	980	43
	60	874	594	1290	55
50	0	0	676	1059	46
	60	0	584	955	24
	0	874	894	1337	39
	60	874	900	1950	42
150	0	0	1015	959	60
	60	0	842	913	38
	0	874	941	1081	73
	60	874	829	1182	48
450	0	0	822	1979	48
	60	0	752	1537	64
	0	874	1034	1432	66
	60	874	818	3024	60

Appendix Table 11. Red clover, birdsfoot trefoil, and flatpea N:S ratios as influenced by P, S, and Mo fertilization in the 1983 field experiment.

Treatment			Red clover	Birdsfoot trefoil	Flatpea
P	S	Mo	N:S	N:S	N:S
kg ha^{-1}	kg ha^{-1}	g ha^{-1}			
0	0	0	14.6	11.2	15.7
	60	0	14.5	9.7	16.2
	0	874	16.2	13.9	18.6
	60	874	15.4	11.7	18.0
50	0	0	16.1	12.1	17.7
	60	0	14.0	8.9	13.5
	0	874	16.3	15.6	19.7
	60	874	15.2	14.0	19.1
150	0	0	15.1	12.0	15.4
	60	0	14.6	12.5	18.1
	0	874	16.6	16.9	18.6
	60	874	15.6	13.7	19.6
450	0	0	14.8	12.4	17.6
	60	0	12.2	9.9	17.7
	0	874	14.7	14.1	19.2
	60	874	14.2	13.1	15.9

Appendix Table 12. Red clover, birdsfoot trefoil, and flatpea tissue concentrations of Ca, Mg, and K as affected by P, S, and Mo fertilization in the 1983 field experiment.

Treatment			Red clover			Birdsfoot Trefoil			Flatpea		
P	S	Mo	Ca	Mg	K	Ca	Mg	K	Ca	Mg	K
kg ha^{-1}	kg ha^{-1}	g ha^{-1}	mg kg^{-1}			mg kg^{-1}			mg kg^{-1}		
0	0	0	10375	1725	19750	9500	1525	24500	8875	1600	17875
	60	0	12060	1825	20750	8500	1775	25125	9750	1850	18125
	0	874	11500	1725	24375	9500	1425	25625	8125	1450	21250
	60	874	11750	1650	18125	10625	1575	24375	9125	1625	16500
50	0	0	12125	1950	20500	8750	1775	24500	8875	1525	18500
	60	0	11000	1700	22375	9625	1925	25500	8750	1550	19500
	0	874	10625	1875	19625	9750	1525	24250	8375	1475	21250
	60	874	10875	1850	24750	13125	1475	20750	8125	1400	22250
150	0	0	11000	1750	28875	9000	1700	20750	9375	1625	19250
	60	0	12125	1800	24125	9750	1700	20750	9500	1600	19125
	0	874	11875	1825	21500	8250	1525	27125	9625	1650	19000
	60	874	12375	1725	24875	10250	1625	17250	9250	1525	21375
450	0	0	11500	2075	18125	11375	1925	24250	8750	1825	21125
	60	0	10875	2100	23250	10375	1775	25000	9750	1750	21100
	0	874	10750	2050	18500	9500	1650	22250	9750	1825	22000
	60	874	10875	2200	23750	10875	1800	21125	8875	1900	17500

Appendix Table 13. Red clover, birdsfoot trefoil, and flatpea height as influenced by P, S, and Mo fertilization during the first eight weeks of growth in the 1984 field experiment.

Treatment			Red clover								Birdsfoot trefoil								Flatpea							
			Week								Week								Week							
P	S	Mo	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8	1	2	3	4	5	6	7	8
kg ha^{-1}	kg ha^{-1}	g ha^{-1}					mm								mm								mm			
0	0	0	50	66	94	121	148	169	190	204	47	68	97	115	146	157	182	189	48	60	68	75	109	121	151	162
	60	0	47	66	100	137	168	211	242	252	52	73	109	137	171	187	212	228	51	64	74	84	101	114	126	140
	0	874	46	66	91	117	143	162	195	199	49	66	98	151	179	195	247	261	59	73	84	95	129	142	162	184
	60	874	45	58	91	123	161	192	214	245	58	79	111	143	166	174	208	221	59	68	80	109	136	142	187	205
50	0	0	46	65	105	131	162	191	226	260	58	81	124	151	183	210	242	259	57	68	68	76	99	104	132	157
	60	0	51	71	109	151	193	224	268	293	56	80	132	161	188	217	241	258	50	57	65	82	99	118	137	148
	0	874	60	78	116	141	178	229	271	282	58	78	125	157	189	225	269	286	52	57	67	86	104	124	151	175
	60	874	52	76	116	157	207	247	303	316	67	88	134	172	218	242	279	292	50	69	85	101	120	133	177	192
150	0	0	41	58	95	125	165	199	241	260	51	70	109	141	167	192	222	250	49	57	66	82	109	122	150	193
	60	0	53	69	108	139	185	220	261	305	58	79	115	151	184	218	254	275	47	62	77	88	117	159	178	217
	0	874	59	79	122	157	204	251	307	326	53	72	111	142	189	218	262	272	55	65	79	111	151	171	194	235
	60	874	61	81	129	169	227	261	312	352	64	89	136	164	196	224	262	287	54	64	74	93	119	144	152	193
450	0	0	45	63	97	125	169	207	212	247	52	73	107	149	189	195	237	245	50	59	77	87	132	163	181	236
	60	0	50	70	113	147	183	232	273	282	61	82	133	167	219	263	300	318	45	54	64	85	118	144	177	209
	0	874	50	68	115	149	199	235	282	302	76	101	152	184	248	268	291	311	47	59	71	91	124	139	180	212
	60	874	43	65	99	132	180	245	293	307	54	77	118	151	199	248	274	291	51	61	72	78	119	144	178	217

Appendix Table 14. Daily precipitation from April to September during the 1984 field experiment.

	Month					
Day	Apr.	May	June	July	Aug.	Sept.
	cm					
1	.†	.	.	.	0.43	.
2	.	0.66	.	0.05	0.08	.
3	.	0.99	.	0.08	.	2.08
4	2.01	1.27	.	0.96	0.18	0.41
5	0.94	.	.	2.54	.	.
6	0.05	3.76	.	1.04	.	.
7	.	2.97	.	0.61	.	.
8	.	0.56	.	.	0.20	.
9	.	.	.	.	.	.
10	.	.	.	.	0.63	.
11	.	.	.	0.46	*‡	.
12	.	0.20	.	0.05	*	.
13	.	0.02	.	.	*	.
14	0.94	.	.	.	3.35	0.81
15	0.08	.	.	0.61	.	0.15
16	0.20	.	0.66	3.28	.	.
17	0.20	.	0.84	1.12	.	.
18	0.18	.	.	0.68	3.12	.
19	0.13	.	.	.	.	.
20	.	.	.	.	.	.
21	1.37	.	.	.	.	.
22	1.70	0.05	.	.	.	.
23	0.15	0.58	0.99	.	0.56	.
24	0.13	.	2.34	.	.	.
25	.	.	.	0.41	.	.
26	.	.	.	0.91	.	.
27	.	0.08	.	0.43	.	*
28	0.43	1.85	0.41	.	0.46	1.17
29	.	0.23	0.41	0.08	.	.
30	0.20	0.05	0.91	.	1.09	0.30
31	M§	.	M	.	.	M
Total	8.71	13.28	6.55	13.31	10.11	4.93

† No rainfall

‡ Precipitation included in next day.

§ Missing data.

Appendix Table 15. Red clover, birdsfoot trefoil, and flatpea dry weight yield as influenced by P, S, and Mo fertilization in the 1984 field experiment.

Treatment			Red clover	Birdsfoot trefoil	Flatpea
P	S	Mo	--------------	yield	--------------
kg ha^{-1}	kg ha^{-1}	g ha^{-1}	--------------	kg ha^{-1}	--------------
0	0	0	685	561	41
	60	0	1365	1004	53
	0	874	803	1501	52
	60	874	1090	939	77
50	0	0	1371	1171	44
	60	0	1747	1331	41
	0	874	1522	1550	55
	60	874	3159	2006	71
150	0	0	1072	938	59
	60	0	1168	1498	76
	0	874	1891	1123	83
	60	874	3023	1786	84
450	0	0	760	1012	68
	60	0	1672	1985	45
	0	874	1953	1978	50
	60	874	1899	1088	71

Appendix Table 16. Red clover, birdsfoot trefoil, and flatpea N:S ratios as influenced by P, S, and Mo fertilization fertilization in the 1984 field experiment.

Treatment			Red clover	Birdsfoot trefoil	Flatpea
P	S	Mo	N:S	N:S	N:S
kg ha^{-1}	kg ha^{-1}	g ha^{-1}			
0	0	0	17.5	9.4	17.3
	60	0	15.6	10.1	16.3
	0	874	17.4	13.2	17.0
	60	874	16.6	10.4	18.7
50	0	0	17.1	12.6	18.0
	60	0	16.5	10.2	17.3
	0	874	18.0	14.9	18.1
	60	874	17.7	13.2	18.7
150	0	0	17.0	11.5	16.7
	60	0	17.4	11.4	18.9
	0	874	18.2	15.5	17.1
	60	874	16.9	13.8	16.5
450	0	0	17.1	11.5	16.6
	60	0	16.6	12.6	15.8
	0	874	17.3	13.8	17.3
	60	874	16.5	12.1	15.2

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