EFFECTS OF MOLYBDENUM AND INTERACTING ELEMENTS ON THE PERFORMANCE OF TROPICAL PASTURE/FORAGE LEGUMES

A DISSERTATION SUBMITTED TO THE GRADUATE DIVISION OF THE
UNIVERSITY OF HAWAII IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY

IN

AGRONOMY AND SOIL SCIENCE

DECEMBER 1984

By

Pichit Pongsakul

Dissertation Committee:

Yoshinori Kanehiro, Chairman
Mitiku Habte
Charles L. Murdoch
A. Sheldon Whitney
Russell S. Yost

We certify that we have read this dissertation and that in our opinion it is satisfactory in scope and quality as a dissertation for the degree of Doctor of Philosophy in Agronomy and Soil Science.

DISSERTATION COMMITTEE

Chairman

ABSTRACT

This investigation was to study the effects of Mo, S and lime on yield, biological nitrogen fixation and nutrient concentration of tropical pasture legumes; to determine the external and internal Mo requirements of tropical pasture legumes; and to evaluate the mycorrhiza-rhizobium-legume association as related to Mo status in acid tropical soils.

In order to fulfill these objectives, the study was conducted in three parts. In the first part, the experiments were conducted in the greenhouse and in the field. In the greenhouse experiment, _Desmodium intortum_ was grown in the Wahiawa (Tropeptic Eutrustox) and Paaloa (Humoxic Tropohumult) soils and _Centrosema pubescens_ was grown in the Wahiawa soil. Treatments were composed of five levels of Mo (none added, seed-applied Mo at 0.1 and 0.5 kg Mo/ha, and soil-applied Mo at 2.0 and 4.0 kg Mo/ha); two levels of S (none added and 50 kg S/ha); and two levels of lime (none added and lime rate so that a pH of soil is raised to 6.0). In the field experiment, the treatments were composed of two legume species (_D. intortum_ and _C. pubescens_) and ten fertilizer treatments (none added; lime; seed-applied Mo at 0.1, 0.3, 0.5 and 0.5 kg Mo/ha plus lime; soil-applied Mo at 1.0, 2.0, 4.0 and 4.0 kg Mo/ha plus lime).

In the second part, _D. intortum_ was grown in the Wahiawa and Paaloa soils in the greenhouse. Seven rates of Mo (0, 0.5, 1.0, 2.0, 4.0, 8.0, and 16.0 kg Mo/ha) were added to the soil. Molybdenum adsorption and desorption curves were also studied.

In the third part, treatments were composed of three legume species (_D. intortum_, _C. pubescens_ and _Stylosanthes humilis_); three Mo levels (none added, seed-applied Mo at 0.3 kg Mo/ha and soil-applied Mo at 2.0 kg Mo/ha); and two mycorrhizal inoculation rates (non-inoculation and inoculation). Mycorrhiza used was Glomus mosseae. Soil was fumigated with methyl bromide one month before planting.

Molybdenum increased dry matter yield of desmodium and centrosema grown in the Wahiawa and Paaloa soils. Soil-applied Mo at 2.0 kg Mo/ha was adequate for legumes grown for one cutting in the greenhouse. However, soil-applied Mo at 4.0 kg Mo/ha should be used for excellent stands of legumes harvested five times a year. Seed-applied Mo at 0.5 kg Mo/ha was as effective as soil-applied Mo at 2.0 and 4.0 kg Mo/ha. Seed-applied Mo at 0.1 kg Mo/ha was less effective than seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 2.0 and 4.0 kg Mo/ha.

Molybdenum did not affect nodule fresh and dry weights of desmodium grown in the Wahiawa soil. However, seed-applied Mo at 0.5 kg Mo/ha decreased nodule fresh and dry weights of desmodium grown in the Paaloa soil. Molybdenum did not affect nodule number and nodule fresh weight of centrosema grown in Wahiawa soil. However, soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased nodule dry weight of the legume.

Nitrogenase activity of desmodium grown in the Wahiawa soil tended to increase when Mo was applied. Seed-applied Mo at 0.5 kg Mo/ha, soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased nitrogenase activity by 29.1%, 37.0% and 31.8%, respectively, above the no-Mo treatment. Molybdenum also increased nitrogenase activity of centrosema grown in the Wahiawa soil.

Molybdenum increased N concentration in the tops of desmodium and centrosema grown in Wahiawa soil. Seed-applied Mo at 0.3 and 0.5 kg Mo/ha and soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha increased N concentration from the first to fifth cutting. At the fifth cutting, plant N concentration of legumes receiving seed-applied Mo at 0.3 kg Mo/ha was lower than those of legumes receiving seed-applied Mo at 0.5 kg Mo/ha and the three soil-applied Mo rates. Seed-applied Mo at 0.1 kg Mo/ha did not increase plant N concentration. The residual effect of seed-applied Mo at 0.5 kg Mo/ha appeared to be less than that of soil-applied Mo at 4.0 kg Mo/ha.

Molybdenum application increased Mo concentration in the tops of legumes grown in the Wahiawa and Paaloa soils. Plant Mo concentrations in the later cuttings decreased when compared to those in the earlier cuttings. Critical Mo concentrations in plant top associated with 80% and 95% maximum yield of desmodium grown in the Wahiawa soil were 0.13-0.20 and 0.22-0.30 ppm, respectively. Comparable Mo concentrations of desmodium with the Paaloa soil were 0.40 and 1.30 ppm, respectively. Critical Mo concentration in the top associated with 80% maximum yield of centrosema grown in the Wahiawa soil was 0.20 ppm.

Molybdenum application also increased Mo concentration in the nodule of legumes. Critical Mo concentrations in nodule associated with 80% and 95% maximum yield of desmodium grown in the Wahiawa soil were 2.2-3.5 and 7.85 ppm, respectively. Comparable Mo concentrations of desmodium with the Paaloa soil were 4.0 and 13.2 ppm, respectively. Critical Mo concentration associated with 80% maximum yield of centrosema grown in the Wahiawa soil was 4.2 ppm.

Molybdenum application increased available Mo in soil. The available Mo associated with 95% maximum yield of desmodium grown in the Wahiawa and Paaloa soils were 0.09 and 0.10 ug Mo/ml, respectively. Predicted quantity of applied Mo required to establish 0.09 ug Mo/ml available Mo for the Wahiawa soil was 4.4 kg Mo/ha. For the Paaloa soil, predicted quantity of applied Mo to established 0.1 ug Mo/ml was 4.7 kg Mo/ha.

Molybdenum had no effect on S concentration in the tops of desmodium and centrosema but tended to increase Cu concentrations. The potential harmful effect of Mo regarding Mo-induced Cu deficiency in animals should not occur since Cu/Mo ratios in plant tops were well above 3, which is the ratio under which Mo-induced Cu deficiency usually occurs.

Sulfur did not increase dry matter yield, nodule number, nodule fresh and dry weights, nitrogenase activity, plant N concentration and other nutrient concentrations of desmodium and centrosema grown in the Wahiawa soil. Sulfur increased yield of desmodium grown in the Paaloa soil. Nodule fresh and dry weights of desmodium grown in limed Paaloa soil and receiving seed-applied Mo with S was higher than that of desmodium grown in the same treatment without S.

Lime increased dry matter yields of desmodium and centrosema grown in the Wahiawa soil. Lime increased plant N, Mo but decreased Cu concentrations of desmodium grown in the Paaloa soil. However, lime increased Cu concentrations in the tops of desmodium and centrosema grown in the Wahiawa soil. Lime increased nodule number, nodule fresh and dry weights, nitrogenase activity and plant N concentration of centrosema grown in the Wahiawa soil. Lime did not increase Mo and S concentrations in the tops of desmodium and centrosema grown in the Wahiawa soil.

Mycorrhizae enhanced plant growth and increased yields of desmodium, centrosema and stylosanthes. Mycorrhizae strongly stimulated nodulation of legumes. Nonmycorrhizal plants did not nodulate. Mycorrhizae did not affect Mo concentration in the tops of desmodium, centrosema and stylosanthes. Seed-applied Mo at 0.3 kg Mo/ha increased the percentage of mycorrhizal infection, suggesting that there is a relationship between Mo concentration and mycorrhizal infection.

TABLE OF CONTENTS

LIST OF TABLES

Table Page

LIST OF FIGURES

Figure Page

GENERAL INTRODUCTION

Pasture/forage production in the tropics is relatively low compared to the production in the temperate zone. Soils of the tropics are generally low in fertility, high acidity, and deficient in phosphorus and some micronutrients such as molybdenum. Increasing forage production on the highly weathered, acid soils of the tropics seem probable with proper fertilizers, soil amendments, and crop management practices.

The vast majority of the grazing lands in the tropics are acid. These acid soils are considered infertile because of low pH, excessive levels of aluminum and/or manganese, low base saturation and deficiencies of calcium, magnesium and molybdenum. These components may affect the performance of rhizobia, the nodulation, the nitrogen fixing process and the host plant. In many situations, it is difficult to determine what factor of soil acidity affecting the symbiotic nitrogen fixing process and the growth of the legume.

One of the important factors affecting pasture/forage yield is molybdenum. Molybdenum is involved in at least two essential plant functions. It is required in all plants for protein synthesis and for symbiotic nitrogen fixation in legumes. Molybdenum deficiency usually occurs in acid soil because its availability decreases as soil pH decreases, and it is adsorbed strongly by iron and aluminum oxides and hydrous oxides which are present in large amount in acid tropical soils.

The role of molybdenum, as well as other micronutrients, has not been evaluated in a systematic and definitive way in food production in the tropics. In a case such as molybdenum where limits between deficiency and toxicity for animals are narrow and where imbalance between micronutrients (e.g. molybdenum and copper) can easily occur due to over-application of one mocronutrient, more refined studies on rates and methods of molybdenum application and on interactions between molybdenum and other nutrients must be carried out. Pronounced interactions between molybdenum and sulfur on the growth of pasture/forage crops have been reported. It is recognized that one direct effect of sulfur addition to the soil is to decrease the molybdenum uptake by plants. Sulfur application, therefore, could be used to overcome molybdenum toxicity.

Lime can be used to decrease excessive aluminum and/or manganese and increase molybdenum availability. However, the use of massive doses of lime would be prohibitive, both economically and from an availability standpoint. Therefore, the main purpose of liming acid tropical soils often is to lower toxicity of aluminum and/or manganese and increase availability of molybdenum.

Molybdenum reaction in soil is similar to that of phosphorus because the available form of both nutrients is an anion and both nutrients are adsorbed by iron and aluminum oxides and hydrous oxides. For phosphorus, the value of the diffusion coefficient depends on the proportion of the labile phosphate which is in the soil solution. This principle should also be applied to molybdate.

There is well-documented evidence that vesicular-arbuscular mycorrhiza aids the host legume by increasing phosphorus uptake in infertile tropical soils. By the same basis of phosphorus uptake, molybdenum uptake by plant should also increase in the system of plant-mycorrhiza symbiosis. There is a need to find out how such association affects uptake of molybdenum as well as plant growth.

This research was conducted: (1) to study the effects of molybdenum sulfur and lime on growth, yields and biological nitrogen fixation of pasture/forage legumes grown in acid tropical soils; (2) to study the effects of lime in alleviating aluminum and/or manganese toxicities and in correcting molybdenum deficiency; (3) to study the effects of molybdenum, sulfur and lime interactions on growth, yield and some nutrient concentrations of pasture/forage legumes; (4) to determine the external molybdenum requirement of pasture/forage legumes grown in acid tropical soils; and (5) to evaluate the influence of mycorrhizae on yield and molybdenum response of pasture/forage legumes.

REVIEW OF LITERATURE

Molybdenum is one of the seven micronutrients essential for normal plant growth and development. This element is somewhat unique when compared to other micronutrients because its availability increases as soil pH increases. Molybdenum is required in small quantities for correcting deficiency in plants. It is involved in at least two essential plant functions: It is required in all plants for protein synthesis and for symbiotic nitrogen fixation in legumes.

Adsorption of Molybdenum in Soil

Molybdenum adsorption in soils has been used as an index of soil potential for decreasing the availability of molybdenum to plants. The molybdenum status of a soil depends not only on the amount of molybdate present in the adsorbed and solution phases but also on the distribution between them.

In considering the molybdenum status of soils it is convenient to subdivide molybdate in soil into three states: (1) molybdate in soil solution, (2) adsorbed molybdate, and (3) molybdate held in the crystal lattice of minerals (firmly-held molybdate). Adsorbed molybdate may be defined as molybdate on the soil solid phase which is in equilibrium with molybdate in soil solution. However, this definition is somewhat ambiguous because equilibrium between solid and solution is not instantaneous. It is also difficult to make a sharp separation between adsorbed molybdate and firmly-held molybdate. Firmly-held molybdate is not in direct equilibrium with the molybdate in solution, but a slow interchange can occur between the adsorbed and firmly-held states. Although these states are hard to define unambiguously, they are nevertheless useful concepts in considering molybdenum status in soil. In the short term, the availability of molybdate depends on the amount in the adsorbed and solution states and on the distribution between them. In the long term, status of availability depends on the rate of transfer between adsorbed molybdate and firmly-held molybdate.

Mechanism of Adsorption of Molybdate

One of the earliest observations on molybdenum status was that the molybdenum availability was increased by raising the pH and decreased by lowering it (Lewis, 1943). Subsequently, it was indicated that adsorption of molybdate by soils or oxides of iron and aluminum was decreased as the pH was raised above 4 (Jones, 1957; Reisenauer et al., 1962). Theng (1971) investigated the adsorption of molybdate by some soil clays containing dominantly allophane or layer lattice silicate minerals (kaolinite and illite) as a function of pH. He found that all soil samples, irrespective of their composition and charge characteristic, exhibit maximum adsorption close to pH 4. He further stated that adsorption could be satisfactorily explained in terms of the theory of the specific adsorption of anions developed by Hingston et al. (1967, 1968) for goethite systems. The theory postulates that the undissociated free acid such as molybdic acid and the most highly charged form of the anion are not adsorbed when present alone, i.e., at low and high pH with respect to pK. For adsorption to occur, the simultaneous presence in solution of proton-donating (e.g. $HMoO_4^-$) and proton-accepting (e.g. MoO_4^{2-}) species, as well as that of the clay surface, are required. Further, it was indicated that for monovalent anions there was an optimum pH for adsorption, and that this pH coincided with the pK of the dissociated acid. With polyvalent anions there were points of inflection in the curves of adsorption versus pH which coincided with the pK's of the acid. According to the theory of specific adsorption of anions (Hingston et al., 1967, 1968), the pH of maximum adsorption should coincide with pK 2. Maximum adsorption of molybdate occurs near pH 4 may be due to the fact that this is close to the pK 2 of molybdic acid (Barrow, 1977).

If the concentration is low enough so that polymeric forms can be ignored, molybdate may be present below pH 4 as H_2MoO_4 and MoO_4^{2-} (Theng, 1971). As the pH increases, the proportion of MoO_4^{2-} ions increases until, at the pK 2 of molybdic acid, the proportion of H_2MoO_4 and MoO_4^{2-} are equal. In an earlier paper by Hingston et al. (1967) it was argued that this proportion provided the best conditions for adsorption because it was necessary to have both proton donor and proton acceptor present. In a later paper (Hingston et al., 1972) it was argued that at this pH, the energy required to dissociate a proton from the acid was at a minimum. In either case, the mechanism envisaged is that the acid donates a proton to the clay surface and thus converts an OH^- group to a water molecule. This water molecule is displaced by the anion. The anion approaches the clay surface closely and forms a chemical bond with it. Specific adsorption can occur on surfaces which are negatively charged, and as a result of adsorption, the negative charge is increased (Barrow, 1977). An alternative view of the adsorption mechanism has been proposed by Bowden et al. (1977) who argue that the apparent maximum adsorption near the pK is due to a balance between positive and negative charges of clay surface: As pH rises the positive charge on the surface decreases but the proportion of negatively-charged ions increases. The rate of change of positive and negative charges on clay surface differ and the net effect is an adsorption maximum near the pK.

Characterization of Adsorption:

Adsorption isotherms for molybdate are always found to be curvilinear. Hence, at least two coefficients are needed to characterize adsorption. A common approach has been to use Langmuir isotherm to describe the curve (Reyes and Jurinak, 1967; Theng, 1971; Gonzelez et al., 1974). The linear form of the Langmuir equation may be written as $C/A = 1/kV + C/V$, where C and A refer to the equilibrium concentration and the amount adsorbed, respectively; k is a constant related to the energy of adsorption; and V is the adsorption maximum (Theng, 1971). The Langmuir isotherm only describes adsorption of molybdate over a limited range of concentration. Reyes and Jurinak (1967) studied the adsorption of molybdate on haematite at pH 4.0 and 7.75 by equilibrium dialysis at 22°and 40° C. They found that there were two distinct adsorption reactions occured at pH 4.0. The first reaction was insensitive to a change in temperature but the second reaction was endothermic. The lack of temperature dependence of the first reaction indicates a definite specific-site adsorption process which is one of the assumption of the Langmuir isotherm. Reyes and Jurinak (1967) also indicated that the potential adsorption sites of haematite occupied an average surface area of 22 to 23 Å ^{2}. Since the area of the tetrahedral form of molybdate ($H_xMoO_4^{x-2}$) closely approaches this value (25 Å ^{2}), it was adsorbed on the basis of geometric fit and possible electrochemical neutrality.

The adsorption maximum may be identified with a "monolayer" composed of molybdate ions and solvent molecules (Gile et al., 1960). The isotherm at pH 5.5 shows that the adsorption maximum decreases in proportion to its value at pH 4 (Theng, 1971). This decrease can be ascribed to a decrease in the affinity of molybdate for the clay surface as is also indicated by the parallel reduction in the initial slope of the respective isotherm. Barrow (1977) noted that the adsorption maximum indicated from within the limited range of concentration by Langmuir isotherm may not predict actual adsorption at higher concentrations because adsorption makes the surface more negative; hence, adsorption of one increment of molybdate makes adsorption of the next increment more difficult, i.e. the affinity term decreases as the amount adsorbed increases. Molybdate adsorption maxima, according to Langmuir equation, obtained from different soils have been reported as: 0.01 to 0.30 mmole/100 g for some Australian soils (Barrow, 1970), 2.6 to 10.0 mmole/100 g for New Zealand soils containing allophane minerals (Theng, 1971), and 2.5 to 19.3 mmole/100 g for volcanic-ash-derived soils in Chile (Gonzalez et al., 1974).

The isotherm for layer lattice silicate minerals (e.g. kaolinite and illite) at pH 4 shows two distinct regions of adsorption (Theng, 1971).

The shape of the isotherm clearly indicates that there are two energetically distinct sites on which adsorption takes place. Theng (1971) suggested that at the first region there was considerable "edge-to-face" aggregation in the original material causing a proportion of such sites to be difficult accessible. This type of particle arrangement is probably promoted by the presence of sesquioxides acting as cementing agents. The commencement of the second region of adsorption may therefore correspond to adsorption on the less accessible sites which gradually become exposed as molybdate begins to penetrate into crystal edges of silicate minerals when its concentration is raised beyond 3.5 ug/ml. Once such penetration is occurred, the presence of a molybdate ion would favor adsorption of the next anion on the adjacent sites (Theng, 1971). Studies on desorption suggested that desorption of molybdate from layer silicate minerals cannot be reversed by simply decreasing the solution concentration at constant pH and ionic strength as is predicted by the general specific adsorption theory of Hingston et al. (1967, 1968). Theng (1971) suggested that a specifically adsorbed anion can only be displaces by either increasing the pH or adding an anion which is capable of making the clay surface more negative than the equilibrium value at which adsorption takes place.

Another equation used to determine the molybdenum adsorption is the Freundlich isotherm. The Freundlich isotherm may be written as $x/M = kC^{1/n}$, where x/M is the amount of molybdenum adsorbed per unit weight of soil, C is the concentration of molybdenum in the equilibrium solution, k and n are constants. This equation was used by Reisenauer et al. (1962) who studied soils from northern California and hydrous oxides of iron, aluminum, and titanium. They indicated that molybdate adsorption by these materials at a given pH follows the Freundlich isotherm. Freundlich isotherm has the advantage of simplicity. Further, it can be shown (Hayward and Trapnell, 1964) that it is appropriate to adsorption in which an affinity term decreases as the amount adsorbed increases. The 1/n coefficient is not constant, but tends to increase with pH (Reisenauer et al., 1962; Barrow and Shaw, 1975). Nevertheless, the variation in 1/n coefficient is fairly small and the k term provides a useful index of the curve. The value of the k term indicates adsorption when the concentration is unity. Its value thus depends on the scale used for concentration. Karimian and Cox (1978) studied the adsorption of molybdenum from aqueous solution of eight soils from the Atlantic Coastal plains and Piedmont regions and reported that the data followed the Freundlich isotherm more consistently than the Langmuir isotherm. Adsorption isotherms for histosols showed that the level of molybdenum adsorbed increased with the organic matter content while adsorption isotherms for mineral soils showed that the amount of molybdenum adsorbed increased with the free iron oxides content.

Factors Affecting Molybdenum Adsorption in Soils

Soil pH

Soil pH is one of the most important factors affecting the adsorption of molybdenum in soils. Molybdate adsorption shows a maximum at a pH close to 4 (Theng, 1971). Above and below this pH the adsorption sharply declines. Reisenauer et al. (1962) indicated an increase in molybdenum adsorption with decreasing pH from 7.75 to 4.45. Reyes and Jurinak (1967) found no marked increase in adsorbed molybdate on haematite beyond 1.0 ppm Mo at pH 7.75. The adsorption of molybdate at pH 7.75 appeared to be completed at an equilibrium concentration of 2.0 ppm Mo. About five times as much molybdenum was adsorbed by haematite near saturation at pH 4.0 as at pH 7.75 (Reyes and Jurinak, 1967). The molybdate concentration in soil increased hundred-fold for each unit increase in pH (Vlek and Lindsay, 1977). Even wulfonite ($PbMoO_4$), one of the most stable minerals likely to form in soils, becomes more soluble as pH increase.

Concentration of Molybdate in Soil Solution

The adsorption of molybdate by soils and hydrous oxides of iron, aluminum and titanium increases exponentially with the equilibrium phase molybdate concentration at constant pH (Reisenauer et al., 1962). They also indicated that the solubility of molybdate in a soil system is a function of the percentage saturation of the molybdenum adsorbing surface and soil reaction. Molybdenum solubility increases with the amount of adsorbed molybdate present and with pH, and decreases with the molybdate adsorbing capacity of the soil.

Soil Iron and Aluminum

Molybdenum is not lost by reaching from acid soils other than sands, but is adsorbed on sesquioxide surfaces and slowly changed into less soluble form (Smith and Leeper, 1969). Jones (1956, 1957) reported that freshly prepared ferric oxides and a soil high in ferric oxides removed large amount of molybdenum from aqueous solution. He also indicated that aluminum oxides are capable of removing molybdenum from aqueous solutions but their effectiveness is much less than that of ferric oxides under the same conditions. It was suggested that molybdenum in ferruginous soils is held on the surface of the colloidal ferric oxides as the MoO_4^{2-} anion, which is replaceable by OH^- ions (Vlek and Lindsay, 1977). Jarrel and Dawson (1978) studied the sorption of molybdenum in soils of western Oregon which had pH ranges of 4.5 to 5.3 and concluded that the amorphous fraction of ferric oxides which was extracted with ammonium oxalate at pH 3.3, can be assigned a major role in molybdenum sorption in these soils. However, Karimian and Cox (1978) indicated that the amounts of molybdenum adsorbed increased with free ferric oxide content. Smith and Leeper (1969) suggested that molybdenum bound to ferric and aluminum oxides can be thought of as a semi-permanent form.

Phosphate, Sulfate, and Hydroxyl Ions

Gorlach et al. (1969) reported that competition from phosphate limited adsorption of molybdate, and also that phosphate could be used to displace some of the previously adsorbed molybdate. It seem possible that the amount of molybdate which could not be displaced by phosphate might indicate the amount converted into a more stable form. Barrow (1973) indicated that phosphate solutions can displace adsorbed molybdate from soil and the specific effect of the phosphate reached a maximum near pH 7 which is near the pK 2 of phosphoric acid. Gonzalez et al. (1974) indicated that adsorption of molybdate was decreased by the presence of phosphate solution when the ratio of Mo/P was 1/10. Increased effectiveness of phosphate in displacing molybdate with increase in phosphate concentration is also evidence for competitive displacement. However, this trend was reversed by further increases of phosphate concentration. It was suggested that very high concentration of phosphate disrupted the surface (Barrow, 1973). This disruption may have exposed further sites for molybdate adsorption or it may have caused some of the adsorbed molybdate to be "trapped" by a change in the molecular arrangement. Gonzalez et al. (1974) reported that sulfate ions did not compete with molybdate for the adsorption sites. High concentrations of hydroxyl ions were also able to displace previously adsorbed molybdate (Barrow, 1973). After prolong incubation of soil plus molybdate, displacement by hydroxyl ion was slow. This is further evidence that the nature of the bond to the surface had changed.

Organic Matter

The level of molybdenum adsorbed is closely related to soil organic matter (Karimian and Cox, 1978). Smith and Leeper (1969) proposed that for the molybdenum that is combined with organic matter, the association may be due to the incorporation of molybdenum into cellular structure by micro-organic activity or due to chelation of some of the molybdenum by poly-dentate organic compounds. Extracellular polysaccharides produced by bacteria have been shown to bind molybdenum with the consequence that less molybdenum was absorbed by plants (Lee and Loutit, 1977). They also reported that the polysaccharides which contain uronic acids appear to be responsible for binding molybdenum. Szalay and Szilagyi (1968) reported that the retention of molybdenum by organic soil is presumably due to the reducing properties of humic acids which transform anionic-$(MoO_4)^{2-}$ to -$(Mo)^{5+}$ that becomes fixed in the cationic form. However, iron oxides have been known to adsorb molybdenum by several workers (Jones, 1956; Reisenauer et al., 1962; Reyes and Jurinak, 1967) and the reaction of organic matter and iron in the soil has been demonstrated (Oades, 1963). It is therefore suggested that iron oxides bound to organic matter is actually responsible for molybdenum adsorption. This mechanism would required no reduction of molybdate to cationic form. Bonding of phosphate to fulvic acid through iron also has been reported (Levesque and Schnitzer, 1967). It is conceivable that a similar mechanism may be operative in molybdenum adsorption. The removal of organic matter from the soil resulted in a decrease in molybdenum retention by soils of 16.8 to 25.9 percents (Misra et al., 1977).

Time

Residual effects of molybdenum applied to soils vary widely. On some soils the effects may persist for several years; on others, further applications may be needed after two years (Smith and Leeper, 1969). This could be due to differences in adsorbing capacity of soils. Smith and Leeper (1969) concluded that, except in very sandy soils, the low residual value observed in some soils was caused by continuing reaction between molybdate and the soil; i.e., the initial adsorption was followed by a slow reorganization of molybdate on the colloid surface of soil particles. This reorganization results in a more-ordered arrangement, and bonds which are more difficult to break. Barrow (1973) reported that with prolonged incubation, the amount of molybdate which could be extracted by the phosphate solution fell at a decreasing rate. After 12 months' contact with the soil, about one-half to two-thirds of the molybdate added to some soils could be displaced by phosphate. Barrow (1977) stated that the adsorption reaction between soil and molybdate is first order with respect to the effect of concentration, i.e., the amount changed is proportional to the amount added. The effect of time differ from that expected for a first order reaction. The period required for each successive halving in concentration is not constant but increases with time, i.e., the reaction rate decreases with time.

Temperature

Whereas high temperatures of incubation resulted in low solution concentrations, high temperatures of measurement had the opposite effect (Barrow and Shaw, 1975). By these results, it was concluded that there were two distinct effects of temperature. High temperatures increased the rate of conversion of molybdate to a more tightly bound form and hence, indirectly resulted in low solution concentrations. However, the adsorption step itself was exothermic and high temperatures favored high solution concentration. These opposing effects of temperature could be separated by the procedure used but can be confounded using other techniques. Reyes and Jurinak (1967) studied the molybdenum adsorption of haematite and reported that at low molybdenum equilibrium concentration (0-15 ug/ml), the adsorption (the first surface reaction) was insensitive to temperature change and was completed at an equilibrium concentration of 10 ppm molybdenum. At high equilibrium concentrations (15-150 ug/ml) the adsorption (the second surface reaction) was dependent on temperature changes and the reaction was completed at about 55 ppm molybdenum.

Measurement of Adsorption

In all methods of measurement the general approach is to mix soil and molybdate solution and to calculate how much molybdate that is adsorbed from the change in solution concentration. In a simplest approach, only one level of molybdate is used. A difficulty with this approach is that if a range of adsorbing capacities is present, there may be some treatments on which most of the added molybdate is adsorbed. Differences between these treatments may then pass unnoticed. A more complete approach is to use several initial levels of molybdate and to plot the amount adsorbed against the final concentration at equilibrium. The resulting isotherm is taken as characteristic of the soil. A problem with this approach is that the position of the isotherm depends on the conditions of measurement. At a given concentration, the amount adsorbed increases with time (Barrow, 1970). The increase is rapid in the first few hours but then becomes slower and the reaction continues, through at an ever decreasing rate, for a long time. This problem is usually dealt with by making the measurements after an arbitrary period, e.g., 72 hours (Reisenauer et al., 1962), 24 hours (Barrow, 1970), 18 hours (Theng, 1971), or 6 hours (Gonzelez et al., 1974; Karimian and Cox, 1978). Adsorption also depends on the concentration of electrolytes in the medium in which adsorption is measured. When calcium chloride is used, adsorption increases as the electrolyte concentration increases (Barrow, 1972). The effect of electrolyte could partly be explained by assuming that the surface near the adsorbed molybdate is negatively charged. The effects of concentration are then seen as affecting the distribution of anions between the layer near the surface and the bulk of the solution (Barrow, 1972).

Absorption and Translocation of Molybdenum in Plants

It was reported that tomato plants absorbed and translocated sub-microgram amounts of radioactively tagged molybdate from single salt solutions of phosphate, chloride, nitrate and sulfate (Stout and Meagher, 1948). The greatest translocation of molybdate took place when present with phosphate salts and least in the presence of sulfates. Stout et al. (1951), who studied the absorption of molybdenum from culture solutions using radioactive isotopes of molybdenum, found that where sulfate salts were used, the lowest molybdenum concentrations were found in stems and blades of tomato plants, and where phosphate salts were used, the highest amounts of molybdenum were found in stems and blades. Since the total accumulation of molybdenum by roots in the presence of either phosphate or sulfate salts was not significantly different, Stout et al. (1951) suggested that there was a direct competition between sulfate and molybdate ions within translocation pathways through which divalent anions are transferred from roots to the conducting system of the plant. Stout and Meagher (1948) found a distinctly different pattern of distribution of absorbed molybdate from the patterns of other absorbed nutrient ions, such as phosphorus, potassium, manganese, and zinc. They reported that molybdenum was preferentially accumulated in interveinal areas of leaves whereas the other nutrient ions were preferentially absorbed in the midribs and veins. Fried (1948) has found precisely the same pattern of distribution of absorbed sulfates in alfalfa (Medicago sativa L.) as Stout and Meagher (1948) discovered for absorbed molybdate with the tomato (Lycopersicum esculentum L.) plant. This similarity may help to explain the competitive effect between molybdate and sulfate ions. Moreover, Fried (1948) reported that the distribution pattern of absorbed phosphates is directly opposite to that of sulfates. Calcium sulfate added to soils exercised a marked depression of molybdenum accumulated by tomatoes and peas (Pisum sativum L.) (Stout et al., 1951). The degree of inhibition could be quantitively assessed at different levels of added calcium sulfate ranging from low application of 219.7 kilograms per hectares to 3496.9 kilograms per hectares. Molybdate absorption by rice (Oryza sativa L.) grown in Hoagland solution was greater than those raised in calcium sulfate (Kannan and Ramani, 1978).

The absorption and transport of molybdate follow a biphasic pattern (Kannan and Ramani, 1978). Molybdate supplied to the primary leaf of rice plant was translocated to other parts, although most of it was transported to stem and root, suggesting that molybdate was mobile in plant. The presence of copper, chloride, and sulfate ions inhibited molybdenum absorption (Kannan and Ramani, 1978). There is also a marked reduction in molybdenum uptake in the presence of manganese and zinc ions. Kannan and Ramani (1978) also reported that the presence of $FeSO_4$ enhanced molybdate uptake and this enhancement did not occur in the presence of FeEDDHA. They commented that molybdate may possibly combine with Fe^{2+} dissociated from $FeSO_4$ and enter as iron molybdate.

Investigations on the absorption and translocation of molybdenum in soybean (Glycine max L. (Merr.)) grown in alluvial soil and Andosols were conducted in the field (Ishizuka, 1982). The molybdenum in the cotyledons was first translocated mainly to the stems, and then to the roots. With growth of the nodules, molybdenum was intensively transferred to the nodules, and more than 50 percents of the molybdenum was found in the nodules at early pod-filling stage. From the pod-setting stage, the molybdenum concentrations in the stems and leaves increased up to maturity. During the pod-filling stage, the molybdenum in the roots and pod shells is translocated to the seeds, and accumulated there. However, translocation of large amounts of molybdenum from the nodules, leaves and stem to the seeds does not appear to occur.

Roles of Molybdenum in Biological Nitrogen Fixation

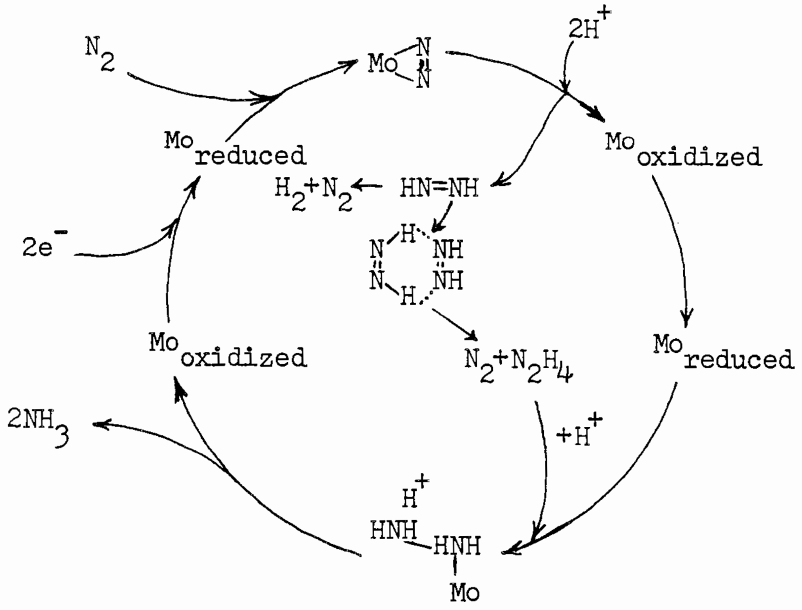
Molybdenum is the only second-row transition element known to possess defined biological functions and has been identified as a cofactor in a variety of bacterial, plant, mammalian, and nonmammalian enzymes (Schrauzer, 1976). The most important molybdenum enzymes are the nitrogenases. Nitrogenase is a key enzyme in the biological nitrogen fixation. It catalyzes the ATP-dependent 6 electron reduction of nitrogen to ammonia. Nitrogenase consists of two dissociating protein components, both of which are required for enzymic activity (Mortenson and Thorneley, 1979). The larger (Mo-Fe) protein (molybdoferredoxin) contains 2 Mo, 28-32 Fe and about 28 acid labile S atoms. The Fe protein is a dimer of molecular weight 57,674-73,000. The Mo-Fe protein contains two Fe-Mo cofactor centers. The Fe-Mo cofactor isolated by Shah and Brill (1979)'s method was extremely O_2-sensitive, but relatively stable under aerobic conditions in non-aqueous solvents.

The nitrogenase system has an absolute requirement of ATP. The nitrogen reduction catalyzed by nitrogenase is energy-demanding; the transfer of each pair of electrons is accomplished by the hydrolysis of four molecules of ATP (Winter and Burris, 1968). Kinetic studies have shown that ATP is required for electron transfer from the Fe protein to Mo-Fe protein (Eady et al., 1980). Under physiological conditions, ferredoxin or flavodoxin serves as a source of electron for the reduction of Fe protein (Ljones and Burris, 1978). Iron proteins transfer electrons to Mo-Fe protein. The Mo-Fe protein then can bind substrate and reduce it. Iron protein is a one-electron transfer agent (Ljones and Burris, 1978). As Mo-Fe protein must have multiples of two electrons to reduce substrates, a single transfer from one electron transfer agent, Fe protein, is insufficient. The transitory complex between the two proteins must dissociate after each electron transfer between them to accomplish the needed buildup of the electron pool on Mo-Fe protein (Hageman and Burris, 1978a). Until the Mo-Fe protein accumulates an adequate supply of electron to a proper state, it is not capable of transferring electrons to substrate.

The nitrogenase system is versatile in regard to the substrates that it can reduce. Not only does it reduce N_2, but it is also capable of reducing azide, cyanide, methyl isocyanide, acetylene, nitrous oxide, cyclopropane, and protons, and some analogous compounds (Burris and Hageman, 1980). All of the substrates apparently bind to Mo-Fe protein and receive their electrons from it; hence, all of them are in competition for electron from a single source. The question arises on how the Mo-Fe protein pool of electrons will be partitioned among substrates when more than one substrate is present at a time. Hageman and Burris (1978b) changed the balance between Mo-Fe protein while maintaining the electron flux through the whole nitrogenase system relatively constant. They found that under conditions of constant electron flux, electron transfer to N_2 was effective when there was approximately a one-to-one balance between Mo-Fe proteins and Fe protein. As the Mo-Fe protein concentration was increased, fewer of the electrons were transferred to N_2 and a higher percentage of them were used to produce H_2 and ethylene. It is also possible to alter the electron flux through the system and to determine how the alteration influences the partitioning of electrons to substrates. Hageman and Burris (1980) found that under conditions of rapid electron flux, N_2 functions effectively as an electron acceptor, whereas at low electron flux N_2 functions poorly and the bulk of electrons are utilized in the reduction of acetylene to ethylene or proton to H_2. They suggested that the electron flux through Fe protein or the concentration of ATP does not directly control electron allocation but rather indirectly controls it via their influence on the electron flux through Mo-Fe protein.

The model studies on mechanism of nitrogenase action established that the substrate chemistry of nitrogenase is molybdenum chemistry (Schrauzer, 1980). All substrate reactions are characteristic of a mononuclear Mo-active site located in a sterically partially obstructed or "pocket" environment. The Mo-active site is accessible to the protons of the medium as well as to substrates other than N_2. ATP hydrolysis is associated with the reduction of the Mo-active site. Substrates are bound either "end-on" or "side-on" (Schrauzer, 1980). Acetylene is a "side-on" substrate.

The stereospecific reduction of acetylene to cis-1,2-dideuterioethylene by nitrogenase in D_2O by analogy suggested that N_2 should be reduced to cis-diazene as the initial product (Schrauzer, 1980). Diazine (or diimide) is a highly reactive, unstable compound which has a great tendency to decompose into H_2 and N_2 or to disproportionate into N_2 and N_2H_2. Schrauzer et al. (1974) formulated the comprehensive diazene mechanism of biological nitrogen fixation as shown schematically in the figure below:



Nitrogen gas is assumed to interact with the reduced Mo catalyst in the side-on fashion just like acetylene (Schrauzer et al., 1974). The resulting nitride type intermediate complex undergoes hydrolysis to yield cis-diazene. Cis-diazene is not a substrate and accumulates in the vicinity of the active site, possibly within a "pocket" and either disproportionates into hydrazine or dinitrogen or decomposes into H_2 and N_2 without leaving the"pocket". Once formed, hydrazine is reduced to NH_3 at the Mo-active site (Schrauzer et al., 1974).

Responses to Molybdenum in Legumes

The criteria for response to added molybdenum in agricultural terms are increases in growth and yield of plant, nitrogen concentration, nitrogenase activity and Mo concentration in plant. Sometimes a visible symptom of deficiency may be present, and its disappearance or prevention form part of response.

Plant species differ in their response to molybdenum (Gladstone et al., 1977). Johansen et al. (1977) studied the response of several tropical pasture legumes, grown with Panicum maximum, to an initial application of MoO_3 over a five year period at six sites in southeastern Queensland. The most responsive legumes were found to be Glycine wightii and Desmodium intortum, followed by Macroptilium atropurpureum and Medicago sativa . Lotononis bainesii and Stylosanthes guianensis were least responsive.

Yield responses to molybdenum were always accompanied by increased nitrogen in plant tops but where a reduction in legume growth could be attributed to Mo deficiency, legumes showed chlorotic symptoms typical of N deficiency (Johansen et al., 1977). For example, desmodium plants growing at 10 percent~~s~~ of their maximum capacity were pale yellow with strong anthocyanin pigmentation. Plants became increasingly green with Mo treatment until maximum yield was reached. Nitrogen concentration of *Desmodium intortum*, *Macroptilium atropurpureum*, *Lotononis bainesii*, and *Glycine wightii* increased when molybdenum was applied (Kerridge et al., 1973). Johansen and Kerridge (1981) studied the relation between dry matter yield and N concentration in shoot samples that were collected for tropical legumes responding to Mo application. They reported that critical N concentration, those at 90% of the maximum yield, varied widely (2.2 to 3.6% N). They concluded that the wide variation of N response curves obtained prevented the establishment of critical N concentration sufficiently precise to indicate whether growth of Siratro, desmodium, glycine or lotononis was being limited by N supply. Only extreme cases of deficiency (less than 2% N) and sufficiency (more than 3.6% N) could be reliably defined. A substantial increase in leaf nitrogen of soybean was obtained from Mo applications regardless of lime treatments (Parker and Harris, 1962). Hagstrom and Berger (1963) also reported that the N content of soybean leaf was positively related to Mo supply.

The accumulation of molybdenum by plants was proportional to the amount added to the soil, with an increase of 5.2 ppm in alfalfa for each kg/ha Mo added to the soil (Jensen and Lesperance, 1971). Depth to water table was also associated with Mo accumulation in plant. Forage legumes grown where the water table was near the soil surface contained more molybdenum than did legumes grown where depth to water table was greater (Jensen and Lesperance, 1971). Molybdenum concentration in forage varied from cutting to cutting. At the 0.88 kg Mo/ha rate, the tissue Mo level of alfalfa and red clover (Trifolium pratense L.) grown on podzolic soils decreased sharply between the first and second cutting whereas only small differences was observed in the subsequent cuttings (Gupta, 1979). However, there was no such decline when lower rate of molybdenum was applied. A sharp decline in the tissue Mo concentration was likely due to the fact that high rates of molybdenum, its absorption by the plants is very rapid (Gupta, 1979). With soil application of 0.88 kg Mo/ha, tissue Mo concentration of greater than 10 ppm occurred in alfalfa and red clover limed to soil pH 5.6, 5.9 and 6.3 (Gupta, 1979). However. with soil application of 0.44 kg Mo/ha, tissue Mo concentration of only 0.92 and 0.31 were found in alfalfa and red clover, respectively. Gupta and MacLeod (1975) reported that applications of 0.5 to 1.0 ppm Mo to podzolic soil produced red clover tissue containing as high as 15 ppm Mo. However, Mo concentration in the second cutting tissue was reduced to 7.46 ppm for 1.0 ppm applied Mo treatment. Application of molybdenum to lateritic podzolic and red-brown earth soils consistently and markedly increased the concentration of molybdenum in subterranean clover (Trifolium subterraneum L.), particularly when applied with phosphorus (Reddy et al., 1981). The effect of phosphorus on the concentration of molybdenum in the plant was dependent on the levels of Mo applied. At low levels of Mo application (0 to 0.15 kg Mo/ha), phosphorus had only a small effect on concentration of molybdenum in the plant, but when 1.5 kg Mo/ha was applied, phosphorus markedly increased the concentration of molybdenum (Reddy et al., 1981). Generally, concentration of molybdenum increased as the level of molybdenum applied previously increased (Hawes et al., 1976). For crops sampled at maturity dates, Mo concentration was higher in plants sampled at earlier dates than when plants were matured. The lower values at mature harvest may be attributed to a dilution effect of dry matter yields (Hawes et al., 1976). Molybdenum content in the plant and total Mo yield of Siratro responded strongly to the two higher rates of Mo application (Ostrowski et al., 1978). Concentration of plant Mo ranged from 0.01 to 5.28 ug/g in alfalfa and from 0.12 to 9.44 ug/g in clover, and generally increased with Mo rate. Maximum yields of alfalfa were associated with plant Mo concentration greater than 0.5 ug/g (Mortvedt and Anderson, 1982). Molybdenum concentrations observed in nodule~~d~~s of *Phaseolus vulgaris* L. were higher than those observed in the root and leaf at the lowest external Mo concentration (Franco and Munns, 1981). At the highest external Mo (1 uM) both stems and leaves were important sinks, each containing about 330 ug/g compared with about 60 ug/g in nodules and roots. The highest correlation coefficient (0.982) was obtained for a linear relationship of molybdenum in nodule with that in stem. They suggested that the critical Mo concentration in nodule is between 3 and 5 ug/g. Franco and Munns (1981) also stated that in general over the whole range of experiments, root Mo evidently varies most with external Mo, and nodule Mo varies the least. Molybdenum application at high rates increased the number of nodules of **Siratro** (Ostrowski et al., 1978). However, there was a large increase in weight of nodules at all levels of applied Mo and Mo content in the nodule tissue increased with Mo application up to 280 g/ha.

Effects of Lime on Molybdenum Response in Legumes

Availability of molybdenum to both the nodule bacteria and plants is dependent on soil pH. Raising the pH of an acid soil to neutral or slightly alkaline levels by liming not only increases the availability of molybdenum to plants but also provides a favorable environmental condition for the growth of rhizobia (Date, 1970). However, nodulation of certain species was depressed when a Hawaiian Oxisol was limed at rates to raise soil pH to above 6 (Munns and Fox, 1976). A variety of reasons has been proposed to explain lime-induced depression of growth in the soil pH range of 6 to 7 (Kamprath, 1971), including induced phosphate deficiency in oxidic soil and direct adverse effects of calcium (Munns and Fox, 1976). Under certain conditions, the amount of lime required for maximum production of legumes may be sharply reduced or eliminated by Mo fertilization (Sim et al., 1974). Anthony (1967) suggested that the combination of lime and molybdenum appears to be the best approach to increase soybean yield. deMooy (1970) stated that the response to molybdenum was not related to soil pH; he reported soybean yield increases from added Mo at pH 6.7 that were of the same magnitude as those at lower pH's. This finding supports the observation by Lavy and Barber (1963) that responses were more related to soil type than to pH. Johansen et al. (1977) reported that soil sites differed markedly in their magnitude of legume response. For example, the most responsive site (xanthozem soil) required 200 g Mo/ha over five years for maximum growth of Siratro whereas there was no response of Siratro to Mo application at another site. However, Franco and Munns (1981) reported that shoot growth, nodulation and N_2 fixation of Phaseolus vulgaris L. grown in an Ultisol subsoil were restricted at pH below 5.9, regardless of Mo level. They indicated that once the inhibitory factor for nodulation or growth was removed by liming to pH 5.9, there was a tendency of Mo application to increase nitrogenase activity. Giddens and Perkins (1960) obtained an alfalfa response from 226.8 grams of sodium molybdate plus 560 kilograms of lime per hectare equivalent to that obtained from 2 tons of lime. Lime increased the Mo content of the alfalfa plants but did not produce a detectable increase in the available soil Mo (Giddens and Perkins, 1960). Liming increased both concentration and uptake of molybdenum in white clover (Trifolium repens L.) (Widdowson and Walker, 1971; Edmeades et al., 1983). Gupta et al. (1971) reported an herbage Mo concentration of 0.18 ug/g on a pH 4.9 soil which increase up to 0.62 ug/g when the soil pH was increased to 6.8 by liming. Lime alone did not increase available Mo to a level adequate for alfalfa production on highly oxidized soils of the Georgia Piedmont (pH 5.8 to 6.0), and additional Mo at 110 g Mo/ha was required to maintain the stands (Giddens and Perkins, 1972).

Lime was needed for successful alfalfa production in Canada on a silty clay loam (1.0 ppm total Mo) and on sandy clay loam (0.35 ppm total Mo) with pH level of 5.0 (Gupta, 1969). Additional Mo did not increase yield on the high-Mo soil, but Mo plus lime significantly increased yield on the low-Mo soils. Application of 650 g Mo/ha to soil (pH 5.0 to 6.0) averaging 0.74 ppm available Mo (by the anion exchange method) resulted in increasing subterranean clover yield by 20 percents, and plant Mo was increased from 0.16 to 0.58 ug/g (Dawson and Bhella, 1972). Application of lime, phosphorus, and molybdenum increased Mo concentration in the subterranean clover-ryegrass (Lolium perenne L.) mixture from 1 to over 5 ug/g (Petrie and Jackson, 1982). In Oregon, James et al. (1968) obtained an alfalfa yield response with lime applied to seven soils and with molybdenum applied to five soils having initial soil pH levels ranging from 5.0 to 5.8.

Effects of Molybdenum Seed Coating on Legumes

Dramatic yield increases are obtained on many soils from application of very small quantities of molybdenum fertilizers. Recommended soil applications of molybdenum is very low as compared to other nutrients, thus introducing application problems. Application of molybdenum in a macronutrient fertilizer has usually been used for establishing legume based pastures (Kerridge et al., 1973). This has generally proved a satisfactory method but there have been some instances of failure to correct deficiency due to uneven distribution of fertilizer and/or inadequate mixing of molybdenum in the macronutrient fertilizer.

Application of molybdenum with inoculated legume seeds has been recommended as an alternative method to soil application. Molybdenum applied as a seed treatment to pea was 30 to 60 times more efficient than equivalent amounts applied to the soil (Reisenauer, 1963). It appeared that application of molybdenum as a seed treatment provided less opportunity for reaction with the soil complex and, as a consequence, the seed-applied-Mo remained in a form usable by the plant. Thompson and Hsieh (1971) indicated that when molybdenum was coated on seed along with $CaCO_3$, alfalfa forage yields in a greenhouse experiment were more than doubled. Kerridge et al. (1973) found that the application of MoO_3 at a rate of 100 g Mo/ha in rock phosphate pellet was as effective as soil application in correcting Mo deficiency. They also reported that nodulation, yield and Mo concentration of Desmodium intortum, Glycine wightii, Lotononis bainsii and Macroptilium atropurpureum were similar whether molybdenum was applied with the seed pellet or to the soil. Hagstrom and Berger (1963) noted that the availability of Mo in the region around the seeds was greatly increased when inoculated seeds were planted in a mixture with peat, lime and Mo. Harris et al. (1971) reported that seed treatment with compounds containing Mo generally increased soybean grain yields on acid soils if the materials were applied as a slurry. No compound used in these studies was superior to sodium molybdate as a source of molybdenum. They also found no evidence of incompatibility between Mo and fungicide used. Giddens (1964) studied the effect of adding Mo compounds to soybean inoculant by mixing 7.09 and 28.35 grams of sodium molybdate, ammonium molybdate, molybdic oxide, or Moly-Gro (commercial material) with 141. 75 grams of inoculant and stored for the periods of 6 hours, 12 days, and 40 days before it was used to coat 27. 18 kilograms of seed. He found that the Mo treatment significantly reduced nodulation. Burton and Curley (1966) also reported that 99 percents of rhizobia was killed when sodium molybdate at 28.5 grams was mixed with 300 g inoculant for 4 days before inoculating to 27.18 kg soybean seed. However, they noted that the harmful effect of sodium molybdate on rhizobia was not manifested quickly. Highly effective nodulation was obtained when the sodium molybdate and inoculant were mixed and applied to the seed just before planting.

Relationships of Molybdenum, Sulfur, and Copper on Nutritional Quality of Forage Legumes

Metabolic disorders in grazing animals known as molybdenosis are widespread (Underwood, 1977). Characteristic symptoms of molybdenosis are diarrhea, listlessness, lost of hair colors and occasionally results in death (Alary et al., 1981). While the disorder may be due to deficiencies of Cu in the diet, they are frequently associated with ingestion of 'normal' amount of Cu. Retention of Cu by the animal is influenced by Mo and S and the latter elements are closely implicated in the development of Cu disorder (Wynne and McClymont, 1955). According to a review by Whitehead (1966), the amount of Mo which will induce Cu deficiency will depend on the Cu content of the diet. For example, when a pasture legume contains 5 ppm Cu, there must be about 7 ppm Mo to cause molybdenosis (Whitehead, 1966). Copper deficiency was observed in sheep when herbage Mo was in the range of 3 to 9 ppm and Cu in the range of 5 to 10 ppm (Wynne and McClymont, 1955). Kubota and Allaway (1972) pointed out that Mo toxicity (Mo-induced Cu deficiency) is an endemic nutritional problem of ruminant animals and is associated with plant accumulation of 10 ppm or more Mo in the presence of 4 to 10 ppm Cu concentrations. The critical Cu/Mo ratio with respect to the incidence of molybdenosis (hypocuprosis) in animal feeds from western Canada was found to be 2 (Miltimore and Mason, 1971), whereas in some English pasture legumes the ratio was reported to be closer to 4 (Allaway, 1973). However, Alary et al. (1981) reported that when the Cu/Mo ratio of fodder was less than 3, the livestock showed symptoms of molybdenosis. On the other hand, Cu toxicity may develop in sheep when the animals graze on pasture legumes containing 10 to 15 ppm Cu and extremely low levels of 0.1 to 0.2 ppm Mo (Underwood, 1971).

All cattle are susceptible to molybdenosis, with milking cows and young stock suffering the most, with sheep next in susceptibility, and with horses and pig being the most tolerant farm livestock (Underwood, 1976). In general nonruminants are less subject to molybdenosis (Allaway, 1968). Thiomolybdate, formed by the reaction between molybdate and sulfate, was proposed to be responsible for the depletion of Cu normally stored in the liver (Suttle, 1980).

Fertilizers can influence the chemical composition of pasture/forage legumes indirectly, by changing the botanical composition of the sward (Rossiter, 1966) or, directly, by influencing uptake of nutrient from the soil. Sulfur has an antagonistic influence on the adsorption of Mo (Stout et al., 1951). Application of S fertilizer has been reported to decrease Mo concentration in alfalfa and red clover (Gupta and MacLeod, 1975). Whereas Mo application of 0.5 to 1.0 ppm to fine sandy loam soil of pH 6.0 produced forage containing as much as 15 ppm Mo, tissue contration was less than 10 ppm when S was applied concurrently at 50 to 200 ppm. In the second and subsequent cuttings, 50 ppm applied S was adequate in alleviating Mo toxicity at all levels of applied Mo; moreover, resulting S concentrations were 0.15 to 0.21 percents. These concentrations are well below the plant tissue level of 0.6 to 0.9% S required to induce hypercuprosis (Gupta and MacLeod, 1975). Application of S at 22.5 and 67.5 kg/ha decreased the concentration of Mo in plants grown in lateritic podzolic and red brown earth soils (Reddy et al., 1981). They also reported that application of Mo to the soil had no effect on the concentration of Cu in the plant, but it consistently and markedly increased the concentration of Mo. The concentration of Cu in the plant was affected by soil type and the amount of S applied (Reddy et al., 1981). Application of S increased the concentration of Cu in plants grown in lateritic podzolic soils but not on those grown on red brown earth and calcareous sand.

Influence of Mycorrhizae on Legumes

Most of the nodulated legumes so far examined are also mycorrhizal (Barea and Azcon-Aguilar, 1983). Two of the three subfamilies included in the family Leguminosae (i.e., Papilionoideae and Mimosoideae) are known to have vesicular-arbuscular mycorrhiza (VAM) and rhizobial nodules. The formations of vesicular-arbuscular fungal entry points and nodules on a legume root occur simultaneously, usually within a few days after seed or seedling inoculation, and it appears that the two endophytes do not compete for infection sites (Smith and Bowen, 1979). Legume nodules are usually not invaded by the VAM fungus (Smith et al., 1979).

Influence of Mycorrhizae on Legume Growth

From the point of view of plant ecology and crop production, legumes are special because they can be supplied with the two major nutrients, P and N by naturally existing biological systems. As a consequence of simultaneous infection with rhizobia and mycorrhizal fungi, legumes can receive growth benefits because of improved P and N supplies (Munns and Mosse, 1980). Redente and Reeves (1981) found a decrease in mean root/shoot ratio in plants infected with both symbionts when compared with either symbiont inoculated alone. This decrease reflects the increased effeciency of the root system in providing the plant with essential levels of P and N for growth of plant. Crush (1974) reported that in phosphate-deficient soils, VAM increased growth of _Centrosema pubescens_, _Styloxanthes guyanensis_, _Trifolium repens_ and _Lotus pedunculatus_. He indicated that the tropical legumes were much more dependent on mycorrhizae for growth than the temperate species and their difference seems to be related to the degree of root hair development. _Lotus pedunculatus_, a temperate legume which has well-developed root hairs, was able to grow well without mycorrhizal infection. The tropical legumes _Centrosema pubescens_ and _Stylosanthes guyanensis_, which form few root hairs, exhibit a strong dependence on mycorrhizae. _Trifolium repens_ hold an intermediate position with regard to both root hair production and response to mycorrhizae.

Influence of Mycorrhizae on Nodulation and Nitrogen Fixation

Vesicular-arbuscular mycorrhizae strongly stimulate nodulation and nitrogen fixation of temperate and tropical legume (Crush, 1974). Using the $^{15}N_2$ tracer technique, Kucey and Paul (1982) confirmed that nodulated root systems of mycorrhizal faba beans (Vicia faba L.) fix more nitrogen than nodulated root systems of non-mycorrhizal plants. An increase in nodule biomass for plants infected with both rhizobia and mycorrhizal fungi was concluded to be the major factor increasing nitrogen fixation rates.

Early observations on this subject suggested several approaches to elucidation of physiological aspect of the legume-rhizobia-mycorrhiza interactions. One of these was whether mycorrhizae enhanced symbiotic nitrogen fixation only through the stimulation of host-plant nutrition, or whether they also exerted a more direct effect on nodulation and nitrogenase activity. The existence of such a direct availability of P to the nodules by mycorrhizal hyphae does not preclude, however, the importance of a suitable P nutrition, as acheived by mycorrhizal inoculation of the host as a condition for effective symbiotic nitrogen fixation (Barea and Azcon-Aguilar, 1983). If the plant is well-nourished, the nodules also will receive a suitable P supply for their function. The existence of a close relationship between host nutritional status and nodule formation has been reported by Mosse et al. (1976) who found that a plant did not nodulate unless its P concentration was at least 0.15%. Mycorrhizal infection helped the plant to reach this required level, and nodulation then occurred. Smith and Daft (1978) also confirmed that VAM increases the rate of nitrogen fixation.

Waidyanatha et al. (1979) reported that nitrogenase activity in Pueraria sp. still increased when the phosphate response growth curve become asymptotic. This finding therefore supports the suggestion that nodule function may be preferentially stimulated by mycorrhizal infection. Smith et al. (1979) indicated that mycorrhizal effect on nodulation, nitrogenase activity, and nodule efficiency (specific nitrogenase activity) occurs before any positive growth response to VAM in a low nutrient soil but not in a more fertile soil. They stated that a steady supply of P to root cells and to adjacent nodules, as would be available from continuous polyphosphate conversion at the fungus-root interface, would be stimulating to the development of effective nodule symbiosis and associated nitrogen fixation. Because there is no contact between the mycorrhizal fungi and rhizobial bacteroids, any phosphate ions must pass through the host cells (Barea and Azcon-Anguilar, 1983). The role played by the VAM-specific phosphatase in the arbuscules developed inside root cells adjacent to nodules was suggested as being of great significance in phosphate transfer to bacteroids (Asimi et al., 1980). Soluble phosphate can replace VAM in increasing nitrogenase activity in nodulated soybeans (Carling et al., 1978). Thus, they suggested that P is almost exclusively responsible for the mycorrhizal effects. However, the increased mycorrhizal uptake of water (Safir et al., 1972) and elements, other than P which are related to the infectivity and/or effectiveness of legume-rhizobia systems, may also be involved.

Influence of Mycorrhizae on Nutrient Uptake of Legumes

Phosphorus is probably the most important nutrient for growth and effective nodulation of legumes (Gibson, 1976). Vesicular-arbuscular mycorrhizae, which are common in legumes, can increase P uptake of plants growing in infertile or high phosphate fixing soils (Mosse, 1973; Powell and Daniel, 1978), mainly due to the capacity of mycorrhizal fungi to absorbed phosphate from soil and transfer it to the host root. In a P-deficient situation of a Tropeptic Eutrustox, plant concentration of P was enhanced by the mycorrhizal association (Yost and Fox, 1979). The levels of soil P at which fumigation cease to make a difference in the P percentage in plants of Glycine max (L.) Merr., Vigna unguiculata L., Leucaena leucocephala, and Stylosanthes hamata were 0.1, 0.2, 1.6, and 1.6 ug P/ml, respectively. They suggested that this P concentration order may be the order in which these legumes depend on mycorrhizae in P-deficient soils. They also noted that at the two lowest soil P levels, P uptake by mycorrhizal plants was 25 times greater than by plants without mycorrhizal associations.

It has been reported that mycorrhizal infection also increases the concentration of nutrients other than P in plant tissues, but it is unclear if this enhancement of nutrient uptake is merely a consequence of improve P supply. Mycorrhizal hyphae will help the plant to overcome uptake limitations in the case of nutrients that diffuse slowly and give way to depleted zones around roots (Barea and Azcon-Aguilar, 1983). Conversely, mycorrhizae will confer little additional advantage for the uptake of ions such as sulfate which may move through soil by mass flow with no rate-limiting step in its way to the root surface. In fact, Cooper and Tinker (1978) observed that the flux of S into clover seedlings was small suggesting that S nutrition may not be aided greatly by VAM. However, Gray and Gerdemann (1973) reported that VAM on red clover absorbed ^{35}S at higher rate than did comparable nonmycorrhizal roots.

Cowpea and soybean grown on a nonfumigated Tropeptic Eutrustox with soil P levels below 0.025 to 0.05 mg P/l contained higher Ca and K percentages than plants grown on fumigated, low P-status soils (Yost and Fox, 1982). However, they indicated that differences in Ca and K percentages in the plant were more closely related to crop growth rates than to VAM infection levels. Yost and Fox (1982) also found that Si percentages of mycorrhizal soybean plants were 0.5 to 0.9% Si while nonmycorrhizal plants contained 0.2 to 0.3% Si. Silicon content of cowpea, however, was not altered by the presence or absence of mycorrhizae. They hypothesized that if mycorrhizae consistently enhance Si uptake by plants, such as soybean, then Si uptake may indicate mycorrhizal activity.

Ross (1971) reported that concentrations of N, P, Ca, and Cu in foliage of mycorrhizal soybean plants were greater than those from nonmycorrhizal plants at the various P levels. However, since concentrations of Ca and N in nonmycorrhizal plants were directly related to phosphate fertilization, accumulation of these elements may be associated with increased phosphate absorption per se rather than with fungal activity. Lambert et al. (1979) indicated that two micronutrients, Cu and Zn, are frequently at higher concentration in shoots of mycorrhizal plants, compared with that in nonmycorrhizal plants. Moreover, P fertilization significantly reduced Cu and Zn concentrations in mycorrhizal soybeans, but concentrations of these nutrients in nonmycorrhizal treatments were not affected.

Hayman and Day (1978), who grew black beans (<u>Phaseolus</u> <u>vulgaris</u> var. Venezuela) in low phosphate (3 ppm P) soil from Brazil, suggested that the mechanism whereby mycorrhizae greatly increase phosphate uptake do not apply to molybdate which seem to be the main factor limiting plant growth under these conditions. They reported that bean plants inoculated with endophyte develop very dense mycorrhizal infection and took up twice as much P as the uninoculated control plants. However, plants in both inoculated and uninoculated treatments remained stunted and only those given monocalcium phosphate nodulated well and grew quite extensively and showed no symptoms of Mo deficiency.

It is clear that direct experimentation on the role of mycorrhizae in the uptake of plant nutrients is required. The formation gained would have application in the field of nutrient deficiencies and crop production.

GENERAL MATERIALS AND METHODS

Soil Used

Wahiawa Series:

This series is classified as a clayey, kaolinitic, isohyperthermic family of Tropeptic Eutrustox. These soils developed in residuum and old alluvium derived from basic igneous rock. They are nearly level to moderately steep. Elevations range from 150 to 365 m. Rainfall amounts to 1000 to 1500 mm annually. The mean annual soil temperature is 22° C. This series consists of well-drained soils on uplands on the island of Oahu. The clay fraction in this soil is abundant in kaolinite, haematite, maghemite, and gibbsite. The soil used in the experiments was collected from the Poamoho Research Station of the Hawaii Institute of Tropical Agriculture and Human Resources (HITAHR), Island of Oahu.

Paaloa Series:

This series is classified as a clayey, oxidic, isothermic family of Humoxic Tropohumult. This soils developed in old alluvium and residuum derived from basic igneous rock. They are gently to moderately sloping. Elevations range from 305 to 518 m. The annual rainfall amounts to 1778 to 2286 mm. The mean annual soil temperature is 21° C. These soils are well-drained. The clay fraction is abundant in haematite, maghemite, gibbsite and goethite. The soil used in the experiments was collected from a field of the Waialua Sugar Co. Inc. located about 8 miles east of Waialua, Island of Oahu.

Plant Species Used

Some characteristics of plant species used in the experiments as described by Bogdan (1977) are as follows:

Desmodium intortum (Mill.) Urb. cv. Greenleaf desmodium:

It is a large perennial herb with erect, ascending or scandent, much branched, and often with reddish-brown stems. Leaves are trifoliate with ovate leaflets which are 2-7 cm long and 1.5-5 cm wide. Brown- or red-speckling usually occurs on the upper side of the leaf. The numerous pink or mauve flowers are in terminal and axillary racemes. Pods are falcate, with 8-12 one-seeded joints. Seeds are small. Tolerance to soil acidity of pH 5 is good. *Desmodium intortum* is highly specific for *Rhizobium* inoculant.

Centrosema pubescens Benth. cv. Centro:

It is a climbing and trailing perennial with slender pubescent stems. The leaves have three leaflets. The leaflets are oblong to ovate-laceolate, 1.5-7 cm long and 0.6-4.5 cm wide, finely pubescent on the lower side, and seldom glabrous. Flowers are in dense and short axillary racemes, on a peduncle up to 6 cm long. Pods are linear, 4-17 cm long and 6-7 mm wide, with hard-raised margins and a narrow straight beak at the apex, and dehiscent at maturity. Seeds are red-brown, streaked with black, 4-5 mm long, 3 mm wide and 2 mm thick. It is slightly less sensitive to Mn toxicity than the majority of other legumes. *Centrosema pubescens* is highly selective in its *Rhizobium* requirement.

Stylosanthes humilis H.B.K. cv. Townsville stylo:

It is an erect or subprostrated annual with stems up to 1 m long. Leaves are trifoliate, with narrow-ovate sharply pointed leaflets up to 2 cm long. Flowers are yellow to orange, in terminal clusters. Pods are 7-10 mm long consisting of two joints, of which the lower part contains one seed, whereas the terminal joint, which is 4-6 mm long, is empty and is hook-shaped. Seeds are kidney-shaped, 2-3 mm long, sometimes yellowish, but mostly brown or purplish-black. *Stylosanthes humilis* tolerates soil acidity up to pH 4 and a high level of Al and Mn. The inoculant of the cowpea group can be used to inoculate stylo seeds.

Seed weight, Mo concentration in seeds and amount of Mo per seed of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* are as follows:

Legume species	Wt of 100 seeds	Conc of Mo	Amt of Mo per seed
	g	ug/g	ug
Desmodium intortum	0.183	1.080	0.002
Centrosema pubescens	2.410	0.597	0.014
Stylosanthes humilis	0.350	0.721	0.003

Soil Test and Plant Analysis

Molybdenum Analysis in Soil and Plant

Methods of soil and plant analyses for Mo including sample preparation are based on methods described by Purvis and Peterson (1956), Sandell(1959), and Reisenauer (1965).

Preparations of Samples for Analysis:

a. Soil Samples

Ten grams of soil were shaken with 100 ml of acid ammonium oxalate solution (pH 3.3) for 12 hours at room temperature. The mixture was centrifuged or filtered and 70 ml of filtrate was evaporated to dryness in Pyrex beaker. The beaker was heated in muffle furnace at 450° C for 4 hours to destroy the oxalate. The residue was dissolved in 20 ml of 6.5 N HCl, and filtered. The filtrate was made up to 100-ml volume for Mo determination.

b. Plant Samples

Eight-gram sample of oven-dried, finely ground plant material was weighed into a porcelain crucible. The crucible was ignite to 500° C for 18 hours in a muffle furnace. The residue was dissolved in 20 ml of 6.5 N HCl containing 0.5 g/l $FeCl_3$. The dissolving residue was covered with a watchglass and was left overnight. The dissolved residue was filtered through a No. 42 filter paper into a 100 ml volumetric flask and was diluted to volume.

c. Nodule Samples

Oven-dried nodule was weighed into a porcelain crucible. The crucible was ignited to 500° C for 8 hours in the muffle furnace. The residue was dissolved in 20 ml of 6.5 N HCl containing 0.5 g/l $FeCl_3$. The dissolved residue was filtered through a No. 42 filter paper into a 100 ml volumetric flask and was diluted to volume.

Determination of Molybdenum

A 100 ml of test solution was transferred to a 125-ml separatory funnel. Ten ml of a mixture of 1:1 carbon tetrachloride-isoamyl alcohol was added and the mixture was shaken vigorously for 100 times to saturate the aqueous phase. The liquid phase was allowed to separate for 15 minutes and the organic phase was removed quantitatively. Two ml of 40% (w/v) potassium thiocyanate was mixed and the mixture was shaken vigorously. Two ml of 40% stanous chloride in 1.5 N HCl was added into the mixture. Exactly ten ml of carbon tetrachloride-isoamyl alcohol mixture was added. The mixture was shaken vigorously for 150 times and the phase was allowed to separate for 15 minutes. The colored extract was transferred to spectrophotometric cell and the optical density of the solution was measured by a Klett-Summerson photoelectric colorimeter using filter # 47 (475 millimicrons).

Sulfur Analysis in Soil

A turbidimetric method for determining sulfates in soil extracts (R. L. Fox, unpublished mimeograph. University of Hawaii, Manoa) was used.

Extraction Procedure:

Two and a half grams of oven-dried soil was weighed into a 50-ml centrifuge tube. Twenty five ml of the phosphate extracting solution (0.04 M $Ca(H_2PO_4)_2$) was added and the mixture was shaken for 16 hours at room temperature. The suspension was centrifuged and then 20 ml of the supernatant solution was decanted into another centrifuge tube. Two-tenth gram of washed charcoal was added. After 30 minutes, the suspension was filtered to obtain a clear extract.

Barium Sulfate Precipitation Procedure

Five to ten ml of the decolorized extract was transferred into a 25-ml beaker and was evaporated to dryness in a drying oven at approximately 100° C. One ml of 0.25 N HCl containing 20 ug SO_4-S was added. The beaker containing the residue was rotated with the acid-sulfate reagent. After the residue has dissolved, one ml of $BaCl_2$, seed crystal-gelatin-reagent was added. The content in the beaker was swirled and the suspension was allowed to stand for 5 minutes and then 10 ml of 1% $BaCl_2$ solution was added. The suspension was swirled again, and after 25 minutes it was titurated with a rubber policeman. The suspension was swirled again and it was immediately transferred to a cuvette. The absorbance at the wavelength of 600 millimicrons was read. Gross SO_4-S was determined from an appropriate standard curve.

Determination of Other Nutrient Elements in Plant

Whole plant materials were analyzed for P, K, Ca, Mg, S, Fe, Mn, Cu, Zn, Al, and Si by x-ray emission spectroscopy. Nitrogen was determined by micro-kjeldahl digestion and a colorimetric procedure of a Technicon Auto-Analyzer.

Acetylene Reduction Assessment of Nitrogen Fixation

The acetylene reduction assay used was modified from the method described by Hardy et al. (1968).

Roots with nodules intact were placed in a 500-ml glass jar that was made air tight with a screw cap fitted with a rubber serum stopper. Ten percents of the air in the jar was withdrawn by hypodermic syringe and replaced with an equivalent amount of C_2H_2. The jars were incubated at room temperature. After 1 hour, 2-ml sample of gas was transferred by a gas-tight syringe from the jars to a 20-ml pyrex vial. A 0.2-ml volume of gas sample was analyzed for C_2H_4 by hydrogen flame ionization in a Perkin-Elmer Model Sigma 3B gas chromatograph.

Lime Requirement Determination

Fifty-g sample of seived air-dried soil was placed in beakers. Graduated quantities of $Ca(OH)_2$ were added to separate soil samples, mixed together and water required to meet 1:2 water to soil ratio was added. The treated soils were equilibrated at room temperature for 2 weeks. The pH of suspension was determined using a glass electrode. Liming standard curves relating pH values to quantity of lime were then prepared from these data and were used to determine the lime required to raise soil pH to any desired level.

CHAPTER 1

EFFECTS OF MOLYBDENUM, SULFUR AND LIME ON GROWTH, YIELD AND NUTRIENT CONCENTRATION OF *DESMODIUM INTORTUM* AND *CENTROSEMA PUBESCENS* GROWM IN ACID TROPICAL SOILS

INTRODUCTION

One way to increase the production of pasture legumes grown in acid tropical soils is to apply Mo fertilizer since Mo availability of these soils is generally low. The typical Mo deficient soil is a very acid (pH 4.5-5.0) highly weathered soil with sesquioxides and kaolin making up much of the soil mineralogy (Kanehiro et al., 1983). Molybdate is adsorbed strongly by oxides of Fe and Al (Jones, 1957; Jarrell and Dawson, 1978). By these findings, Oxisols and Ultisols should be quite low in available Mo.

Response of legumes to Mo vary from place to place. While there was no response when total Mo in soils exceeds 1.64 ppm for Louisiana soils (Boswell, 1980) there was a response of legumes grown in Hawaiian Oxisols containing 2.8 ppm Mo (Sherman and Kapteyn, 1966; Younge and Takahashi, 1953). Recommendation rates of Mo fertilizer also vary from location to location. In Australia the recommendation rate for forage legumes is 140 g Mo/ha (Ostowski et al., 1978), whereas in the southern states of the U.S. the rate is 50 to 100 g Mo/ha (Mortvedt and Anderson, 1982). These rates are quite low when they are compared to Mo fertilizer rates of 1 kg Mo/ha and 2.24 kg Mo/ha for Hawaiian Ultisols and Oxisols, respectively (Taal, 1979; Younge and Takahashi, 1953).

Dramatic yield increases are obtained on many soils from application of very small quantities of Mo fertilizers. Recommended soil applications of the nutrient used in such small quantities introducing application problems. The importance of seed reserves in supplying plant needs for the element (Peterson and Purvis, 1961; Lavy and Barber, 1963) and the response reported from soaking seed in molybdate solution (Donald and Spencer, 1952) suggest seed application as an alternate method of applying Mo fertilizers.

Seed coating is ~~one of the~~ an effective method ~~recommended~~ for Mo application. Application of Mo as a seed treatment provides less opportunity for Mo reaction with the soil complex and as a consequence the seed-applied Mo remains in a form available to plant. However, it is known that Mo salt may kill rhizobia and consequently reduce nodulation, ~~although~~ the detrimental effect of Mo salt may not manifest quickly if it is coated to the seed just before planting. Coating seed with Mo is also beleived to be responsible for high Mo concentration in pasture legumes because of the high concentration of Mo around seed. High Mo in plant in turn may cause molybdenosis when the ratio of Cu/Mo is lower than 2-4. However, high rate of S fertilizes has been used to decrease the toxic level to animal of Mo (Gupta and MacLeod, 1975) as well as to increase plant growth, application of S along with seed-applied Mo should increase plant growth and prevent molybdenosis.

Liming is also one of the practical methods used to correct Mo deficiency. Liming increases soil pH and as a consequence increases Mo availability. Lime also alleviates Mn and/or Al toxicity which is a serious problem in many acid tropical soils ~~if left uncorrected~~. Manganese and/or Al toxicity may limit response to Mo in legumes. Liming therefore not only increases the availability of Mo to plants but also provides a favorable environmental condition for the growth of rhizobia.

Since Mo, S and lime are important factors affecting growth and chemical composition of legumes in the tropics, studies on these components as affecting response to Mo and nutritional quality of pasture/forage legumes become necessary.

This part of the study was conducted in the greenhouse and in the field with the following objectives:

1. To study the effects of Mo, S, and lime on growth, yield and biological nitrogen fixation of *Desmodium intortum* and *Centrosema pubescens* grown in acid tropical soils;

2. To study the effect of lime in alleviating Mn and/or Al toxicity and in correcting Mo deficiency; and

3. To study the effects of Mo, S and lime on some nutrient concentrations in *Desmodium intortum* and *Centrosema pubescens*.

MATERIALS AND METHODS

a. Greenhouse Experiments

[illegible]

a.1. Procedures

Pot experiments were conducted at the Agronomy and Soil Science Field Laboratory, Mauka campus of the University of Hawaii at Manoa. Desmodium intortum was grown in the Wahiawa and Paaloa soils. Centrosema pubescens was grown in the Wahiawa soil. Some chemical properties of both soils are shown in Table 1.1.

Treatments were composed of five levels of Mo (none added, Mo seed coating at rates of 0.1 and 0.5 kg Mo/ha, and Mo soil application at the rates of 2.0 and 4.0 kg Mo/ha); two rates of S (none added and 50.0 kg S/ha); and two rates of lime (none added and lime rate so that the soil pH is raised to 6.0). These treatments made up the 5x2x2 factorial arrangement in a randomized complete block design with three replications. The legumes were inoculated with the appropriate Rhizobium strains. Phosphorus, K, Mg, Zn, and B were added to all treatments as part of the basal fertilizer application. Thirty percent gum arabic solution was used as adhesive material for Rhizobium inoculation and Mo seed coating treatment.

Fifteen treated seeds of a legume were planted in a pot. After three weeks from planting, the seedlings were thinned to three uniformly growing plants per pot. Throughout the experiment the pots were watered to 100% field capacity. The plants were harvested at

Table 1.1.

Some chemical properties of soils used in the greenhouse and field experiments.

Chemical properties	Wahiawa soil	Paaloa soil
pH (1:1 soil:water)	5.20	4.67
Organic carbon (%)	1.12	3.26
Available P (modified Truog) (ug/g)	146.0	-
Water soluble P (ug/ml)	0.07	-
Exchangeable bases (NH_4OAc extraction) (meq/100 g)		
K	0.65	0.20
Ca	4.13	1.14
Mg	1.30	0.70
Na	0.17	0.25
Exchangeable Al (KCl extraction) (meq/100 g)	0.05	2.60
Effective C.E.C. (meq/100 g)	6.25	4.89
KCl extractable Mn (ppm)	26.0	17.25
Extractable Mo (ppm)	0.38	0.30
Available S (ppm)	49.2	80.0

60 days after planting. They were cut approximately 1 cm above the soil surface and the roots were carefully removed from the soil with a minimum detachment of nodules. The roots with nodules were put into a glass jar for acetylene reduction assessment. Plant samples were washed and oven-dried at 70° C in air-draft oven for three days for determining dry matter yield. The oven-dried samples were ground(20 mesh) in a stainless steel Wiley Mill and kept for elemental analysis.

a.2. Treatment Preparation

a.2.1. Fertilizers and Lime Used

Fertilizers and lime used in the greenhouse experiments were as follows.

Basal fertilizers:

- Calcium phosphate, monobasic ($Ca(H_2PO_4)_2$ at 600 kg P/ha.
- Potassium chloride (KCl) at 200 kg K/ha.
- Magnesium chloride ($MgCl_2.6H_2O$) at 200 kg Mg/ha.
- Zinc chloride ($ZnCl_2$) at 20 kg Zn/ha.
- Boric acid (H_3BO_3) at 2 kg B/ha.

Fertilizers and lime used for treatments:

- Sodium molybdate ($Na_2MoO_4.2H_2O$) as source of Mo.
- Magnesium sulfate ($MgSO_4.7H_2O$) as source of S.
- Calcium hydroxide ($Ca(OH)_2$) as source of lime.

For the Wahiawa **soil** , basal fertilizers were used in the solid form. Each chemical was weighed separately at the required amount for each pot and the five chemicals were mixed together before applying to the soil. The basal fertilizers were thoroughly mixed with the soil.

The fertilized soil was then put back into the pot and it was readied for seeding.

For the Paaloa soil, basal fertilizers except the phosphate fertilizer were used in the solution form. Each chemical was weighed at the required amount for the whole treatment, mixed together in solution form and made up to 1.8-l volume. The solution fertilizer was used at 30 ml per pot. Phosphate fertilizer was weighed separately at the required amount for each pot and mixed with soil along with the solution fertilizer.

a.2.2. Preparation of Seed-Coated Treatment

Molybdenum-coated seeds were prepared by adding the required amount of sodium molybdate to gum arabic solution, mixing together, and then coating the seed. *Rhizobium* bacteria were inoculated to the seed after the seed was coated with sodium molybdate-gum arabic solution mixture. The treated seeds were spread to dry before they were used for planting. These Mo-coated seeds were prepared just before planting.

a.2.3. Seed Preparation

Seeds of *Centrosema pubescens* were scarified by immersing in concentrated H_2SO_4 for 10 minutes. After draining the acid, the seeds were washed many times with deionized water. The scarified seeds were spread to dry before they were treated with sodium molybdate and inoculated with the appropriated *Rhizobium* strains. Seeds of *Desmodium intortum* needed not to scarify.

a.2.4. Soil Preparation

The soil was sieved through a 1-cm sized sieve to preclude gravels, stones and large roots and spread on benches to dry. After air-drying and thoroughly mixing to attain homogeneity, a sample was taken to determine moisture content as well as to conduct soil test for fertilizer application, e.g. test for pH, lime requirement, available P, etc. Six kilograms of oven-dried soil was put into 8-l plastic pots for subsequent liming and fertilizer application. For lime treatment, $Ca(OH)_2$ at the amount needed to raise soil pH to 6.0 according to the lime requirement curve was mixed with soil and the limed soil was incubated for 2 weeks before fertilizer application and seeding.

b. Field Experiment

A field experiment was conducted at the Poamoho Research Station of the Hawaii Institute of Tropical Agriculture and Human Resources (HITAHR), Island of Oahu. Two pasture legumes were grown in the Wahiawa soil at Field F of the Station. The experimental design used was a 2x10 factorial arrangement in a randomized complete block design with three replications. The treatments were as follows.

Crops:

- *Desmodium intortum*
- *Centrosema pubescens*

Molybdenum and lime treatments:

- No Mo.

- $CaCO_3$ (at the rate so that the soil pH was raised to 6.0).
- Seed-applied Mo at 0.1 kg Mo/ha.
- Seed-applied Mo at 0.3 kg Mo/ha.
- Seed-applied Mo at 0.5 kg Mo/ha.
- Seed-applied Mo at 0.5 kg Mo/ha plus lime.
- Soil-applied Mo at 1.0 kg Mo/ha.
- Soil-applied Mo at 2.0 kg Mo/ha.
- Soil-applied Mo at 4.0 kg Mo/ha.
- Soil-applied Mo at 4.0 kg Mo/ha plus lime.

A plot consisted of four rows, 3.66 m in length; the rows were spaced 0.91 m apart. $CaCO_3$ was applied one month before planting. Seeding rate was approximately 52 seeds per 1 m of row. Seeds were planted by hand and seedlings were thinned to 13 plants per 1 m of row 48 days after planting. ~~Furrow~~ Irrigation water was ~~done routinely~~ in furrows as needed every week. The legumes were cut five times. The first cutting was ~~done~~ taken when the legumes were 112 days old. Second, third and fourth cuttings were ~~done~~ taken at 60-day intervals. The fifth cutting was ~~done~~ taken at 90 days after the fourth cutting. The Area harvested was the two middle rows of 3.05 m in length.

c. Measurements of Responses

c.1. Yield Response

Plants were observed for deficiency and/or toxicity symptoms. Dry matter yield was determined at harvest. Efficiency of Mo applied as seed coating and soil treatment regarding dry matter yield

define

was compared. The interaction of Mo, S and lime on yield was examined. The relationship between yield and Mo concentration in plant was determined.

c.2. Nodulation

The nodule number and weight of legumes from the greenhouse experiments were determined at harvest in order to assess the effects of Mo, S and lime on nodulation.

c.3. Nitrogenase Activity

Nitrogenase activity of nodules from the greenhouse experiments was evaluated by using acetylene reduction technique. Nodule efficiency (specific acetylene-reducing activity of nitrogenase) was determined. The relationship between nitrogenase activity and Mo concentration was evaluated.

c.4. Chemical Composition of Plant

Plant tops at harvest were analyzed for N, P, K, Ca, Mg, S, Fe, Mn, Cu, Zn, Si, Al, and Mo. Nodules were analyzed for Mo. The relationship between Mo concentration in plant tops and Mo concentration in nodules was determined. The relationship between some nutrient elements, especially N, S and Cu and Mo concentration in plant tops and nodules was, also determined. A Cu/Mo ratio in plant tops was calculated ~~in order~~ to help evaluate the nutritional quality, ~~of legumes~~.

c.5. Chemical Composition in Soil

Soil samples were analyzed for organic **carbon,** pH, C.E.C., P, K, Ca, Mg, Na, S, Al, Mn, and Mo in order to evaluate the fertility status of soils used in the experiments. The relationships between extractable Mo and dry matter yield, nitrogenase activity and some nutrient elements in plant, e.g. N, S, and Cu were also determined.

RESULTS AND DISCUSSION

a. Greenhouse Experiments

a.1. Effects of Mo, S and Lime on Growth, Yield and Nutrient Concentration of *Desmodium intortum* Grown in the Wahiawa Soil

Yield Response

Applications of Mo as seed coating and soil treatment significantly increased dry matter yield of desmodium grown in the Wahiawa soil (Table 1.2). Yield of desmodium grown from seed coated with 0.5 kg Mo/ha was significantly increased 25.3% above that of desmodium grown without Mo application. Yields of desmodium applied with Mo as soil treatment at 2.0 and 4.0 kg Mo/ha were significantly increased 27.3% and 29.4%, respectively, above those of desmodium receiving no Mo. However, yield obtained from the 0.1 kg Mo/ha seed-applied treatment was increased only 6.9% above that obtained from the no-Mo treatment and this difference was not significant. Yield obtained from the 0.5 kg Mo/ha seed-applied treatment was not significantly different from that obtained from 2.0 or 4.0 kg Mo/ha soil-applied treatment, suggesting that seed-applied Mo at this rate provides adequate amount of Mo for plant need. Yield obtained from the 0.1 kg Mo/ha seed-applied treatment was significantly lower than that obtained from the 0.5 kg Mo/ha seed-applied treatment, 2.0 and 4.0 kg Mo/ha soil-applied treatments, indicating that coating seed with Mo at 0.1 kg Mo was not enough for the requirement of desmodium grown in the Wahiawa soil.

Table 1.2

Effects of Mo, S, and lime on dry matter yield of *Desmodium intortum* grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	g/pot				
No Mo added	25.3	35.1	33.1	34.6	32.0 b*
0.1 kg Mo/ha (seed applied)	33.4	33.0	34.2	36.3	34.2 b
0.5 kg Mo/ha (seed applied)	34.6	40.3	43.7	41.7	40.1 a
2.0 kg Mo/ha (soil applied)	37.1	40.0	46.1	39.4	40.7 a
4.0 kg Mo/ha (soil applied)	43.5	38.9	39.9	43.4	41.4 a
	34.8	37.5	39.4	39.1	
Mean	36.1 b		39.2 a		
	No S		S		
Mean	37.1 a		38.3 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Sulfur application at the rate of 50 kg S/ha slightly increased desmodium dry matter yield (3.2%) above the no-S treatment when averaged over all Mo and lime rates. Lack of response to S fertilizer was due to sufficient amount of available S in soil (Table 1.1).

Application of lime at a rate such that soil pH was raised to 6.0 significantly affected dry matter yield of desmodium (Table 1.2). When averaged over all Mo and S rates, yield obtained from the lime treatment was significantly higher (8.6%) than that obtained from the no-lime treatment. When no Mo and S were applied, lime greatly increased yield (30.8%) above the no-lime treatment. The influence of lime tended to decline as the rate of Mo was increased, especially for the Mo soil application (Fig. 1.1), suggesting that there may be an interaction between Mo and lime, although the interaction effect between these two factors was not statistically significant. Lime increased yield of desmodium when applied with both levels of Mo as seed coating (Fig. 1.1). However, when lime was not applied along with the Mo-coated seeds, the rate of increase of yield decreased as the level of Mo was increased, i.e. the increment of increase of yield due to lime at 0.5 kg Mo/ha was higher than that of yield at 0.1 kg Mo/ha. Effect of lime in increasing yield may in part be due to the influence of lime on nodulation.Seed-applied Mo at 0.5 kg Mo/ha tended to reduce nodulation (Table 1.4). Molybdenum salts were reported to reduce nodulation if they are mixed with inoculated seed (Giddens, 1964; Burton and Curley, 1966). Lime therefore may alleviate the detrimental effect of Mo salt since it provides favorable condition for <u>Rhizobium</u> growth.

In order to compare the efficiency of seed-applied Mo and soil-applied

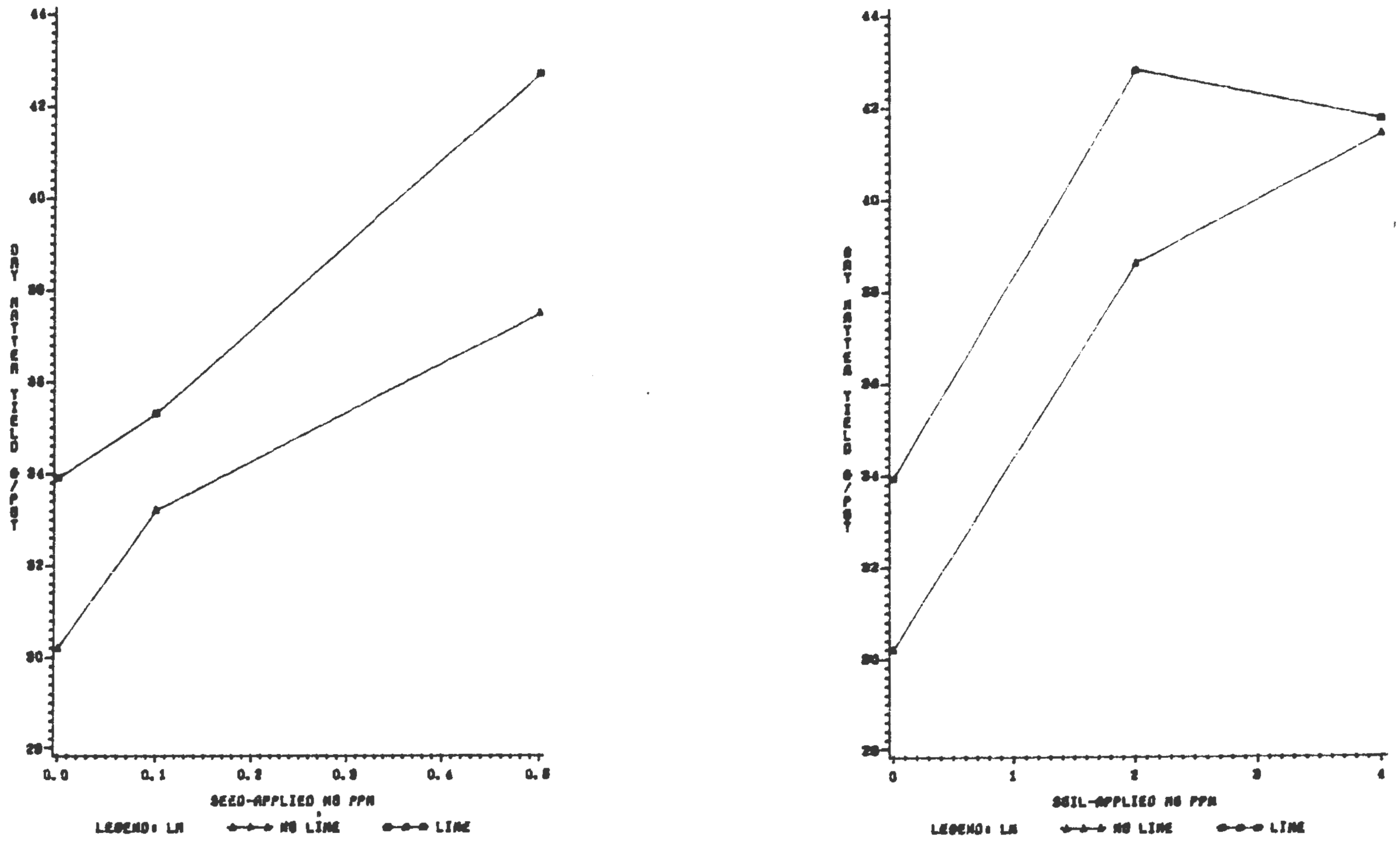


Fig. 1.1. Effects of seed-applied and soil-applied Mo, with or without lime, on dry matter yield of <u>Desmodium intortum</u> grown in the Wahiawa soil.

Mo, dry matter yields that would be obtained by using other rates of seed-applied or soil-applied Mo (e. g. 0.08 and 0.2 kg Mo/ha seed-applied or 0.5 and 1.0 kg Mo/ha soil-applied) were estimated from yield response curves (Fig. 1.2 and 1.3). According to the curves it can be estimated, for example, that yield of 33.8 g/pot would be obtained from seed-applied Mo at 0.08 kg Mo/ha or soil-applied Mo at 0.3 kg Mo/ha. The quantities of seed-applied or soil-applied Mo that would produce the same yield including the ratio of seed-applied to soil-applied Mo were shown in Table 1.3. These ratios were plotted against dry matter yields (Fig. 1.4). At the low levels of Mo, it was observed that **the** ratio of seed-applied to soil-applied Mo yield is low indicating that Mo applied is not adequate for plant need. At increasing rates of Mo applied, as the ratio increases, yield increased highly suggesting that at these Mo levels, seed coating is more efficient than soil application. At low rates of Mo applied, seed-applied Mo was adequate for plant need since it was concentrated around the seed, whereas soil-applied Mo was inadequate because Mo concentration around the seed was low. However, rate of yield increase leveled off as the ratios were increased above 0.28, suggesting that seed-applied Mo at high rate as compared to soil-applied Mo may be less efficient than seed-applied Mo at low rates. At high rates of Mo applied, soil-applied Mo became efficient since Mo concentration in soil was sufficient for plant need. The implication of this demonstration is that seed-applied Mo is more efficient than soil-applied Mo when low rates of Mo are used.

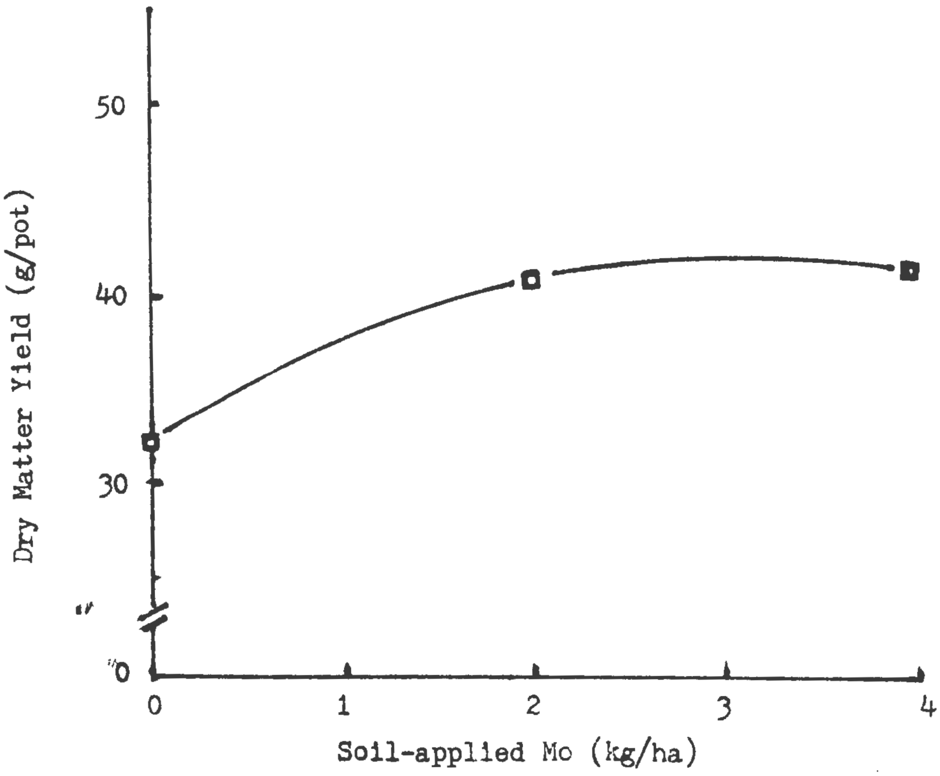


Fig.1.2. Effect of soil-applied Mo on dry matter yield of <u>Desmodium intortum</u> grown on the Wahiawa soil.

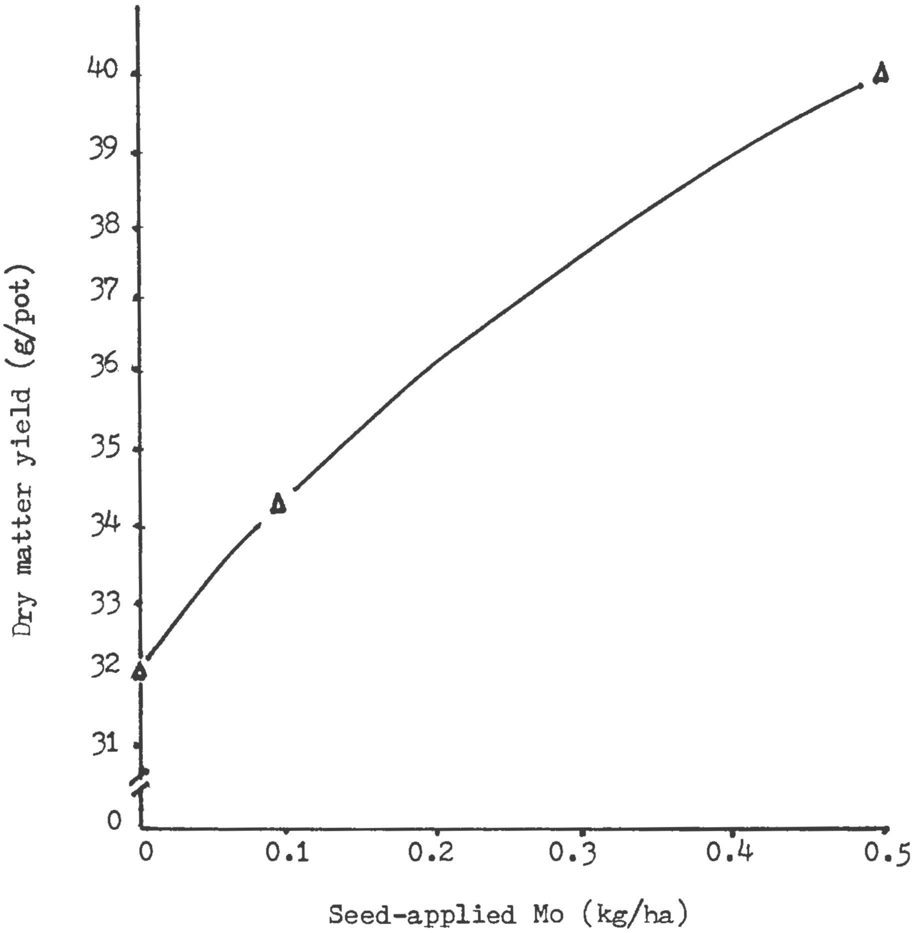


Fig. 1.3. Effect of seed-applied Mo on dry matter yield of Desmodium intortum grown in the Wahiawa soil.

Table 1.3.

Dry matter yield, estimation of Mo applied and ratio of seed-applied to soil-applied Mo obtained from Mo response curves.

	Mo fertilizer rate		Ratio of
Dry matter yield	Seed-applied	Soil-applied	Seed- to soil-applied Mo
g/pot	---------- kg/ha -----------		
33.0	0.04	0.17	0.235
33.5	0.06	0.23	0.261
34.0	0.09	0.34	0.265
34.5	0.11	0.40	0.275
35.0	0.14	0.50	0.280
35.5	0.17	0.60	0.283
36.0	0.20	0.70	0.286
36.5	0.24	0.80	0.300
37.0	0.27	0.95	0.284
37.5	0.31	1.00	0.310
38.0	0.35	1.15	0.304
38.5	0.385	1.30	0.296
39.0	0.42	1.45	0.289
39.5	0.46	1.55	0.297
40.0	0.495	1.75	0.283

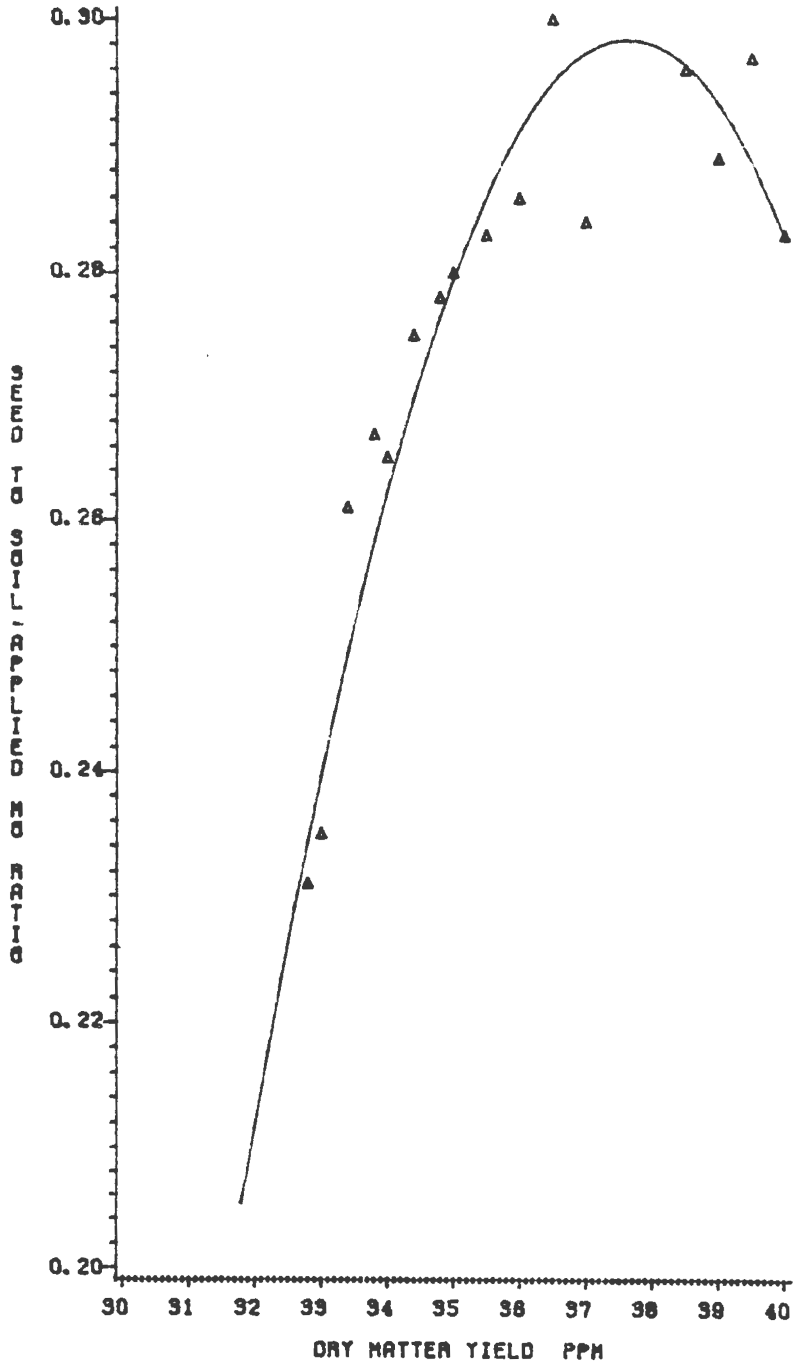


Fig. 1.4. Relationship between the ratio of seed-applied to soil-applied Mo and dry matter yield of Desmodium intortum grown in the Wahiawa soil.

Nodulation Response

Nodule Fresh Weight

There was no significant difference in nodule fresh weight of desmodium applied with or without Mo although soil-applied Mo had a tendency to increase nodule weight whereas seed-applied Mo tended to decrease nodule weight (Table 1.4). A tendency to decrease nodule weight with seed-applied Mo was due to the detrimental effect of Mo salt to rhizobia since it has been reported that Mo salts can kill rhizobia and reduce nodulation (Giddens, 1964; Burton and Curley, 1966). Lime and S tended to increase nodule fresh weight although the magnitude of increase was small. However, comparison of the control treatment (no Mo, no S and no lime) with lime treatment (no Mo and no S) indicated that lime increased fresh weight by 66.5% above the control treatment. When averaged over all Mo rates without S application, lime had a positive effect on nodule weight. Sulfur, when averaged over all Mo and lime rates, did not significantly increase nodule weight. However, S tended to increase nodule weight when no lime was applied and the effect of S disappeared when lime was applied. It is probable that lime could have at least partially eliminated the S deficiency since it has been indicated that sorbed sulfate not readily available to plants becomes increasingly available as the pH of the soil is increased (Elkins and Ensminger, 1971).

Nodule Dry Weight

Although there was no significant difference in nodule dry weight of desmodium applied with or without Mo, soil-applied Mo tended to increase nodule weight whereas seed-applied Mo tended to decrease nodule

Table 1.4

Effects of Mo, S, and lime on nodule fresh weight of *Desmodium intortum* grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	g/plant				
No Mo added	3.88	5.90	6.46	5.40	5.41 a
0.1 kg Mo/ha (seed applied)	4.82	5.09	5.55	5.94	5.35 a
0.5 kg Mo/ha (seed applied)	4.32	6.10	4.78	6.03	5.31 a
2.0 kg Mo/ha (soil applied)	5.75	5.66	7.10	5.15	5.92 a
4.0 kg Mo/ha (soil applied)	6.65	5.07	6.33	5.91	5.99 a
	5.08	5.56	6.04	5.69	
Mean	5.32 a		5.86 a		

	No S	S
Mean	5.56 a	5.63 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

weight (Table 1.5.). This trend was similar to the tendency observed with nodule fresh weight. It should be noted that nodule size of desmodium receiving Mo was larger than that of desmodium receiving no Mo. This result is likely due to the vigorous growth of plants receiving Mo application. A vigorously growing plant can supply a large amount of photosynthates to nodule such that nodule size is increased.

Nitrogenase Activity **Response** .

There was no significant difference in nitrogenase activity in desmodium applied with various rates of Mo, S, and lime (Table 1.6). Acetylene reduction among replicated samples was quite variable and although treatment differences were sizable, treatment effects were not statistically significant. Much of the variability was probably caused by variation in the degree of nodulation among plants. Despite a lack of significant difference, some definite trends were evident. Nitrogenase activity tended to be higher where Mo was applied, either by seed coating or soil application. Soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased nitrogenase activity by 37.0% and 31.8%, respectively, above that of the no-Mo treatment. Seed-applied Mo at 0.1 and 0.5 kg Mo/ha increased nitrogenase activity by 21.3% and 29.1%, respectively, above that of the no-Mo treatment. Seed-applied Mo at 0.5 kg Mo/ha appeared to be as effective as the two soil-applied Mo treatments although it tended to decrease nodulation as discussed before. Nitrogenase activity of plant from the S treatment was increased by 5.5% above that of plant from the no-S treatment. Nitrogenase activity of plant from the lime treatment was increased by 7.6% from the no-lime treatment. It was observed that

Table 1.5

Effects of Mo, S, and lime on nodule dry weight of <u>Desmodium intortum</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	---------- g/plant ----------				
No Mo added	0.65	1.17	1.17	0.90	0.97 a
0.1 kg Mo/ha (seed applied)	0.79	0.87	0.99	1.13	0.95 a
0.5 kg Mo/ha (seed applied)	0.73	1.04	0.88	1.07	0.93 a
2.0 kg Mo/ha (soil applied)	1.07	1.02	1.20	0.87	1.04 a
4.0 kg Mo/ha (soil applied)	1.19	1.15	0.81	0.98	1.03 a
	0.89	1.05	1.01	0.99	
Mean	0.97 a		1.00 a		

	No S	S
Mean	0.95 a	1.02 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.6

Effects of Mo, S, and lime on nitrogenase activity of <u>Desmodium intortum</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	µ mole C_2H_2/plant/hr				
No Mo added	3.70	4.86	5.72	5.72	4.84 a
0.1 kg Mo/ha (seed applied)	5.18	5.33	6.72	6.24	5.87 a
0.5 kg Mo/ha (seed applied)	5.14	7.70	4.47	7.70	6.25 a
2.0 kg Mo/ha (soil applied)	8.46	5.54	6.43	6.12	6.63 a
4.0 kg Mo/ha (soil applied)	5.66	6.22	6.90	6.74	6.38 a
	5.63	5.93	6.05	6.38	
Mean	5.78 a		6.22 a		

	No S	S
Mean	5.84 a	6.16 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

there was an interaction trend between Mo and S on nitrogenase activity. When no S was added, nitrogenase activity of plant receiving high rates of Mo (0.5 kg Mo/ha seed-applied and 4.0 kg Mo/ha soil-applied) was lower than that of plant receiving low rates of Mo counterpart (0.1 kg Mo/ha seed-applied and 2.0 kg Mo/ha soil-applied) (Fig. 1.5). However, when S at 50 kg S/ha was added, nitrogenase activity of plant receiving high rates of Mo was higher than that of plant receiving low rates of Mo counterpart. It may be speculated from these results that Mo/S ratio in nodule may have a significant effect on nitrogenase activity since both Mo and S are constituents of nitrogenase enzyme. It was shown in Table 1.13 and 1.18 that while S concentrations in plant top were similar among treatments receiving various rates of Mo, Mo concentration was significantly different. Molybdenum and S concentration in plant top may reflect Mo and S in nodule since it was observed that Mo concentration in plant top was correlated with Mo concentration in nodule (Fig. 1.6).

Since the nodule is the location where nitrogen fixation takes place, it is worthwhile to evaluate nodule efficiency by determination of specific acetylene-reducing activity of nitrogenase (acetylene reduced per unit weight of nodule). There was no significant difference in specific acetylene reduction of plant receiving different rates of Mo, S and/or lime (Table 1.7). However, Mo tended to increase specific acetylene reduction. Seed-applied Mo at 0.1 and 0.5 kg Mo/ha increased specific acetylene reduction by 15.7% and 24.1%, respectively, above the no-Mo treatment. Soil-applied Mo at 2.0 and 4.0 kg Mo/ha also increased acetylene reduction by 26.4% and 20.1%, respectively, above the no-Mo treatment. This result indicated that nodules fixed nitrogen efficiently when they received an adequate

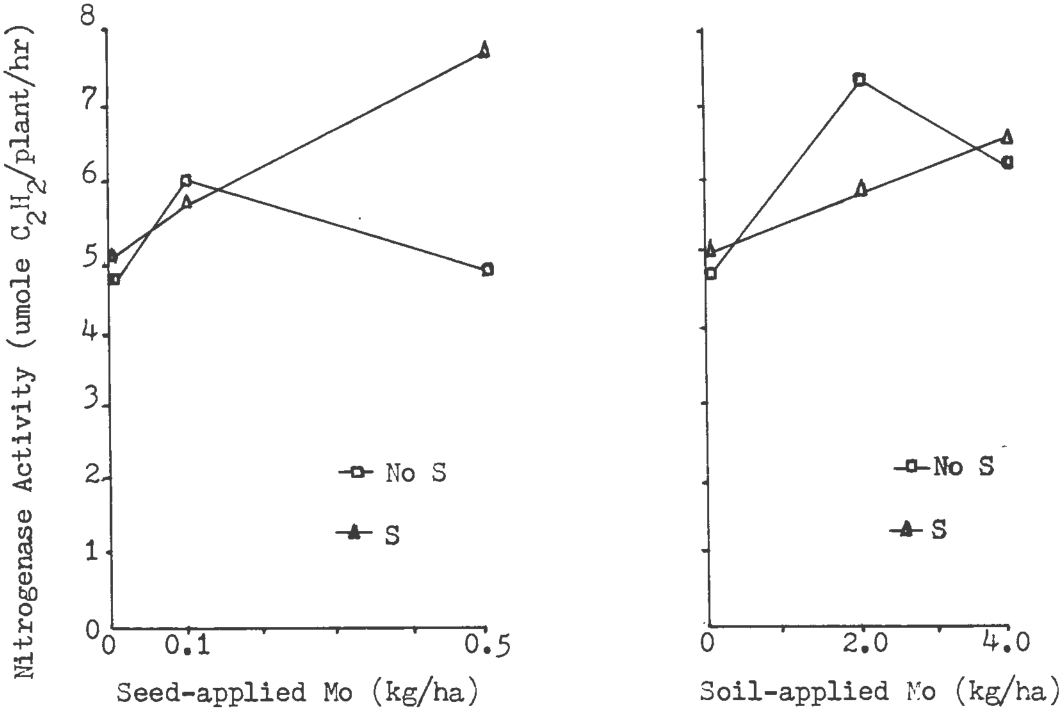


Fig. 1.5. Nitrogenase activity of Desmodium intortum grown in the Wahiawa soil.

Table 1.7

Effects of Mo, S, and lime on specific nitrogenase activity of Desmodium intortum grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	u mole C_2H_2/hr/g				
No Mo added	5.56	4.74	5.04	5.74	5.27 a
0.1 kg Mo/ha (seed applied)	7.03	5.61	6.15	5.59	6.10 a
0.5 kg Mo/ha (seed applied)	6.73	7.41	5.06	6.96	6.54 a
2.0 kg Mo/ha (soil applied)	8.11	5.60	5.50	7.42	6.66 a
4.0 kg Mo/ha (soil applied)	4.61	5.71	8.41	6.60	6.63 a
	6.41	5.81	6.03	6.46	
Mean	6.11 a		6.24 a		
	No S		S		
Mean	6.22 a		6.14 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

amount of Mo.

Influences of Mo, S and Lime on Some Chemical Compositions in Plant

Nitrogen

Soil-applied Mo at 2.0 and 4.0 kg Mo/ha significantly increased N concentration in plant (Table 1.8). Nitrogen concentrations of legume receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were 37.9% and 42.9%, respectively, higher than those of legume receiving no Mo. Soil-applied Mo at both above rates also significantly increased N concentration above seed-applied Mo at 0.1 kg Mo/ha. Nitrogen concentration of legume receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from soil-applied Mo at both rates, indicating that seed-applied Mo at 0.5 kg Mo/ha supplied an adequate amount of Mo to the requirement of legume for nitrogen fixation. Seed-applied Mo at 0.1 kg Mo/ha did not supply adequate amounts of Mo to plant needs since N concentration obtained from legume receiving seed-applied Mo at this rate was significantly lower than that of legume receiving seed-applied Mo at 0.5 kg Mo/ha or soil-applied Mo at both rates. Moreover, N concentration of plant receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly higher than that of plant receiving no Mo strongly suggesting that it is necessary to apply more than 0.1 kg Mo/ha if the seed coating method is to be used for pasture/forage legume production. However, seed-applied Mo at 0.5 kg Mo/ha appeared to be as effective as soil-applied Mo at 2.0 and 4.0 kg Mo/ha in increasing N concentration of plant.

Sulfur and lime had little effect on N concentration of plant. No

Table 1.8

Effects of Mo, S, and lime on N concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	%				
No Mo added	1.59	1.84	1.84	1.79	1.77 b
0.1 kg Mo/ha (seed applied)	2.28	1.71	2.00	1.67	1.92 b
0.5 kg Mo/ha (seed applied)	2.28	2.24	2.45	2.18	2.29 a
2.0 kg Mo/ha (soil applied)	2.60	2.36	2.48	2.32	2.44 a
4.0 kg Mo/ha (soil applied)	2.55	2.79	2.71	2.06	2.53 a
	2.26	2.19	2.30	2.00	
Mean	2.23 a		2.15 a		
	No S		S		
Mean	2.28 a		2.10 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

interaction among Mo, S and lime was observed in this experiment.

Phosphorus

Soil-applied Mo at 2.0 and 4.0 kg Mo/ha significantly decreased P concentration in plant top (Table 1.9). Phosphorus concentration in legume receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from that in legume receiving soil-applied Mo at both rates. Phosphorus concentration in legume receiving seed-applied Mo at 0.5 kg Mo/ha was also not significantly different from that in legume receiving seed-applied Mo at 0.1 kg Mo/ha. Phosphorus concentration in legume receiving seed-applied Mo at both rates was not significantly different from that in legume receiving no Mo. Lime also significantly decreased P concentration but S had little effect on P concentration in this experiment.

The reason that Mo or lime decreased P concentration may be due to a "dilution effect" of plant growth since Mo or lime significantly increased dry matter yield of desmodium (Table 1.2). It should be noted that P concentration in plant top was rather high compared to the critical P concentration of 0.21 to 0.24% for desmodium as suggested by Andrew and Robins (1969a).

Potassium

The K concentrations in desmodium receiving various rates of Mo was not significantly different from each other (Table 1.10). Sulfur and lime also had no effect on K concentration in plant top. However, it should be noted that K concentrations in plant were quite high compared to the critical K concentration of 0.7% to 0.8% as suggested by Andrew and Robins (1969b). Such high K concentrations in the plant top reflect an adequate

Table 1.9

Effects of Mo, S, and lime on P concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	0.43	0.40	0.41	0.37	0.40 a
0.1 kg Mo/ha (seed applied)	0.42	0.37	0.39	0.35	0.38 ab
0.5 kg Mo/ha (seed applied)	0.37	0.35	0.35	0.35	0.36 abc
2.0 kg Mo/ha (soil applied)	0.35	0.35	0.30	0.38	0.35 bc
4.0 kg Mo/ha (soil applied)	0.34	0.34	0.31	0.32	0.33 c
	0.38	0.36	0.35	0.35	
Mean	0.37 a		0.35 b		
	No S	S			
Mean	0.37 a	0.36 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.10

Effects of Mo, S, and lime on K concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	%				
No Mo added	2.22	2.26	2.19	2.17	2.21 a
0.1 kg Mo/ha (seed applied)	2.35	2.13	2.26	2.05	2.20 a
0.5 kg Mo/ha (seed applied)	2.27	2.28	2.27	2.30	2.28 a
2.0 kg Mo/ha (soil applied)	2.46	2.28	2.30	2.22	2.32 a
4.0 kg Mo/ha (soil applied)	2.25	2.40	2.22	2.20	2.27 a
	2.31	2.27	2.25	2.19	
Mean	2.29 a		2.22 a		

	No S	S
Mean	2.28 a	2.23 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

supply of K in the soil.

Calcium

Molybdenum application significantly decreased Ca concentration in plant top (Table 1.11). The Ca concentration in tissue of plant receiving seed-applied Mo at 0.5 kg Mo/ha, soil-applied Mo at 2.0 or 4.0 kg Mo/ha was significantly lower than that of plant receiving no Mo. The Ca concentration in plant receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly different from that in plant receiving no Mo or other rates of Mo. Sulfur had no significant effect on Ca concentration in plant. Lime significantly increased Ca concentration above the no-lime treatment. This effect is due to the fact that lime directly supplies Ca to the soil.

The influence of Mo in decreasing Ca concentration in plant top may be due to a "dilution effect" of growth since Mo significantly increased dry matter yield (Table 1.2). Molybdenum *per se* may not directly influence Ca concentration in plant top.

Magnesium

Magnesium concentration in the top of plant receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at **2.0 kg Mo/ha rate was** significantly lower than that of plant receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo (Table 1.12). Magnesium concentration in plant receiving seed-applied Mo at 0.1 kg Mo/ha was significantly lower than that in plant receiving no Mo. The decrease in Mg concentration in plant top as affected by Mo application may be due to a "dilution effect" of growth because yield of desmodium increased as rates of Mo applied increased (Table 1.2). Sulfur application did not significantly affect Mg concentration in plant

Table 1.11

Effects of Mo, S, and lime on Ca concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	1.25	1.11	1.32	1.24	1.23 a
0.1 kg Mo/ha (seed applied)	1.14	1.10	1.19	1.20	1.16 ab
0.5 kg Mo/ha (seed applied)	1.06	1.04	1.13	1.16	1.10 b
2.0 kg Mo/ha (soil applied)	1.06	1.06	1.09	1.16	1.09 b
4.0 kg Mo/ha (soil applied)	0.96	1.02	1.19	1.15	1.08 b
	1.09	1.07	1.18	1.18	
Mean	1.08 b		1.18 a		

	No S	S
Mean	1.14 a	1.13 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.12

Effects of Mo, S, and lime on Mg concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	------------------------- %		------------------------------------		
No Mo added	0.28	0.24	0.23	0.24	0.248 a
0.1 kg Mo/ha (seed applied)	0.24	0.25	0.22	0.22	0.233 b
0.5 kg Mo/ha (seed applied)	0.23	0.22	0.19	0.22	0.213 c
2.0 kg Mo/ha (soil applied)	0.22	0.24	0.20	0.21	0.219 c
4.0 kg Mo/ha (soil applied)	0.22	0.24	0.22	0.21	0.221 bc
	0.24	0.24	0.21	0.22	
Mean	0.24 a		0.22 b		

	No S	S
Mean	0.22 a	0.23 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

top. Liming significantly decreased Mg concentration in plant top. The effect of lime in decreasing Mg concentration is in accord with the result reported by Andrew and Johnson (1976). They indicated that in some tropical and temperate pasture legumes an increase in Ca resulted in a marked decrease in Mg concentration in plant top. They also noted that the change in Mg concentration in the plant top due to Ca application may not be due to a direct effect, but rather to the capacity of the plant to preserve cation balance.

Sulfur

Since it was observed that S concentrations in plant receiving various rates of Mo were similar, it can be concluded that Mo had no effect on S concentration in plant top (Table 1.13). Application of S at 50 kg S/ha did not increase S concentration in plant top. Sulfur concentration in plant remained the same even where S supply in soil was higher in the S treatment than in the no-S treatment. The reason for this non-increase is due to the adequate amount of available S in the Wahiawa soil; even plants receiving no S showed a higher S concentration than the critical S concentration of 0.15-0.18% as suggested by Andrew (1977). Bouma (1975), in a review of published information, mentioned that sulfate uptake is proportional to its supply until maximum growth is reached, with little or no further uptake at higher levels. Lime had little effect on S concentration in plant top.

Iron

Although Mo, S or lime have no significant effect on Fe concentration

Table 1.13

Effects of Mo, S, and lime on S concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	-%				
No Mo added	0.25	0.23	0.23	0.22	0.23 a
0.1 kg Mo/ha (seed applied)	0.22	0.25	0.24	0.22	0.23 a
0.5 kg Mo/ha (seed applied)	0.22	0.20	0.19	0.21	0.21 a
2.0 kg Mo/ha (soil applied)	0.23	0.24	0.20	0.20	0.22 a
4.0 kg Mo/ha (soil applied)	0.21	0.23	0.20	0.21	0.21 a
	0.23	0.23	0.21	0.21	
Mean	0.23 a		0.21 a		

	No S	S
Mean	0.22 a	0.22 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

in plant top, there was a slight effect of S on Fe concentration since S tended to decrease Fe concentration (Table 1.14).

Zinc

Molybdenum had no significant effect on Zn concentration in plant top, although there was a tendency that Zn concentration decreased when Mo was applied (Table 1.15). Sulfur also had little effect on Zn concentration in plant top but lime significantly decreased Zn concentration.

Manganese

Molybdenum and S had no significant effect on Mn concentration in plant top (Table 1.16). Lime significantly decreased Mn concentration. Manganese concentration in plant applied with lime was 12.5% lower than that in plant receiving no lime. The decrease in Mn concentration reflected liming effect in decreasing Mn availability in soil. This negative effect of lime on Mn availability in soil is one of the important beneficial effects of lime applied in acid tropical soils since many of these soils contain toxic levels of Mn.

Silicon

Molybdenum application significantly decreased Si concentration in plant top (Table 1.17). Seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 2.0 and 4.0 kg Mo/ha significantly decreased Si concentration. Silicon concentration of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from that of plant receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha. Silicon concentration in plant receiving seed-applied Mo at 0.1 kg Mo/ha was significantly lower than that in plant

Table 1.14

Effects of Mo, S, and lime on Fe concentration in the tops of *Desmodium intortum* grown in the Wahiawa soil

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	216.3	191.0	198.7	180.3	196.6 a
0.1 kg Mo/ha (seed applied)	193.3	177.7	211.0	178.7	190.2 a
0.5 kg Mo/ha (seed applied)	200.7	174.3	173.7	172.0	180.2 a
2.0 kg Mo/ha (soil applied)	172.7	197.0	168.0	177.7	178.8 a
4.0 kg Mo/ha (soil applied)	186.0	183.3	187.3	208.3	191.2 a
	193.8	184.7	187.7	183.2	
Mean	189.3 a		185.5 a		
	No S		S		
Mean	190.8 a		184.0 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.15

Effects of Mo, S, and lime on Zn concentration in the tops of <u>Desmodium</u> <u>intortum</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	ppm				
No Mo added	67.3	55.7	47.3	47.3	54.4 a
0.1 kg Mo/ha (seed applied)	62.3	54.0	49.7	42.3	52.1 a
0.5 kg Mo/ha (seed applied)	54.0	50.7	39.7	51.7	49.0 a
2.0 kg Mo/ha (soil applied)	54.3	55.7	38.7	43.3	48.0 a
4.0 kg Mo/ha (soil applied)	49.3	53.3	43.3	45.3	47.8 a
	57.4	53.9	43.7	46.0	
Mean	55.7 a		44.9 b		

	No S	S
Mean	50.6 a	50.0 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.16

Effects of Mo, S, and lime on Mn concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	309.7	278.7	277.0	313.3	294.7 a
0.1 kg Mo/ha (seed applied)	342.0	342.7	273.4	277.3	309.1 a
0.5 kg Mo/ha (seed applied)	325.7	264.0	244.3	287.5	280.4 a
2.0 kg Mo/ha (soil applied)	294.7	321.7	247.0	283.0	286.6 a
4.0 kg Mo/ha (soil applied)	305.7	293.0	296.0	237.0	282.9 a
	315.6	300.0	267.7	279.6	
Mean	307.8 a		273.7 b		

	No S	S
Mean	291.7 a	289.8 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.17

Effects of Mo, S, and lime on Si concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	0.45	0.41	0.30	0.29	0.36 a
0.1 kg Mo/ha (seed applied)	0.43	0.38	0.32	0.26	0.35 b
0.5 kg Mo/ha (seed applied)	0.35	0.31	0.25	0.29	0.30 c
2.0 kg Mo/ha (soil applied)	0.38	0.38	0.23	0.26	0.31 c
4.0 kg Mo/ha (soil applied)	0.34	0.37	0.27	0.27	0.31 c
	0.39	0.37	0.27	0.27	
Mean	0.38 a		0.27 b		
	No S	S			
Mean	0.33 a	0.32 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

receiving no Mo but significantly higher than that in **plant receiving** other levels of Mo. The reason that Si concentration decreased as Mo level increased was likely due to a "dilution effect" since dry matter yield increased. In this situation, desmodium responded to Mo which is the limiting element and dry matter production increased. Uptake of Si probably proceeds more slowly than dry matter accumulation, so Si concentration in plant decreases.

Sulfur had no effect on Si concentration in plant top but lime significantly decreased Si concentration.

Copper

The Cu concentration in desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was significantly higher than that in plant receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo (Table 1.18). The Cu concentration in plants receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly higher than that in plants receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha. Although Cu concentrations in plant receiving soil-applied Mo at both rates were higher than those in plant receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo, the differences among these concentrations were not significant. Sulfur slightly increased Cu concentration. Gupta and MacLeod (1975) who worked with alfalfa and red clover, also reported that Mo and S slightly increased Cu concentration in forage tissue. However, they noted that such increases did not appear to be consistent at all levels of Mo and S applied.

Lime significantly increased Cu concentration. This result was not expected since Cu in soil is more available at a lower pH. The

Table 1.18

Effects of Mo, S, and lime on Cu concentration in the tops of Desmodium intortum grown in the Wahiawa soil.

Mo treatment	No lime No S	No lime S	Lime No S	Lime S	Mean
	ppm				
No Mo added	7.0	8.7	10.3	10.3	9.1 b
0.1 kg Mo/ha (seed applied)	9.7	8.0	10.0	9.7	9.3 b
0.5 kg Mo/ha (seed applied)	10.0	10.3	11.3	12.7	11.1 a
2.0 kg Mo/ha (soil applied)	10.0	9.0	10.7	11.3	10.3 ab
4.0 kg Mo/ha (soil applied)	9.7	10.3	10.3	10.7	10.3 ab
	9.3	9.3	10.5	10.9	
Mean	9.3 b		10.7 a		
	No S		S		
Mean	9.9 a		10.1 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

possible reason that lime increased Cu concentration in this experiment is that soil pH raised to 6.0 may not be high enough to significantly decrease Cu availability. Raising soil pH to 6.0 provides a favorable condition for plant growth and, as a consequence, may increase Cu absorption. Increased Cu absorbtion influenced by the vigorous growth of plant may also be the reason that Mo increased Cu concentration.

Influences of Mo, S and Lime on Mo Concentrations **in Plant and Soil**

Molybdenum Concentration in Plant Top

Molybdenum fertilizer significantly increased Mo concentration in plant top (Table 1.19). The Mo concentration in plant receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were significantly higher than that in plant receiving no Mo or seed-applied Mo at 0.1 and 0.5 kg Mo/ha. The Mo concentration in plant receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly different from that in plant receiving no Mo. Although Mo concentration in plant receiving seed-applied Mo at 0.5 kg Mo/ha was 153.1% and 166.7% higher than that in plant receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo, respectively, the differences were not significant. It should be noted that the recovery of Mo, measured by plant Mo concentration, from seed-applied Mo was higher than that from soil-applied Mo. For example, Mo concentration in plant from soil-applied Mo at 4.0 kg Mo/ha treatment was four times higher than that in plant from seed-applied Mo at 0.5 kg Mo/ha. However, the quantity of Mo added to soil by soil-applied Mo at 4.0 kg Mo/ha was eight times higher than that added to soil by seed-applied at 0.5 kg Mo/ha. The results indicated that addition of Mo by seed

Table 1.19

Effects of Mo, S, and lime on Mo concentration in the tops of *Desmodium intortum* grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	0.06	0.10	0.09	0.12	0.09 c
0.1 kg Mo/ha (seed applied)	0.07	0.09	0.14	0.09	0.10 c
0.5 kg Mo/ha (seed applied)	0.31	0.21	0.26	0.22	0.25 c
2.0 kg Mo/ha (soil applied)	0.39	0.76	0.33	0.40	0.47 b
4.0 kg Mo/ha (soil applied)	1.14	1.00	0.92	1.22	1.07 a
	0.39	0.43	0.35	0.41	
Mean	0.41 a		0.38 a		

	No S	S
Mean	0.37 a	0.42 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

coating is more effective in increasing Mo absorption of plant than by soil-application. The reason for this is that although the amount of Mo added by seed coating was low compared to soil application, nevertheless the Mo was concentrated around the seeds. Moreover, although the amount of Mo added by soil application was higher than by seed coating, much of this amount was adsorbed strongly by oxides and hydrous oxides of Fe and Al found in large amounts in the Wahiawa soil.

Where is your evidence of this?

Sulfur and lime had no effect on Mo concentration in plant top. Sulfur fertilizer at the 50 kg S/ha rate used in this experiment might not enough to decrease Mo concentration in plant top as regard to the potential toxic effect of high Mo application rates to livestock. However, the potential toxic effect of Mo ~~was likely~~ probably did not occurred since desmodium tops contained small amounts of Mo. Even the plant receiving Mo at 4.0 kg Mo/ha contained only 1.07 ppm Mo while Mo concentration in plant top of alfalfa and red clover receiving Mo at the same 4.0 kg Mo/ha rate contained 41.14 and 78.24 ppm Mo, respectively (Gupta and MacLeod, 1975). Such low concentration in desmodium top may be due to low accumulation of Mo in the top or to the requirement of Mo being lower in desmodium than in alfalfa and red clover. High Mo adsorption in the Wahiawa soil ~~is likely~~ probably reduced ~~affect~~ Mo concentration in plant. Even though Mo fertilizer was applied at the ~~at~~ 4.0 kg Mo/ha unusually high rate of ~~rate was used~~, much of this ~~amount~~ apparently was adsorbed by soil and only a relatively small part of it ~~was~~ remained available to the plant.

Molybdenum Concentration in Nodule

Molybdenum fertilizer significantly increased Mo concentration in nodule of desmodium (Table 1.20). Molybdenum concentration ~~of~~ in plants receiving no Mo was significantly lower than that of plant receiving seed-applied Mo. The Mo concentration in nodule of plant receiving seed-applied Mo at 0.5 kg Mo/ha was significantly higher than that of plant receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo. The Mo concentration in nodule of plant receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than that of plant receiving other levels of Mo applied. The Mo concentration in nodule of plant receiving soil-applied Mo at 2.0 kg Mo/ha was not significantly higher than in that of plant receiving seed-applied Mo at 0.5 kg Mo/ha, suggesting that plant receiving Mo at both rates accumulated similar amounts of Mo in nodule.

here on I haven't marked the plant when it should be plants

Sulfur had little effect on Mo concentration in nodule since Mo concentration in nodule of plant receiving 50 kg S/ha was slightly lower than that of plant receiving no S. Sulfur applied at this rate may not high enough to decrease Mo concentration in plant as it has been reported by Gupta and MacLeod (1975) who worked with alfalfa and red clover.

Liming did not significantly increased Mo concentration in nodule when concentration was averaged over all Mo and S treatments (Table 1.20). However, when no Mo and S was applied, lime increased Mo concentration in nodule 100.0% above the no-lime treatment.

The Mo concentration in nodule was higher than that in plant top. Higher Mo concentrations in nodules than in other plant parts has been observed in French bean (<u>Phaseolus vulgaris</u> L.) (Franco and Munns, 1981), alfalfa and clover (Jensen, 1946).

Table 1.20

Effect* of Mo, S, and lime on Mo concentration in nodules of <u>Desmodium intortum</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	ppm				
No Mo added	1.05	1.78	2.10	1.08	1.50 d
0.1 kg Mo/ha (seed applied)	2.07	2.43	3.50	2.78	2.70 c
0.5 kg Mo/ha (seed applied)	5.41	4.45	5.89	4.96	5.18 b
2.0 kg Mo/ha (soil applied)	6.16	6.51	4.58	5.78	5.76 b
4.0 kg Mo/ha (soil applied)	7.79	7.98	8.10	6.85	7.68 a
	4.50	4.63	4.83	4.29	
Mean	4.57 a		4.56 a		

	No S	S
Mean	4.67 a	4.46 a

---

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Extractable Mo in Soil

Extractable Mo in soil with soil-applied Mo at 4.0 kg Mo/ha was significantly higher than that in soil with other rates of Mo (Table 1.21). Extractable Mo in soil treated with soil-applied Mo at 2.0 kg Mo/ha and seed-applied Mo at both 0.1 and 0.5 kg Mo/ha rates were not significantly different. Extractable Mo obtained from the no-Mo treatment was significantly lower than that obtained from the 0.1 kg Mo/ha seed-applied Mo and 2.0 kg Mo/ha soil-applied Mo treatments but was not significantly lower than that obtained from the 0.5 kg Mo/ha seed-applied Mo treatment. The reason that extractable Mo obtained from 0.5 kg Mo/ha seed-applied Mo treatment was not significantly different from that obtained from 0.1 kg Mo/ha seed-applied or no-Mo treatment may be due to the high Mo concentration in nodule and the top of plants receiving seed-applied Mo at 0.5 kg Mo/ha. Desmodium receiving seed-applied Mo at 0.5 kg Mo/ha absorbed more Mo from soil than plants receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo (Table 1.19 and 1.20).

Sulfur application had little effect on extractable Mo in soil after harvesting. When averaged over all Mo and S treatments, lime had no effect on extractable Mo. However, when Mo and S were not applied, lime increased extractable Mo 14.0% above the no-lime treatment. This effect suggests that lime had some influence on the Mo status in soil.

It should be noted that extractable Mo was not really Mo that is readily available to plant. The amount of Mo that the plant absorbed at one time should be less than the amount extracted by acid ammonium oxalate solution. This happens because this extracting solution also dissolves the fraction of Fe and Al oxides and in doing so it extracts Mo in

Table 1.21

Effects of Mo, S, and lime on extractable Mo in soil after harvesting of <u>Desmodium</u> <u>intortum</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	ppm				
No Mo added	0.37	0.30	0.43	0.26	0.34 c
0.1 kg Mo/ha (seed applied)	0.46	0.50	0.41	0.47	0.46 b
0.5 kg Mo/ha (seed applied)	0.47	0.45	0.36	0.48	0.44 bc
2.0 kg Mo/ha (soil applied)	0.64	0.52	0.56	0.35	0.52 b
4.0 kg Mo/ha (soil applied)	0.69	0.73	0.61	0.67	0.68 a
	0.53	0.50	0.48	0.44	
Mean	0.51 a		0.46 a		

	No S	S
Mean	0.50 a	0.47 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

combination with the oxides. Molybdenum held at the Fe and Al oxide surfaces is not readily available to plant. Smith and Leeper (1969) suggested that the Mo bound to Fe and Al oxides could be thought of as a semi-permanent form. Although extractable Mo may not represent readily available Mo, it is still useful in evaluating Mo status of soil (e.g. the capacity of soil to supply Mo) for plant need since the amount of Mo in soil solution is so small that it is very difficult to detect by the conventional thiocyanate method.

Relationships between Mo Concentrations in Plant and Soil

Relationship between Mo Concentration in Plant Top and Mo Concentration in Nodule

There was a highly significant correlation between Mo concentration in plant top and Mo concentration in nodule (Fig. 1.6). According to the curve, plant Mo concentration at very low nodule Mo concentration (e.g. less than 1.0 ppm) was higher than that at nodule Mo concentration in the range of 1.0 to 3.0 ppm, suggesting that Mo in plant top was transferred to the nodule. A similar translocation pattern was also observed in soybean (Ishizuka, 1982). During the early growing period, the Mo in soybean cotyledons was at first translocated mainly to stems, and then to roots. With growth of the nodules, the Mo was intensively transferred to the nodules.

As the Mo concentration in nodule increased, the Mo concentration in the desmodium top increased exponentially (Fig. 1.6), suggesting that when Mo concentration in nodule was adequate for nitrogenase activity, excess Mo was transferred to the plant top. Plant top then

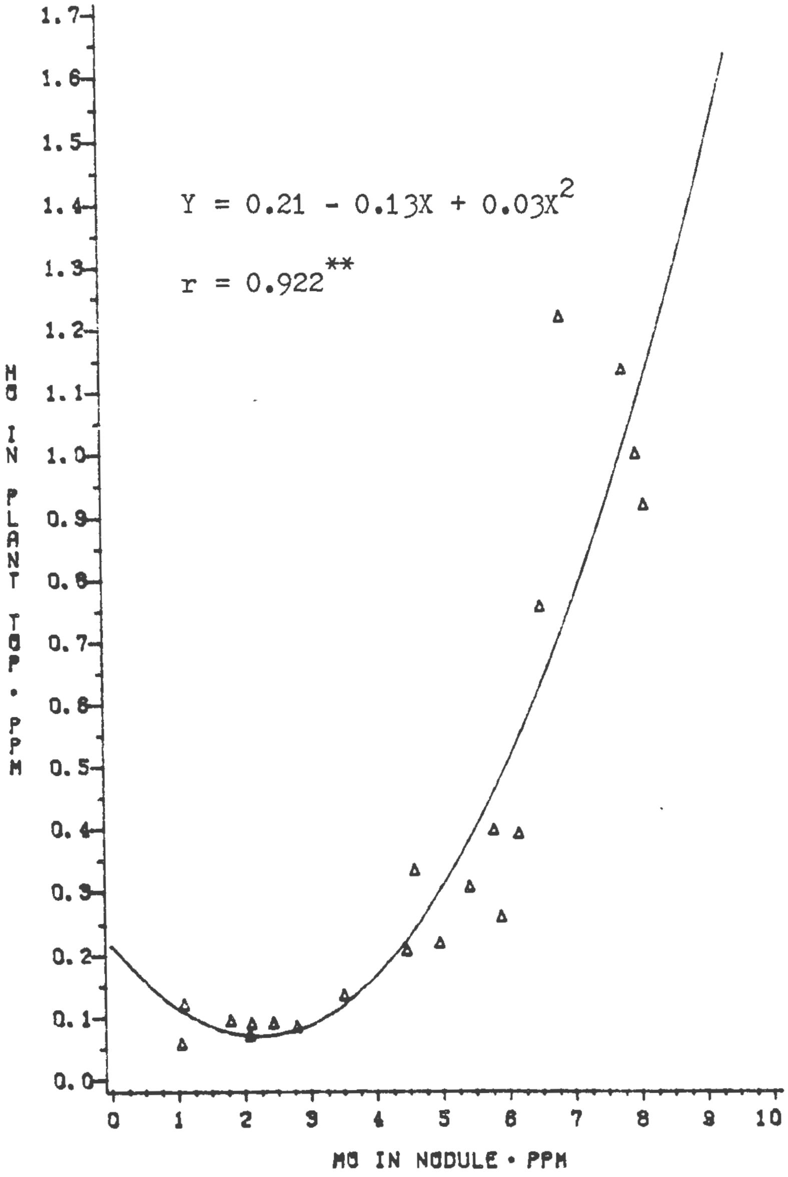


Fig. 1.6. Relationship between Mo concentration in plant top and Mo concentration in nodule of Desmodium intortum grown in the Wahiawa soil.

was the main sink for Mo.

Relationship between Mo Concentration in Plant Top and Extractable Mo in Soil

There was a highly significant correlation between Mo concentration in plant top and extractable Mo in the Wahiawa soil after harvesting (Fig. 1.7). Molybdenum concentration in the top increased as extractable Mo increased. This result indicated that Mo accumulation in the top depend on Mo concentration in soil which may be considered as external Mo. Although extractable Mo is not Mo in soil solution as discussed before, it can be useful as one of the criteria to assess Mo response in plant since it is correlated with Mo concentration in plant top.

Relationship between Mo Concentration in Nodule and Extractable Mo in Soil

There was a highly significant correlation between Mo concentration in nodule and extractable Mo in soil (Fig. 1.8). Molybdenum concentration in nodule linearly increased as extractable Mo increased suggesting that Mo concentration in nodule also depended on Mo concentration in soil. Franco and Munns (1981) who worked with <u>Phaseolus vulgaris</u> concluded that root Mo evidently varied the most with external Mo, whereas nodule Mo varied the least.

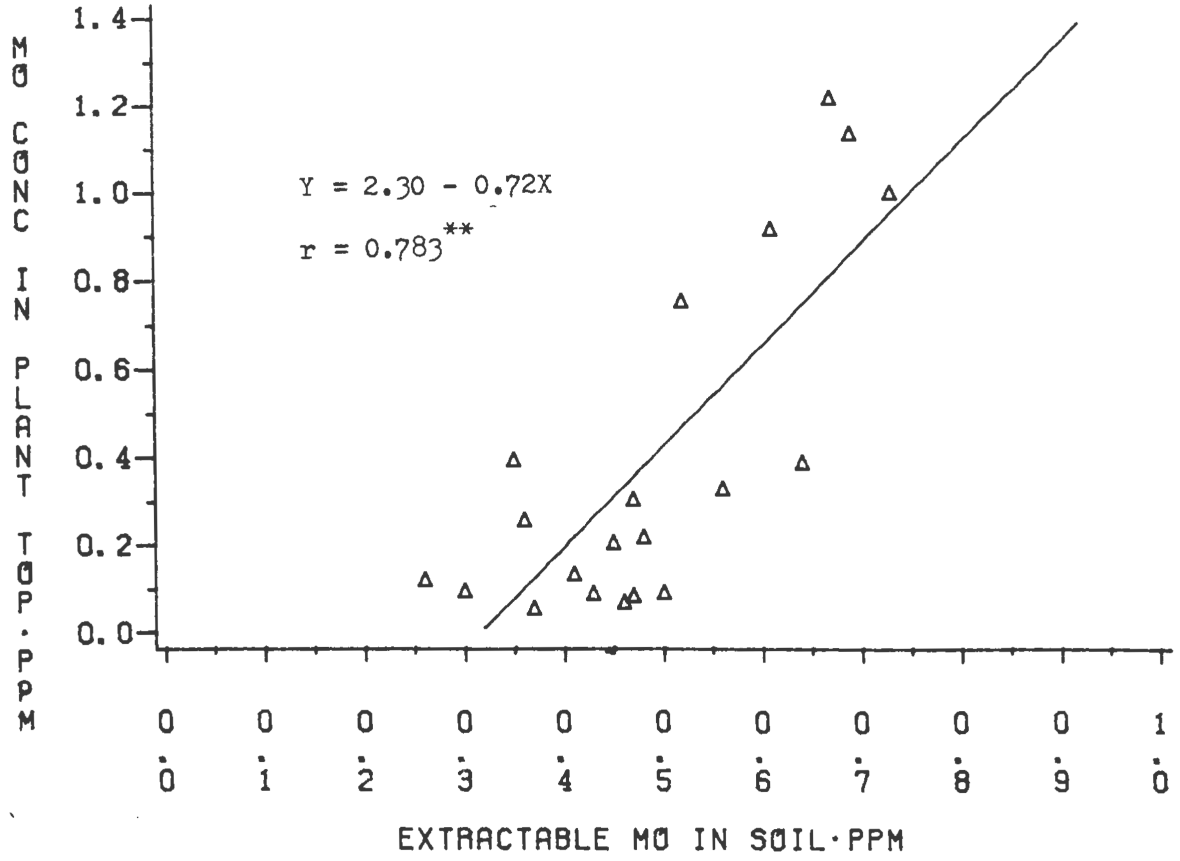


Fig. 1.7. Relationship between Mo concentration in the top of *Desmodium intortum* and extractable Mo in the Wahiawa soil.

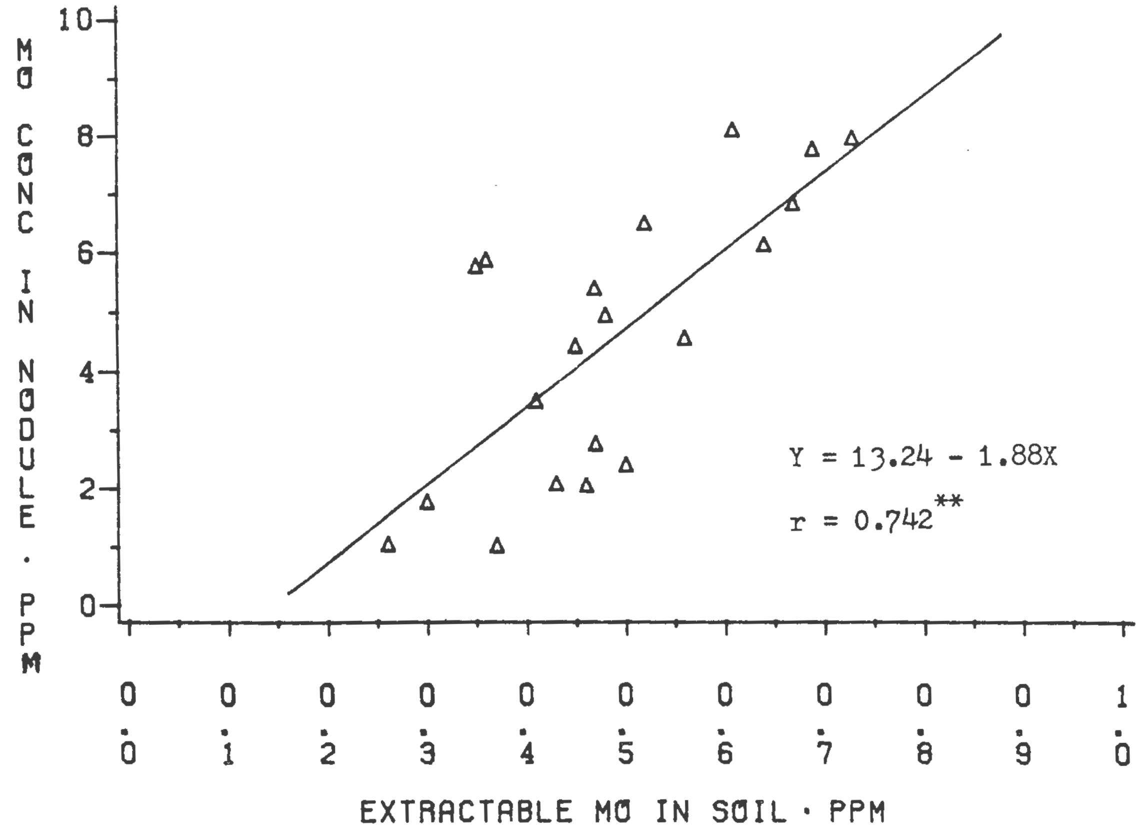


Fig. 1.8. Relationship between Mo concentration in nodule of *Desmodium intortum* and extractable Mo in the Wahiawa soil.

Relationship between Dry Matter Yield and Mo Concentrations

Relationship between Dry Matter Yield and Mo Concentration in Nodule

There was a **highly significant correlation between dry matter yield and Mo concentration in nodule of desmodium (Fig. 1.9).** The yield increased linearly as the Mo concentration in nodule increased. The results suggest that Mo concentration in nodule can be used as one of the criteria for determination of plant response to Mo.

Dry matter yield was also expressed as relative yield (percentage of the maximum yield). Expressing the yield data as relative yield eliminated variability due to growth conditions (e. g. heat, available soil moisture, etc.) (Maynard et al., 1983). Relative yields were plotted against Mo concentrations in nodule. There was a highly significant correlation between relative yield and Mo concentration in nodule (Fig. 1.10). Critical value was taken to be the Mo concentration associated with the point where relative yield was 20% less than the maximum yield since it was observed that the regression line was considerably flat. Using critical value of Mo concentration associated with the point where relative yield was 10% less than the maximum as it was suggested by Spencer and Freney (1980) for S is not possible.

be careful only certain types of variation are removed this way.

Critical value of Mo concentration in nodule required to obtain optimum yield (yield of 80% maximum yield) was 3.5 ppm. Franco and Munns (1981) suggested that the critical Mo concentration in nodule of *Phaseolus vulgaris* L. was between 3 to 5 ppm.

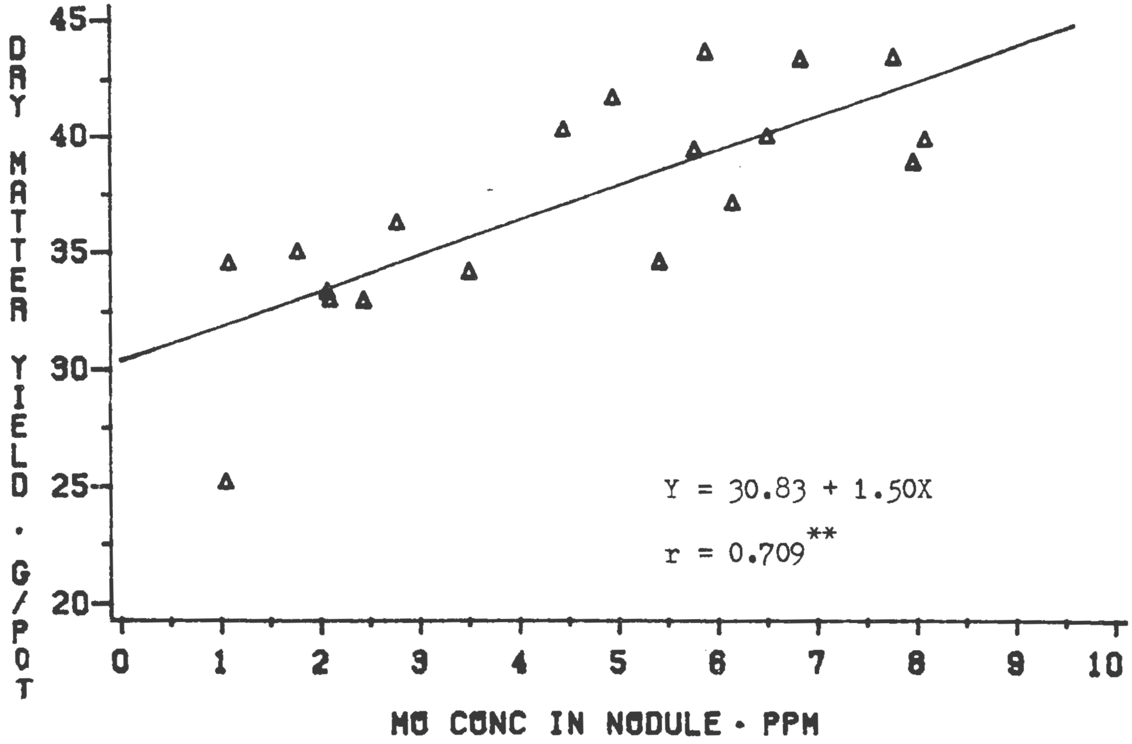


Fig. 1.9. Relationship between dry matter yield and Mo concentration in nodule of *Desmodium intortum* grown in the Wahiawa soil.

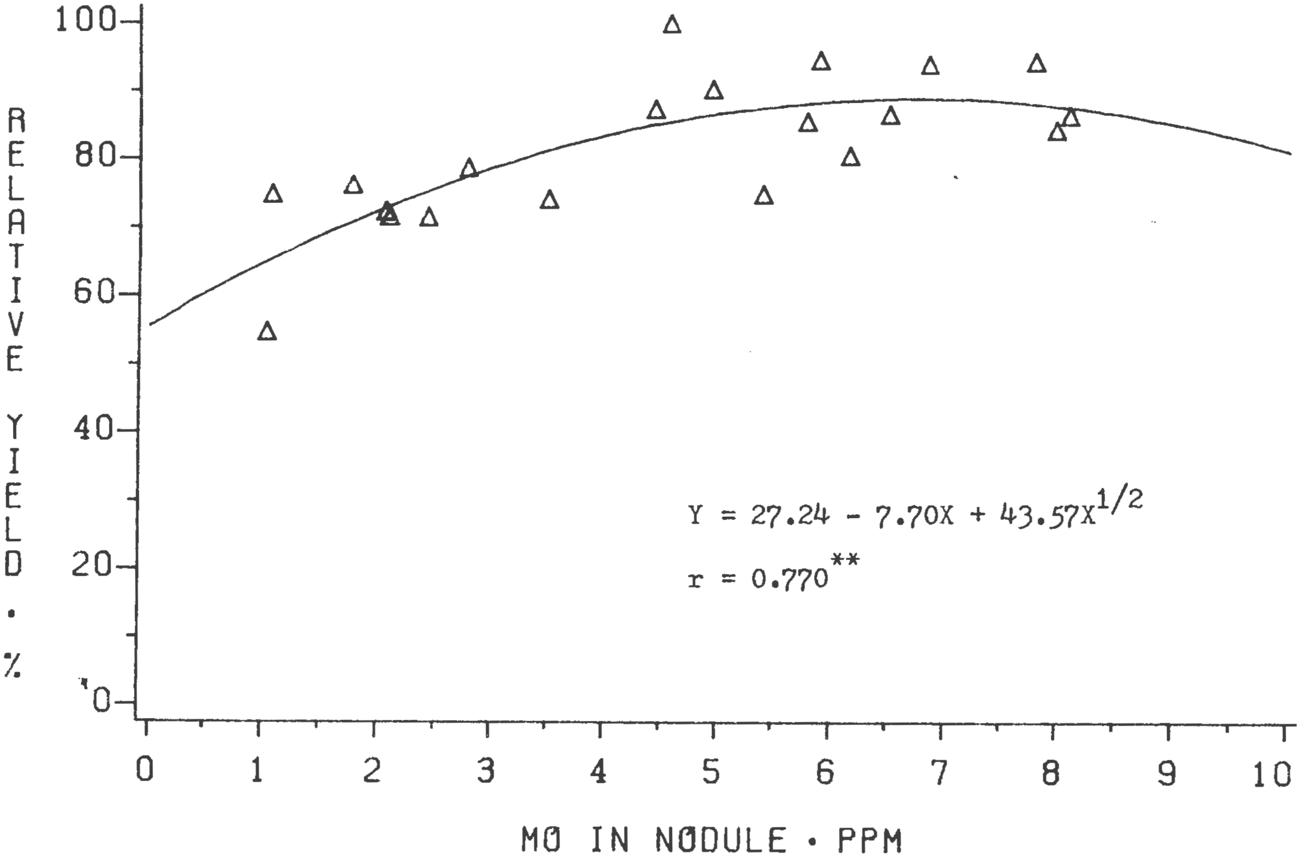


Fig. 1.10. Relationship between relative yield and Mo concentration in nodule of <u>Desmodium intortum</u> grown in the Wahiawa soil.

Relationship between Dry Matter Yield and Mo Concentration in Plant Top

There was a highly significant correlation between dry matter yield and Mo concentration in plant top (Fig. 1.11). Yield of desmodium increased as Mo concentration in the top increased. However, when Mo concentration was higher than 0.9 ppm yield tended to decline, suggesting that high level of Mo would have a negative effect on plant growth. There was also a highly significant correlation between relative yield and Mo concentration in the top (Fig. 1.12). The critical Mo concentration in plant top required to obtain an optimum yield was 0.2 ppm.

Relationship between Dry Matter Yield and Extractable Mo in Soil

There was a significant correlation between dry matter yield and extractable Mo in soil (Fig. 1.13). Dry matter yield of desmodium increased as extractable Mo increased with a correlation coefficient of 0.452. This coefficient is smaller than that obtained from the relationship between yield and Mo concentration in plant top or in nodule. There was a significant correlation between relative yield and extractable Mo in soil (Fig. 1.14). The extractable Mo in soil required to obtain an optimum yield of desmodium was 0.47 ppm.

In considering the relative importance of the correlations between relative yield and Mo concentration in nodule, Mo concentration in plant top, and extractable Mo, the coefficients of determination and the degree of scatter observed in Fig. 1.10, 1.12, and 1.14 indicated that the Mo concentration in nodule was the most highly correlated index with relative yield.

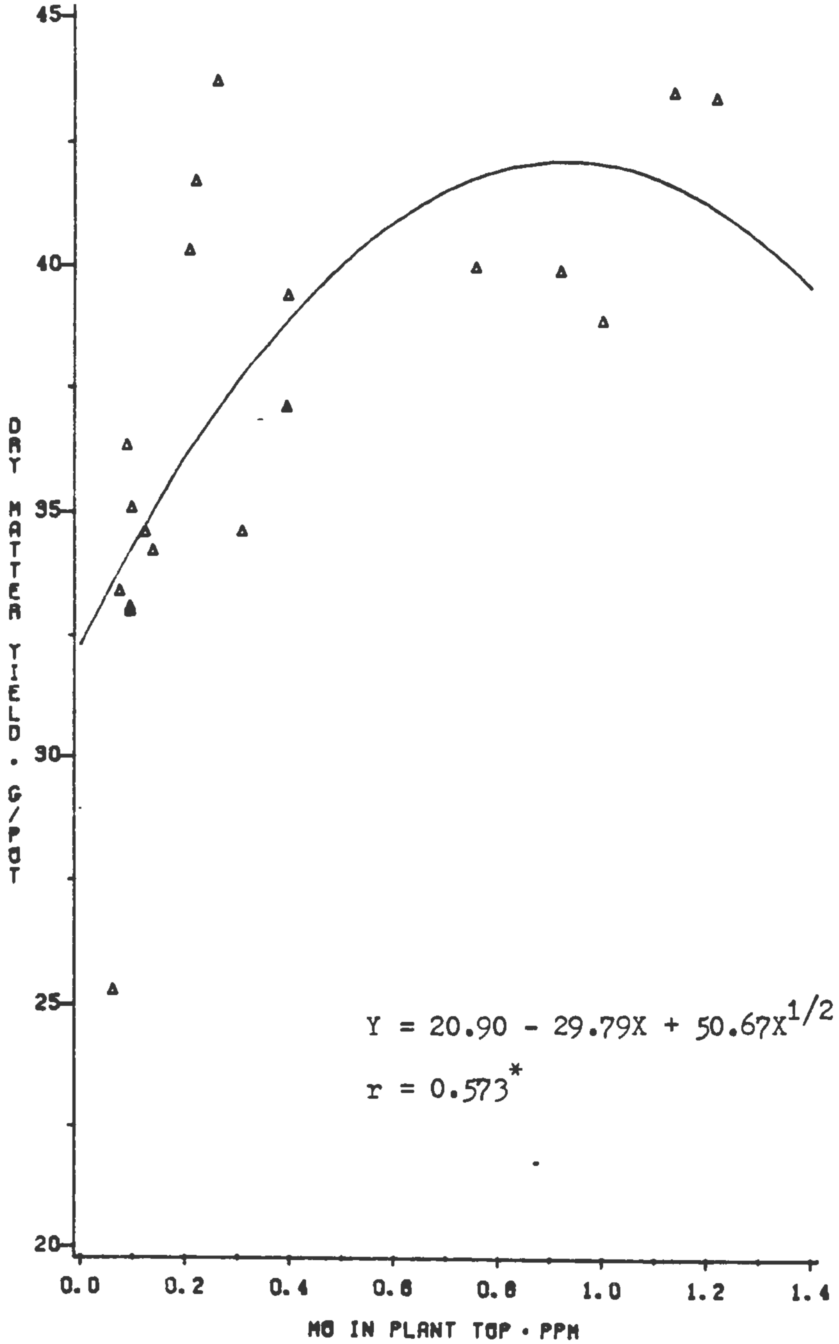


Fig. 1.11. Relationship between dry matter yield and Mo concentration in the top of Desmodium intortum grown in the Wahiawa soil.

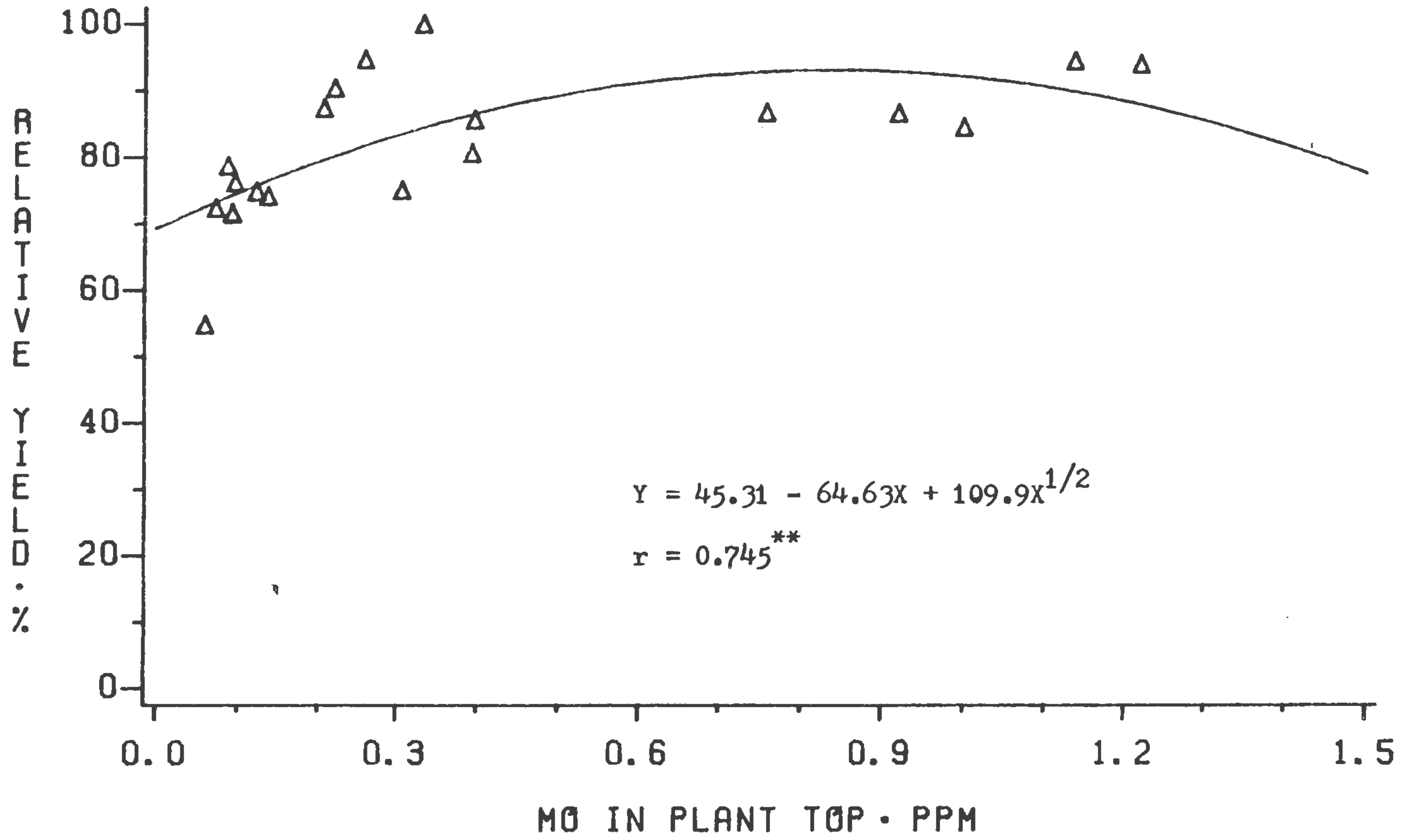


Fig. 1.12. Relationship between relative yield and Mo concentration in the top of <u>Desmodium intortum</u> grown in the Wahiawa soil.

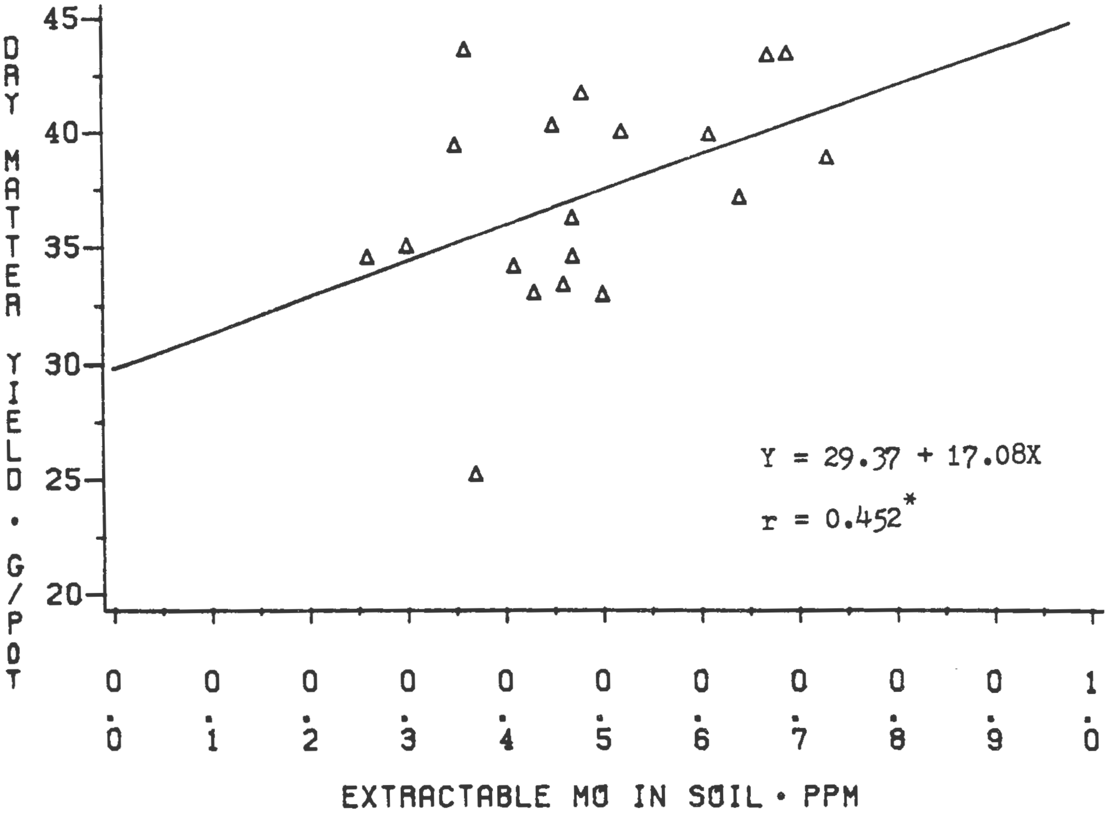


Fig. 1.13. Relationship between dry matter yield of *Desmodium intortum* and extractable Mo in the Wahiawa soil.

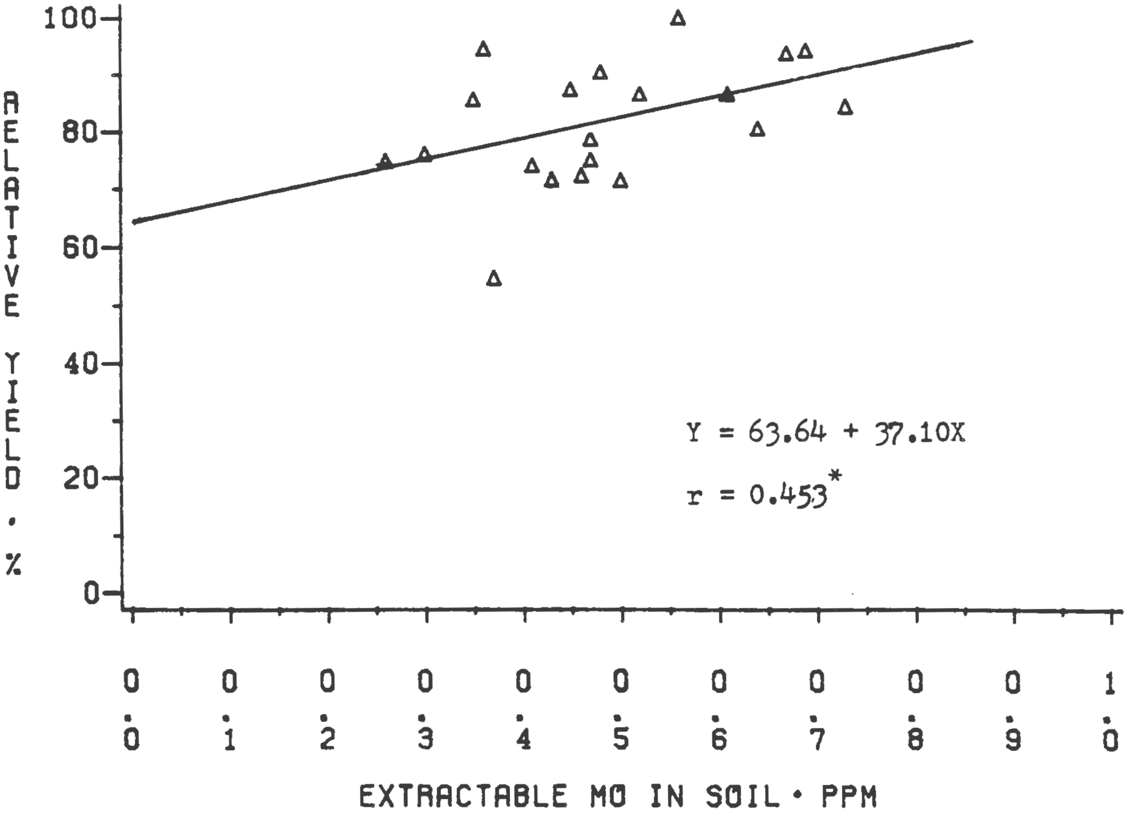


Fig. 1.14. Relationship between relative yield of Desmodium intortum and extractable Mo in the Wahiawa soil.

Relationship between N and Mo Concentrations

Relationship between Plant N Concentration and Mo Concentration in Nodule

There was a highly significant correlation between N concentration in plant top and Mo concentration in nodule (Fig. 1.15). Nitrogen concentration increased as Mo concentration increased, indicating that N concentration in plant top depends on Mo concentration in nodule. Nitrogen fixation in nodules is believed to increase as Mo concentration increases since Mo is required for nitrogenase enzyme responsible for nitrogen fixation. As nitrogen fixation increases, N concentration in plant top should increase.

Relationship between Plant N and Mo concentrations

There was a highly significant correlation between N and Mo concentrations in plant top. Nitrogen concentration increased as Mo concentration in the top increased (Fig. 1.16). However, when Mo concentration was higher than 0.7 ppm N concentration decreased, suggesting that high concentration of Mo is likely to have a negative effect on nitrogen fixation. The reason that the correlation coefficient between N and Mo concentrations in the top was lower than the coefficient between plant N concentration and Mo concentration in nodule is due to the fact that Mo in nodule directly influence N concentration through its role in nitrogen fixation in nodule. In legumes Mo plays a more critical role in nodule than in plant top.

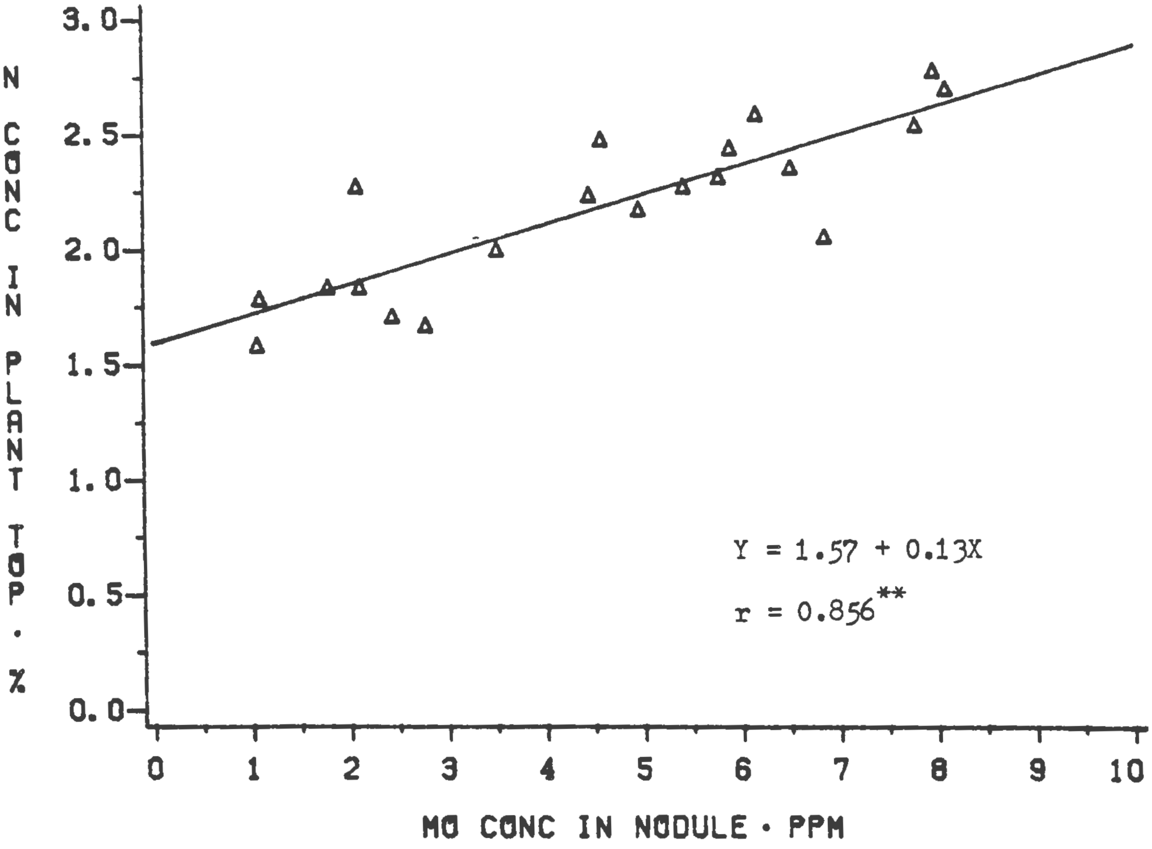


Fig. 1.15. Relationship between N concentration in plant top and Mo concentration in nodule of Desmodium intortum grown in the Wahiawa soil.

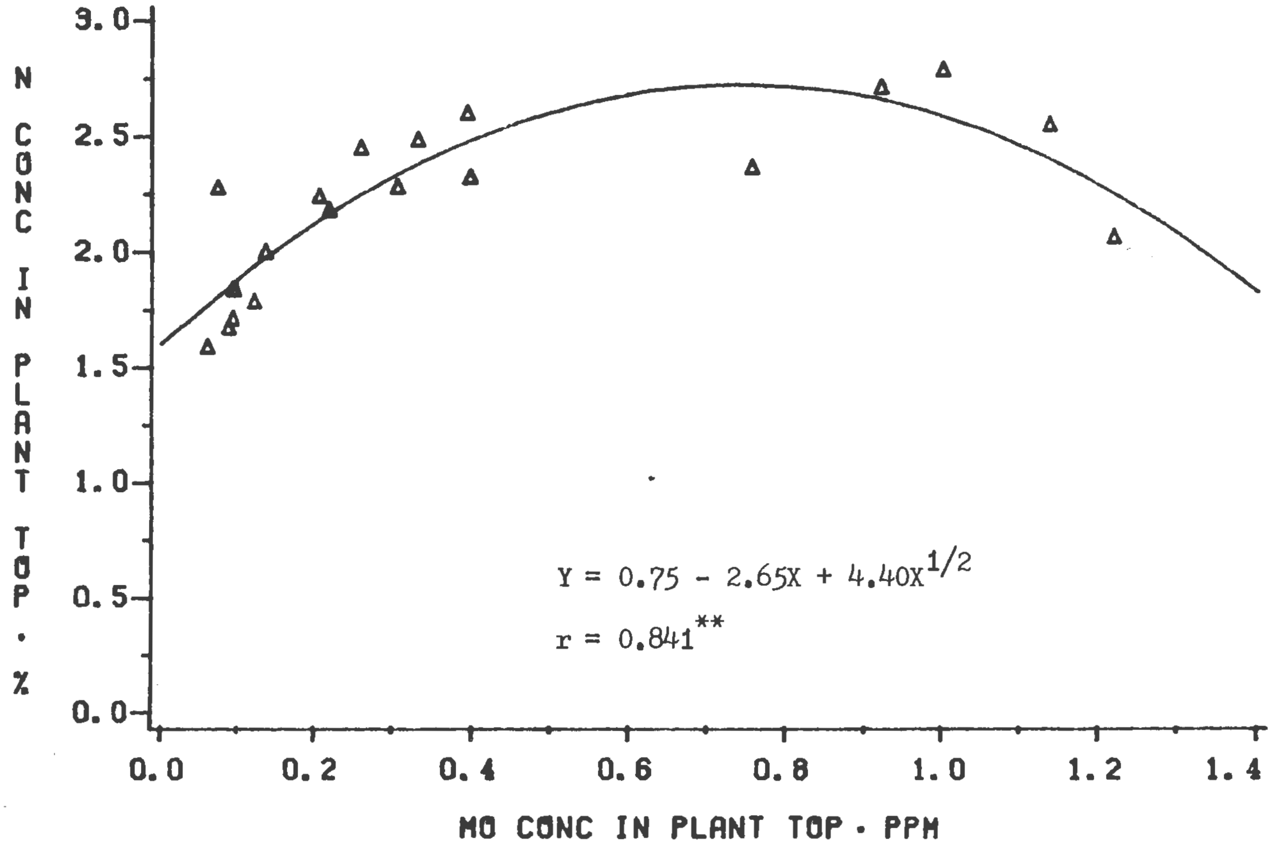


Fig. 1.16. Relationship between N and Mo concentrations in the top of <u>Desmodium</u> <u>intortum</u> grown in the Wahiawa soil.

Relationship between Plant N Concentration and Extractable Mo in Soil

There was a highly significant correlation between N concentration in plant top and extractable Mo in soil (Fig. 1.17). Nitrogen concentration increased as extractable Mo increased. The correlation coefficient of this relationship (0.620) was lower than that of the relationship between N concentration and Mo concentration in plant top (0.841) and that of the relationship between N concentration and Mo concentration in nodule (0.856).

In considering the correlation coefficients of the relationships between N concentration and Mo concentration in nodule, Mo concentration in plant top, and extractable Mo, it was indicated that the Mo concentration in nodule was the most highly correlated index to N concentration in plant. The high correlation between Mo concentration in nodule and N concentration was due to the nitrogen fixation system that requires Mo locates in the nodule.

Relationship between Nitrogenase Activity and Mo Concentrations

Relationship between Nitrogenase Activity and Mo Concentration in Nodule

There was no significant correlation between nitrogenase activity and Mo concentration in nodule. However, at low concentration of Mo in nodule (i.e. from 1 to 4 ppm), nitrogenase activity increased as Mo concentration in nodule increased (Fig. 1.18). This relationship

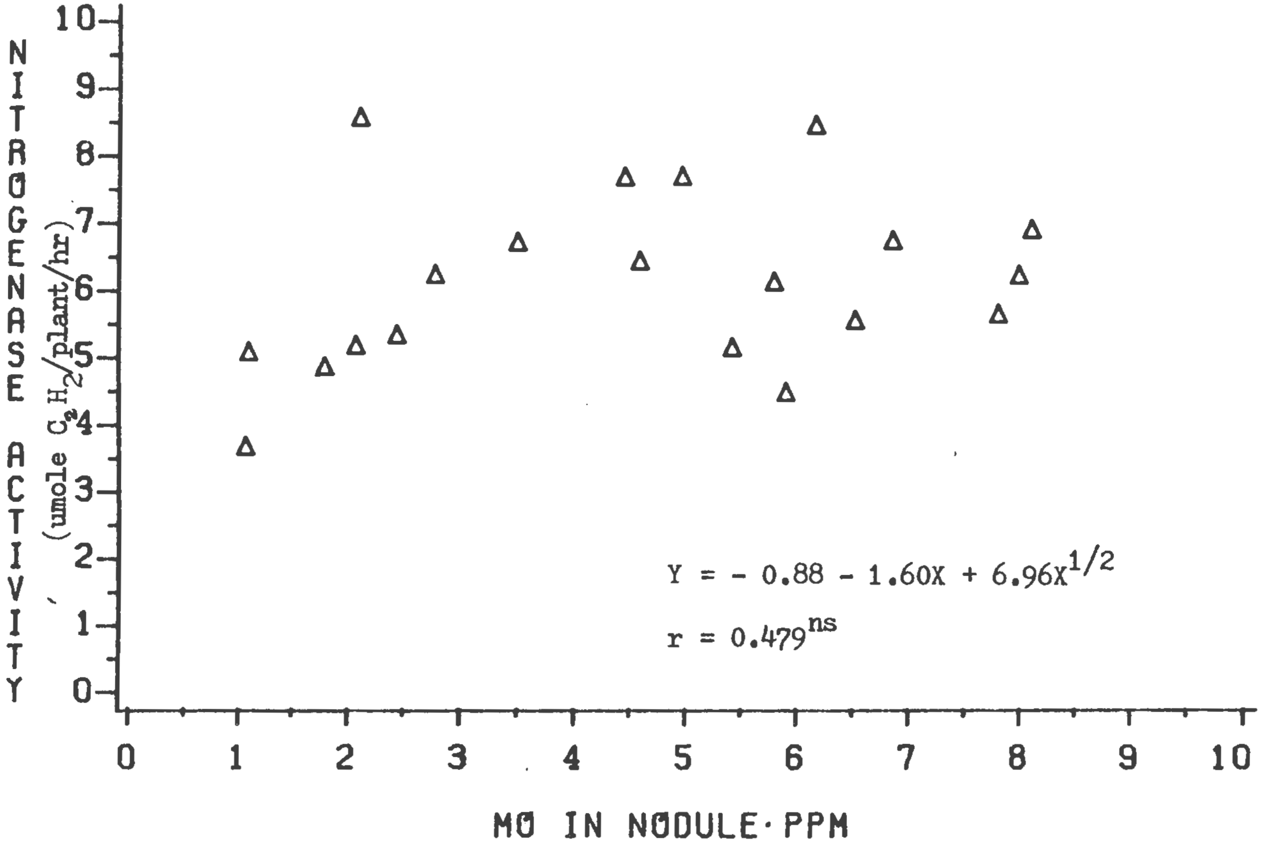


Fig. 1.18. Relationship between nitrogenase activity and Mo concentration in nodule of *Desmodium intortum* grown in the Wahiawa soil.

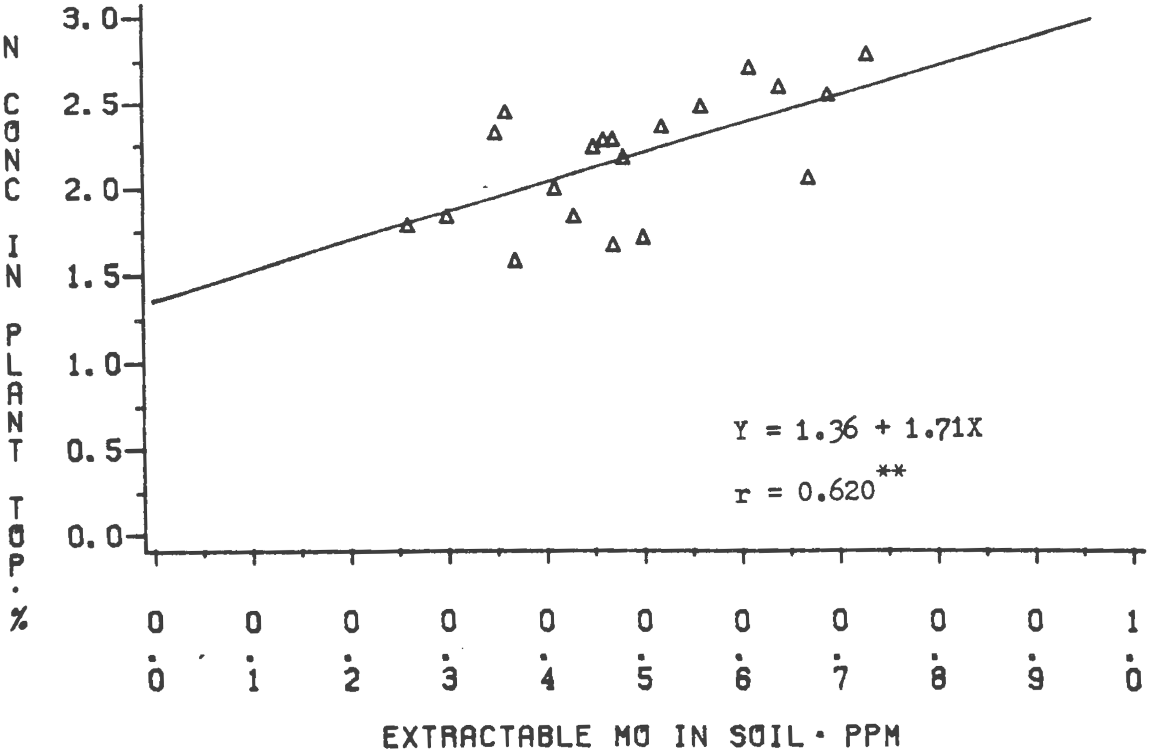


Fig. 1.17. Relationship between N concentration in the top of *Desmodium intortum* and extractable Mo in the Wahiawa soil.

revealed that when Mo concentration in nodule was low and seems inadequate for the requirement of nitrogenase enzymes, increasing Mo concentration in nodules increased nitrogenase activity up to a critical point where Mo concentration might be adequate for nitrogenase activity. When Mo concentration in nodule was adequate, the correlation between Mo concentration and nitrogenase activity became insignificant. According to a scatter diagram, ~~it may~~ approximate ~~that the~~ critical Mo concentration in nodule for high activity of nitrogenase was 4 ppm. For other legumes, Jensen (1946) reported that the critical Mo concentration in nodules for full activity of nitrogen fixation in alfalfa and clover was 10 and 4.8 ppm, respectively.

Relationship between Nitrogenase Activity and Mo Concentration in Plant Top

There was no significant correlation between nitrogenase activity and Mo concentration in plant top. The correlation coefficient of this relationship (0.126) was lower than that between nitrogenase activity and Mo concentration in nodule (0.479). Lack of the relationship between nitrogenase activity and Mo concentration in plant top suggests that Mo concentration in this plant part does not reflect the influence of Mo on nitrogenase activity of legume.

Relationship between Nitrogenase Activity and Extractable Mo in Soil

There was no significant correlation between nitrogenase activity and extractable Mo in soil. However there tended to be a relationship between nitrogenase activity and extractable Mo. The correlation coefficient of this relationship was 0.427 (p = 0.06).

Lack of a significant correlation between nitrogenase activity and Mo concentration in plant and soil may partly be due to the variation of acetylene reduction values and Mo concentration. Acetylene reduction among replicated samples was quite variable. Molybdenum concentrations in the top and nodule of plant as well as extractable Mo in soil were also quite variable due to the very low concentration of Mo which made analysis difficult.

Relationship between S and Mo Concentrations

Relationship between S Concentration in Plant Top and Mo Concentration in Nodule

There was a significant correlation between S concentration in plant top and Mo concentration in nodule with a correlation coefficient of 0.446. The relationship obtained by a scatter diagram (Fig. 1.19) indicated that Mo concentration decreased as S concentration in plant top increased. Sulfur concentration in plant top could reflect S concentration in root; i.e. S concentration in plant top is high when S concentration in plant root is high (Bouma, 1975). Therefore, S in the root may suppress Mo translocation from root to nodule, such that Mo concentration in nodule is decreased. The competitive effect between Mo and S was suggested by Stout et al. (1951). More over, Fried (1948) has found precisely the

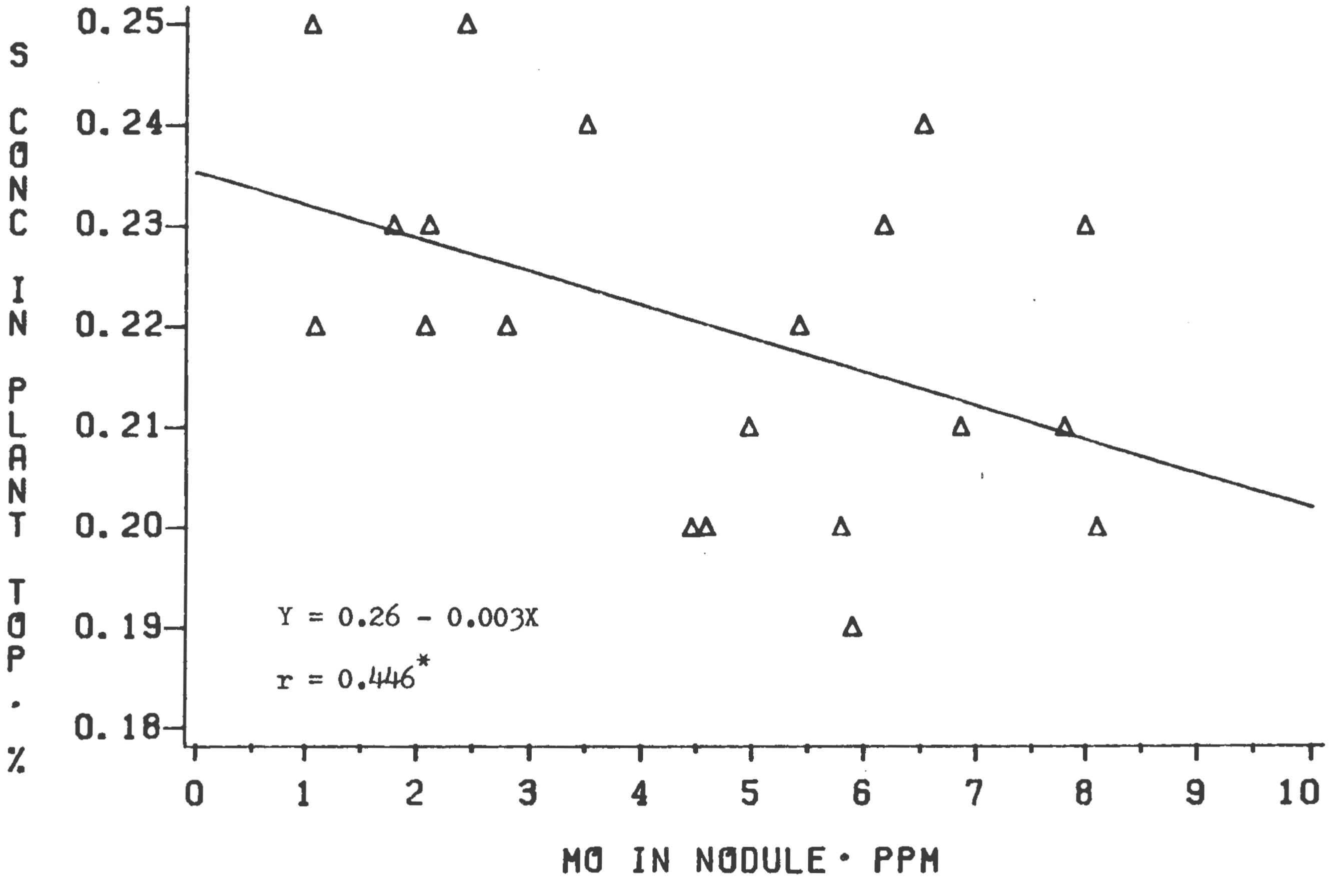


Fig. 1.19. Relationship between S concentration in plant top and Mo concentration in nodule of <u>Desmodium intortum</u> grown in the Wahiawa soil.

same pattern of distribution of absorbed sulfates in alfalfa as Stout and Meagher (1948) discovered for absorbed molybdate with tomato plant. These findings may help to explain the competitive effect of sulfate and molybdate in plant.

Relationship between S and Mo Concentrations in Plant Top

There was no significant correlation between S and Mo concentrations in plant top (r. 0.253). The relationship between S and Mo concentrations in plant top, therefore, should not be used to determine the competitive effect between Mo and S. The effect of S in decreasing Mo concentration as reported by many researchers (Stout et al., 1951; Stout and Meagher, 1958; Gupta and MacLeod, 1975) should be due to the competitive effect of S at the root. A high concentration of S at the root may block the translocation of Mo to the top while S concentration in the top remains unchanged. The total S concentration of red clover receiving S fertilizer up to 200 ppm S was not changed (Gupta and MacLeod, 1975).

Relationship between S Concentration in Plant Top and and Extractable Mo in soil

There was no significant correlation between S concentration in plant top and extractable Mo in soil. The correlation coefficient of the relationship was very low (0.067). This result suggested that molybdate in soil had no effect of sulfate absorption by plant.

Relationship between Cu and Mo Concentrations

Relationship between Cu Concentration in Plant Top and Mo Concentration in Nodule

Although there was no significant correlation between Cu concentration in plant top and Mo concentration in nodule ($r = 0.427$ and $p = 0.06$), Cu concentration in plant top tended to increase as Mo concentration in Nodule increased.

Relationship between Cu and Mo Concentrations in Plant Top

There was no significant correlation between Mo and Cu concentrations in plant top. The correlation coefficient was 0.180 ($p = 0.45$). The potential harmful effect of Mo regarding Mo-induced Cu deficiency in animals should not occur because Mo concentration in desmodium top is not exceptionally high and Cu concentration is in sufficient range. Copper to Mo ratio was well above three, which was the ratio under which Mo-induced Cu deficiency usually occurs (Alary et al., 1981). However, as the Mo application rate increased, Cu/Mo ratio decreased significantly. At the 4.0 kg Mo/ha soil-applied treatment, Cu/Mo ratio dropped from 153 to 11 (Table 1.22). Although the ratio of 11 was still safe for animals, this ratio was obtained only one cutting. For subsequent cutting of desmodium, the Cu/Mo ratio may be lowered until it approaches the harmful level. Thus the Cu/Mo ratio in legume tissue should be determined for each cutting in order to monitor and control it within a safe range for animals.

Table 1.22

Effects of Mo, S, and lime on the Cu/Mo ratio in the tops of <u>Desmodium intortum</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	Cu/Mo ratio				
No Mo added	287	97	115	115	153 a
0.1 kg Mo/ha (seed applied)	142	85	79	138	111 ab
0.5 kg Mo/ha (seed applied)	33	60	45	61	50 bc
2.0 kg Mo/ha (soil applied)	30	13	35	34	28 c
4.0 kg Mo/ha (soil applied)	10	13	11	8	11 c
	100	53	57	71	
Mean	77 a		64 a		

	No S	S
Mean	79 a	62 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Relationship between Cu Concentration in Plant Top and Extractable Mo in Soil

There was no significant correlation between Cu concentration in plant top and extractable Mo. The correlation coefficient of the relationship was 0.105 ($p = 0.66$). This result suggested that Mo in soil had no effect on Cu absorption by desmodium.

a.2. Effects of Mo, S and Lime on Growth, Yield and Nutrient Concentration of <u>Desmodium intortum</u> Grown in **the Paaloa Soil**

Yield Response

Liming significantly increased desmodium dry matter yield (Table 1.23). When averaged over all Mo and S rates, yield of desmodium grown in limed soil was increased 200.4% above that of desmodium grown in unlimed soil. When Mo and S were not added, yield of desmodium grown in limed soil was increased 650.2% above that of desmodium grown in unlimed soil. The results indicated that lime is very much essential for desmodium grown in Paaloa soil which has low pH and high Al (Table 1.1). A definite lime response was observed 2 to 3 weeks after the plants emerged. Plants which received no lime were light green, stunted and root extension was restricted. Roots of stunted plants were shortened and swollen. The entire root system had a characteristic stubby appearance.

what happ to Al [illegible]

Sulfur application significantly increased the dry matter yield of desmodium (Table 1.23). When averaged over all Mo and lime rates, yield of desmodium receiving S was increased 40.3% above that of plant receiving no S.

Although Mo did not significantly increase dry matter yield of desmodium when averaged over all S and lime rates, it tended to increase the yield of desmodium grown in limed soil with S application. The yields of desmodium receiving seed-applied Mo at 0.1 and 0.5 kg Mo/ha were 71.9% and 98.3%, respectively, above that of plant receiving no Mo. The yield of desmodium receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were 22.0%

Table 1.23.

Effects of Mo, S and lime on the dry matter yield of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	g/pot				
No Mo added	2.63	6.53	19.73	15.63	11.13 a*
0.1 kg Mo/ha (seed applied)	7.46	7.77	12.90	26.87	13.75 a
0.5 kg Mo/ha (seed applied)	8.63	6.93	17.50	31.00	16.02 a
2.0 kg Mo/ha (soil applied)	7.50	11.53	13.50	19.07	12.90 a
4.0 kg Mo/ha (soil applied)	5.43	5.17	20.70	32.17	15.88 a
	6.33	7.59	16.87	24.95	
Mean	6.96 b		20.91 a		

	No S	S
Mean	11.66 b	16.27 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

and 105.8%, respectively, above that of desmodium receiving no Mo. The results suggested that when growth of legume was limited by Al toxicity and/or S deficiency, the plant may not respond to Mo application. When Al toxicity was alleviated by liming and S fertilizer was applied, legume responded to Mo. Lime and S are the important factors limiting the dry matter yield of desmodium grown in the Paaloa soil.

Nodulation Response

Number of nodule

Lime significantly increased the number of nodules of desmodium grown in the Paaloa soil (Table 1.24). Desmodium grown in unlimed soil did not nodulate whereas that grown in limed soil nodulated very well indicating that lime was the most critical factor essential for nodulation of desmodium grown in the Paaloa soil.

Although Mo had no significant effect on nodule number of desmodium when averaged over all S and lime rates, there was a trend indicating that seed-applied Mo had a detrimental effect on <u>Rhizobium</u> inoculant since nodule number of legume receiving seed-applied Mo was less than that of legume receiving no Mo or soil-applied Mo at both 2.0 and 4.0 kg Mo/ha rates.

Sulfur applications did not significantly increase nodule number of desmodium when averaged over all Mo and lime rates. It was noted that while S had no effect on nodule number of desmodium grown in limed soil and receiving soil-applied Mo, it tended to increase nodule number of desmodium grown in limed soil and receiving seed-applied Mo. Nodule number of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha with S was 160.8%

Table 1.24.

Effects of Mo, S and lime on number of nodules of Desmodium intortum grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	number				
No Mo added	0	0	598	496	274 a*
0.1 kg Mo/ha (seed applied)	0	0	293	764	265 a
0.5 kg Mo/ha (seed applied)	2	0	130	456	147 a
2.0 kg Mo/ha (soil applied)	0	1	561	264	207 a
4.0 kg Mo/ha (soil applied)	0	0	483	484	242 a
	0	0	413	493	
Mean	0 b		453 a		

	No S	S
Mean	207 a	247 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

above that of desmodium receiving the same rate of Mo without S. Nodule number of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha with S was 250.8% above that of desmodium receiving the same rate of Mo without S. Although the difference in nodule number was not statistically significant, a definite trend is evident. Nodule number of desmodium receiving seed-applied Mo with S tended to be higher than that of desmodium receiving seed-applied Mo without S. It was also observed that nodule number of plant receiving seed-applied Mo at 0.1 kg Mo/ha with S was significantly higher than that of plant receiving seed-applied Mo at 0.5 kg Mo/ha without S. The results suggested that S alleviated the detrimental effect of Mo salt to <u>Rhizobium</u> inoculant. Sulfur may help to increase the multiplication of rhizobia that were not killed by Mo salt so that the population of rhizobia is high enough to infect plant roots effectively.

Fresh Weight of Nodule

Lime significantly increased nodule fresh weight of desmodium grown in the Paaloa soil (Table 1.25). The increase in nodule weight as affected by lime was due to the influence of lime in increasing nodule number (Table 1.24).

Although Mo had no significant effect on nodule fresh weight of desmodium when average over all S and lime rates, Mo had a significant effect on nodule weight of desmodium grown in limed soil without S application. Nodule weight of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was significantly lower than that of desmodium receiving no Mo. Although it was not significantly different, nodule weight of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha was 64.2% lower than that of

Table 1.25.

Effects of Mo, S and lime on nodule fresh weight of *Desmodium intortum* grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	g/pot				
No Mo added	0 d*	0 d	6.78 ab	4.23 abcd	2.75 a
0.1 kg Mo/ha (seed applied)	0 d	0.01 d	2.43 bcd	8.47 a	2.73 a
0.5 kg Mo/ha (seed applied)	0.01 d	0 d	1.56 cd	6.84 ab	2.10 a
2.0 kg Mo/ha (soil applied)	0 d	0 d	5.74 abc	3.80 abcd	2.39 a
4.0 kg Mo/ha (soil applied)	0 d	0 d	6.92 ab	7.40 a	2.58 a
	0.002	0.002	4.69	6.15	
Mean	0.002 b		5.42 a		

	No S	S
Mean	2.34 a	3.08 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

desmodium receiving no Mo. These results indicated that Mo salt had a harmful effect on rhizobia.

Although S had no significant effect on nodule fresh weight of desmodium when averaged over all Mo and lime rates, S significantly increased nodule weight of desmodium grown in limed soil and receiving seed-applied Mo. Nodule fresh weight of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha with S was significantly higher than that of legume receiving the same rate of Mo without S. Nodule weight of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha with S was significantly higher than that of legume receiving Mo at the same rate without S. The increase in weight of nodule as affected by S may, partly, be due to the influence of S in increasing nodule number (Table 1.24). Sulfur did not significantly increase weight of nodules ~~of~~ desmodium ~~grown~~ in limed soil and receiving soil-applied Mo at both 2.0 and 4.0 kg Mo/ha rates. [illegible]

Dry Weight of Nodules:

Lime significantly increased the nodule dry weight of desmodium (Table 1.26). The increased nodule weight of desmodium grown in limed soil was due to the influence of lime in increasing the nodule number (Table 1.24) since there was almost no nodules formed for desmodium grown in unlimed Paaloa soil. Although Mo had little effect on dry weight of nodule when average over all lime and S rates, Mo had a significant effect on nodule weight when lime and no S were applied. Nodule weight of desmodium which had receiv~~ing~~ed seed-applied Mo at 0.5 kg Mo/ha without S was significantly lower than that of legume ~~receiving~~ with no Mo. Despite the lack of significant difference, weight of desmodium receiving seed-applied

Table 1.26.

Effects of Mo, S and lime on nodule dry weight of <u>Desmodium intortum</u> grown in the Paaloa soil

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	g/pot				
No Mo added	0 d*	0 d	1.14 ab	0.69 abcd	0.46 a
0.1 kg Mo/ha (seed applied)	0 d	0 d	0.42 bcd	1.43 a	0.46 a
0.5 kg Mo/ha (seed applied)	0 d	0 d	0.21 cd	1.13 ab	0.34 a
2.0 kg Mo/ha (soil applied)	0 d	0 d	0.97 abc	0.66 abcd	0.41 a
4.0 kg Mo/ha (soil applied)	0 d	0 d	1.16 ab	1.24 ab	
	0	0	0.78	1.03	
Mean	0 b		0.91 a		
	No S		S		
Mean	0.39 a		0.52 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Mo at 0.1 kg Mo/ha was 63.2% lower than that of legume receiving no Mo. Nodule weight of desmodium receiving soil-applied Mo at both rates was not significantly different from that of legume receiving no Mo.

When S was applied to desmodium grown in limed soil, there was no significant difference in nodule dry weight of legume receiving various rates of Mo. However, S significantly increasd nodule weight of desmodium receiving seed-applied Mo at both 0.1 and 0.5 kg Mo/ha rates. Nodule weight of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha with S was significantly higher than that of legume receiving the same rate of Mo without S. Nodule weight of desmodium receiving 0.5 kg Mo/ha with S was also significantly higher than that of legume receiving the same rate of Mo without S. The results suggested that although seed-applied Mo had a detrimental effect on rhizobia and reduced nodulation, S ~~fertilizer~~ seems to ~~be a factor~~ alleviat~~ing~~e that effect by increasing the nodulation of desmodium grown in limed Paaloa soil.

Influences of Mo, S and Lime on Some Chemical Composition in Plant

Nitrogen:

Liming significantly increased N concentration in plant top (Table 1.27). When averaged over all Mo and S rates, lime increased N concentration by 40.3% above unlimed treatment. Molybdenum did not significantly increase N concentration in plant top. Nitrogen concentration in the top of desmodium grown in unlimed soil and receiving various rates of Mo was not significantly different among the treatments. Molybdenum fertilizer had little effect on N concentration in the top of

Table 1.27.

Effects of Mo, S and lime on N concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

Mo treatment	No lime No S	 S	Lime No S	 S	Mean
			%		
No Mo added	2.24	2.03	3.44	2.98	2.67 a*
0.1 kg Mo/ha (seed applied)	2.50	2.39	2.98	3.08	2.74 a
0.5 kg Mo/ha (seed applied)	1.90	2.32	3.05	2.96	2.56 a
2.0 kg Mo/ha (soil applied)	2.52	1.88	3.41	2.82	2.66 a
4.0 kg Mo/ha (soil applied)	2.21	2.06	2.93	3.37	2.65 a
	2.28	2.14	3.16	3.04	
Mean	2.21 b		3.10 a		

	No S	S
Mean	2.72 a	2.59 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

desmodium grown in unlimed soil because the plant had no nodule. Therefore, N concentration in plant top was likely to have been obtained from soil N and not from the nitrogen fixation process which requires Mo. Sulfur had no significant effect on N concentration in the top of plants.

Phosphorus:

There was no significant difference in P concentration in the tops of desmodium receiving various rates of Mo, S and lime (Table 1.28). Molybdenum did not significantly increase P concentration in plant tops. Sulfur and lime also had little effect on P concentration. Due to the lack of nodules in legume grown in unlimed soil, it should be noted, therefore, that P concentrations in the tops of non-nodulated and nodulated desmodium were similar.

Potassium:

Molybdenum application did not significantly increase K concentration in the tops of desmodium grown in Paaloa soil (Table 1.29). Liming significantly decreased K concentration. The effect of lime was in accord with the results reported by Andrew and Johnson (1976). The change in K concentration in plant tops as influenced by lime may be due to the effect of Ca on K absorption or to the capacity of plant to preserve cation balance.

Calcium:

Molybdenum application did not significantly increase Ca

Table 1.28.

Effects of Mo, S and lime on P concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
			%		
No Mo added	0.23	0.28	0.28	0.32	0.28 a*
0.1 kg Mo/ha (seed applied)	0.32	0.32	0.29	0.24	0.29 a
0.5 kg Mo/ha (seed applied)	0.27	0.35	0.35	0.22	0.30 a
2.0 kg Mo/ha (soil applied)	0.34	0.27	0.33	0.32	0.32 a
4.0 kg Mo/ha (soil applied)	0.32	0.31	0.27	0.28	0.30 a
	0.30	0.31	0.30	0.28	
Mean	0.30 a		0.29 a		

	No S	S
Mean	0.30 a	0.29 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.29.

Effects of Mo, S and lime on K concentration in the top of Desmodium intortum grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	2.81	3.24	2.49	2.72	2.82 a*
0.1 kg Mo/ha (seed applied)	3.73	3.89	2.99	2.14	3.19 a
0.5 kg Mo/ha (seed applied)	3.64	3.33	2.63	1.98	2.90 a
2.0 kg Mo/ha (soil applied)	3.69	3.39	3.41	2.52	3.26 a
4.0 kg Mo/ha (soil applied)	3.74	4.05	2.19	2.72	3.05 a
	3.53	3.58	2.74	2.32	
Mean	3.55 a		2.53 b		

	No S	S
Mean	3.13 a	2.95 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

concentration in plant tops of desmodium grown in Paaloa soil (Table 1.30). Liming significantly increased Ca concentration while S had little effect.

Magnesium:

Molybdenum had no significant effect on Mg concentration in the top of desmodium (Table 1.31). Magnesium concentration did not change when Mo was applied as seed coating or as soil treatment. Sulfur also did not affect Mg concentration. Lime, however, significantly decreased Mg concentration. The effect of lime in decreasing Mg concentration was likely due to the capacity of the plants to preserve cation balance (Andrew and Johnson, 1976).

Sulfur:

Molybdenum had no significant effect on S concentration in plant top (Table 1.32). Sulfur also had no significant effect on S concentration. Lime significantly decreased S concentration in plant top. Sulfur concentration in the tops of desmodium grown in limed soil was 25.0% lower than that of desmodium grown in unlimed soil. The reason that S concentration in the top of desmodium grown in limed soil was lower than that of legume grown in unlimed soil may be due to a "dilution effect" of growth since yield of desmodium grown in limed soil was much higher than that of legume grown in unlimed soil. It should be noted that although S concentration in desmodium grown in limed soil was lower than that of plant grown in unlimed soil, S concentration was still high when

Table 1.30.

Effects of Mo, S and lime on Ca concentration in the top of *Desmodium intortum* grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
			%		
No Mo added	0.60	0.85	1.54	1.93	1.23 a*
0.1 kg Mo/ha (seed applied)	1.35	1.02	1.52	1.46	1.34 a
0.5 kg Mo/ha (seed applied)	0.86	1.33	1.54	1.47	1.30 a
2.0 kg Mo/ha (soil applied)	1.21	0.87	1.52	1.73	1.33 a
4.0 kg Mo/ha (soil applied)	1.04	1.00	1.54	1.57	1.29 a
	1.01	1.01	1.53	1.63	
Mean	1.01 b		1.58 a		

	No S	S
Mean	1.27 a	1.32 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.31.

Effects of Mo, S and lime on Mg concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

Mo treatment	No lime No S	No lime S	Lime No S	Lime S	Mean
	------------	------------	%	------------	------------
No Mo added	0.47	0.45	0.22	0.30	0.36 a*
0.1 kg Mo/ha (seed applied)	0.42	0.49	0.32	0.22	0.36 a
0.5 kg Mo/ha (seed applied)	0.41	0.44	0.30	0.22	0.34 a
2.0 kg Mo/ha (soil applied)	0.43	0.43	0.36	0.28	0.37 a
4.0 kg Mo/ha (soil applied)	0.46	0.51	0.22	0.26	0.36 a
	0.44	0.47	0.28	0.25	
Mean	0.45 a		0.36 b		

	No S	S
Mean	0.36 a	0.36 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.32.

Effects of Mo, S and lime on S concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	-----------------------		%	-----------------------------	
No Mo added	0.38	0.42	0.30	0.39	0.37 a*
0.1 kg Mo/ha (seed applied)	0.47	0.47	0.37	0.27	0.39 a
0.5 kg Mo/ha (seed applied)	0.42	0.44	0.37	0.24	0.37 a
2.0 kg Mo/ha (soil applied)	0.48	0.41	0.44	0.35	0.42 a
4.0 kg Mo/ha (soil applied)	0.47	0.49	0.29	0.31	0.39 a
	0.44	0.45	0.35	0.31	
Mean	0.44 a		0.33 b		

	No S	S
Mean	0.40 a	0.38 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

compared to the S critical level of 0.15% to 0.18% in desmodium as suggested by Andrew (1977). Despite the fact that S concentration in plant top was higher than their suggested critical level, desmodium still responded to S application (Table 1.22). Sulfur concentration in plant top alone appears to be not enough for evaluation of plant response.

Iron:

Molybdenum and S had no significant effect while lime significantly decreased Fe concentration in desmodium tops (Table 1.33). Iron concentration in plants grown in limed soil was decreased 30.1% from those grown in unlimed soil. The decrease in Fe concentration was likely due to a "dilution effect" of growth and/or the decrease of Fe availability in soil as the soil pH was raised.

Manganese:

Manganese concentration in the tops of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was significantly lower than in those of desmodium receiving no Mo or seed-applied Mo at 0.1 kg Mo/ha but was not significantly lower than in those of desmodium receiving soil-applied Mo at both 2.0 and 4.0 kg Mo/ha (Table 1.34). Manganese concentration in the tops of desmodium receiving no Mo, seed-applied Mo at 0.1 kg Mo/ha and the two rates of soil-applied Mo were not significantly different among them. Sulfur had little effect on Mn concentration. Lime significantly decreased Mn concentration. Manganese concentration in desmodium grown in limed soil was 54.0% lower from that in legume grown in unlimed soil when averaged over all Mo and S rates.

Table 1.33.

Effects of Mo, S and lime on Fe concentration in the top of Desmodium intortum grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	190	230	129	165	159 a*
0.1 kg Mo/ha (seed applied)	164	267	169	130	154 a
0.5 kg Mo/ha (seed applied)	178	162	135	142	154 a
2.0 kg Mo/ha (soil applied)	289	213	200	123	167 a
4.0 kg Mo/ha (soil applied)	178	221	121	149	154 a
	200	219	151	142	
Mean	209 a		146 b		

	No S	S
Mean	175 a	180 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.34.

Effects of Mo, S and lime on Mn concentration in the top of <u>Desmodium intortum</u> grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
			ppm		
No Mo added	197	178	72	64	128 a*
0.1 kg Mo/ha (seed applied)	160	197	110	66	128 a
0.5 kg Mo/ha (seed applied)	137	114	60	44	89 b
2.0 kg Mo/ha (soil applied)	126	132	107	78	111 ab
4.0 kg Mo/ha (soil applied)	162	174	62	81	120 ab
	156	159	82	63	
Mean	158 a		72 b		
	No S	S			
Mean	119 a	111 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Copper:

There were no significant differences among Cu concentrations in desmodium tops receiving various rates of Mo by seed coating or by soil application (Table 1.35). Sulfur had no effect on Cu concentration. Lime, however, significantly decreased Cu concentration. When averaged over all Mo and S rates, Cu concentration in tops of desmodium grown in limed soil was 19.5% lower than that of plant grown in unlimed soil.

Zinc:

Molybdenum had no significant effect on Zn concentration in desmodium tops since the concentration did not change when various rates of Mo was applied (Table 1.36). Sulfur also had no significant effect on Zn concentration. Lime significantly decreased Zn concentration.

Silicon:

Silicon concentration in desmodium tops receiving soil-applied Mo at 2.0 kg Mo/ha was significantly higher than in those receiving seed-applied Mo at 0.5 kg Mo/ha but was not significantly higher than in those receiving other rates of Mo (Table 1.37). The reason that Si concentration in plant receiving seed-applied Mo at 0.5 kg Mo/ha was lower than in those receiving other rates of Mo was probably due to a "dilution effect" of growth since the dry matter yield of this treatment was higher than other treatments (Table 1.22). Silicon concentration in desmodium tops receiving no Mo, seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at both rates were not significantly different from each other.

Table 1.35.

Effects of Mo, S and lime on Cu concentration in the top of Desmodium intortum grown in the Paaloa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
			ppm		
No Mo added	11.7	9.3	6.7	6.3	8.5 a*
0.1 kg Mo/ha (seed applied)	7.3	8.7	7.3	7.3	7.7 a
0.5 kg Mo/ha (seed applied)	10.0	8.7	8.3	7.0	8.5 a
2.0 kg Mo/ha (soil applied)	9.3	10.3	7.7	7.7	8.8 a
4.0 kg Mo/ha (soil applied)	8.0	7.7	7.7	7.3	7.7 a
	9.3	8.9	7.5	7.1	
Mean	9.1 a		7.3 b		

	No S	S
Mean	8.4 a	8.0 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.36.

Effects of Mo, S and lime on Zn concentration in the top of *Desmodium intortum* grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	54.6	56.7	39.0	44.7	48.8 a*
0.1 kg Mo/ha (seed applied)	53.0	53.7	46.0	36.0	47.2 a
0.5 kg Mo/ha (seed applied)	53.3	56.7	47.3	36.0	48.3 a
2.0 kg Mo/ha (soil applied)	55.0	49.3	47.0	43.3	48.7 a
4.0 kg Mo/ha (soil applied)	62.3	56.3	39.7	40.0	49.6 a
	55.7	54.5	43.8	40.0	
Mean	55.1 a		41.9 b		

	No S	S
Mean	49.7 a	47.3 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.37.

Effects of Mo, S and lime on Si concentration in the top of Desmodium intortum grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	%				
No Mo added	1.15	1.34	0.95	1.07	1.13 ab*
0.1 kg Mo/ha (seed applied)	1.50	1.66	1.28	0.83	1.32 a
0.5 kg Mo/ha (seed applied)	1.15	1.77	1.10	0.77	1.05 b
2.0 kg Mo/ha (soil applied)	1.42	1.38	1.21	1.10	1.28 a
4.0 kg Mo/ha (soil applied)	1.40	1.43	0.89	1.04	1.19 ab
	1.32	1.40	1.08	0.97	
Mean	1.36 a		1.03 b		

	No S	S
Mean	1.21 a	1.18 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Lime significantly decreased Si concentration. This similar effect of lime on Si concentration was also observed in desmodium grown in Wahiawa soil at the previous greenhouse experiment. Sulfur had no significant effect on Si concentration.

Aluminum:

Alumunum concentration in desmodium tops receiving soil-applied Mo at 4.0 kg Mo/ha was significantly lower than those receiving seed-applied Mo at 0.5 kg Mo/ha (Table 1.38). Aluminum concentrations in plant receiving no Mo, seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at both rates were not significantly different from each other. Sulfur tended to reduce Al concentration. Lime also tended to reduce Al concentration. It should be noted that Al concentration in the top of desmodium grown in unlimed soil was not significantly higher than in that of plant grown in limed soil although Al toxicity symptom was observed in plants grown in unlimed soil. Those results suggest that Al concentration in plant tops is not a good criterion to assess Al toxicity level.

Influences of Mo, S and Lime on Mo Concentration in Plant and Soil

Molybdenum Concentration in nodule

Desmodium did not nodulate when lime was not applied. Molybdenum concentration in nodule, therefore, could not be determined. For limed soil, Mo application significantly increased Mo concentration in nodule (Table 1.39). Molybdenum concentration in nodule of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than

Table 1.38.

Effects of Mo, S and lime on Al concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

Mo treatment	No lime: No S	No lime: S	Lime: No S	Lime: S	Mean
	ppm				
No Mo added	488	260	222	315	321 ab*
0.1 kg Mo/ha (seed applied)	243	231	340	306	280 ab
0.5 kg Mo/ha (seed applied)	488	288	406	335	379 a
2.0 kg Mo/ha (soil applied)	280	324	279	195	270 ab
4.0 kg Mo/ha (soil applied)	171	241	105	203	180 b
	334	269	271	271	
Mean	301 a		271 a		
	No S	S			
Mean	302 a	270 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.39.

Effects of Mo, S and lime on Mo concentration in nodule of *Desmodium intortum* grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	-	-	2.33	2.75	1.27 c*
0.1 kg Mo/ha (seed applied)	-	-	2.88	3.07	1.49 c
0.5 kg Mo/ha (seed applied)	-	-	2.81	6.39	2.30 c
2.0 kg Mo/ha (soil applied)	-	-	8.23	9.83	4.52 b
4.0 kg Mo/ha (soil applied)	-	-	12.29	14.04	6.58 a
	-	-	5.71	7.22	
Mean	-		6.46 a		
	No S		S		
Mean	2.85 a		3.61 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

those of desmodium receiving other rates of Mo. Molybdenum concentration in nodule of desmodium receiving soil-applied **Mo at 2.0 kg Mo/ha was significantly** higher than those of plant receiving no Mo and seed-applied Mo at both rates. Although Mo concentration of plant receiving seed-applied Mo at 0.5 kg Mo/ha was 81.1% above that of plant receiving no Mo, the different was not significant. Molybdenum concentration of plant receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly higher than that of **plant** receiving no Mo.

Lime significantly affected Mo concentration in nodule. However, there was no nodule development in plants grown in unlimed soil. Therefore the effect of lime was due to its influence on nodulation. Sulfur did not significantly affect Mo concentration in nodule of desmodium grown in Paaloa soil.

Molybdenum Concentration in Plant Top

The stunted growth of plants from unlimed treatment required the pooling of treatment replications for Mo analysis, thereby precluding statistical evaluation of the data. The trends and differences, however, tended to accord with the previous study on the effects of Mo, S and lime on desmodium grown in Wahiawa soil. Molybdenum concentration in the top of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha was the highest (Table 1.40). Molybdenum concentration of plant receiving soil-applied Mo at 2.0 kg Mo/ha was 69.8%, 329.4%, and 1116.7% higher than those of plant receiving seed-applied Mo at 0.5 kg Mo/ha, 0.1 kg Mo/ha and no Mo, respectively. **Lime** also increased **Mo** concentration in plant top. Sulfur had little effect on Mo concentration.

Table 1.40.

Effects of Mo, S and lime on Mo concentration in the top of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	0.01	0.08	0.09	0.08	0.06
0.1 kg Mo/ha (seed applied)	0.24	0.17	0.13	0.13	0.17
0.5 kg Mo/ha (seed applied)	0.35	0.76	0.99	0.82	0.43
2.0 kg Mo/ha (soil applied)	0.26	0.19	0.47	0.79	0.73
4.0 kg Mo/ha (soil applied)	0.73	1.23	1.92	1.88	1.44
	0.32	0.49	0.72	0.74	
Mean	0.40		0.73		

	No S	S
Mean	0.52	0.61

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

When Mo concentrations in the top of desmodium grown in unlimed (nonnodulated plant) and limed (nodulated plant) soil were plotted against the amounts of Mo applied by seed coating or soil application, a similar trend of relationship between Mo concentration and seed-applied or soil-applied was observed (Fig. 1.20). At the same rate of soil-applied Mo, Mo concentration in the top of nodulated plant (limed treatment) was higher than in nonnodulated plant (unlimed treatment). At no-Mo and 0.5 kg Mo/ha seed-applied Mo treatment, Mo concentration in nodulated plant (limed treatment) was also higher than in nonnodulated plant (unlimed treatment) (Fig. 1.20). The results suggested that nodulated plant absorbed more Mo from soil than nonnodulated plant. The reason for this effect was probably due to the high requirement of Mo for nitrogen fixation of nodulated plant.

Extractable Mo in Soil after Harvesting

Molybdenum application significantly increased extractable Mo in soil (Table 1.41). Extractable Mo in soil with soil-applied Mo at 4.0 kg Mo/ha was significantly higher than in soil added with Mo at other rates. Extractable Mo in soil with soil-applied Mo at 2.0 kg Mo/ha was not significantly higher than that in soil with seed-applied Mo at 0.5 kg Mo/ha but was significantly higher than that in soil with soil-applied Mo at 0.1 kg Mo/ha and no Mo. Extractable Mo obtained from the 0.5 kg Mo/ha seed-applied treatment was not significantly higher than that obtained from the 0.1 kg Mo/ha seed-applied treatment but was significantly higher than that obtained from the no-Mo treatment. Sulfur and lime had no effect on extractable Mo in soil after harvesting.

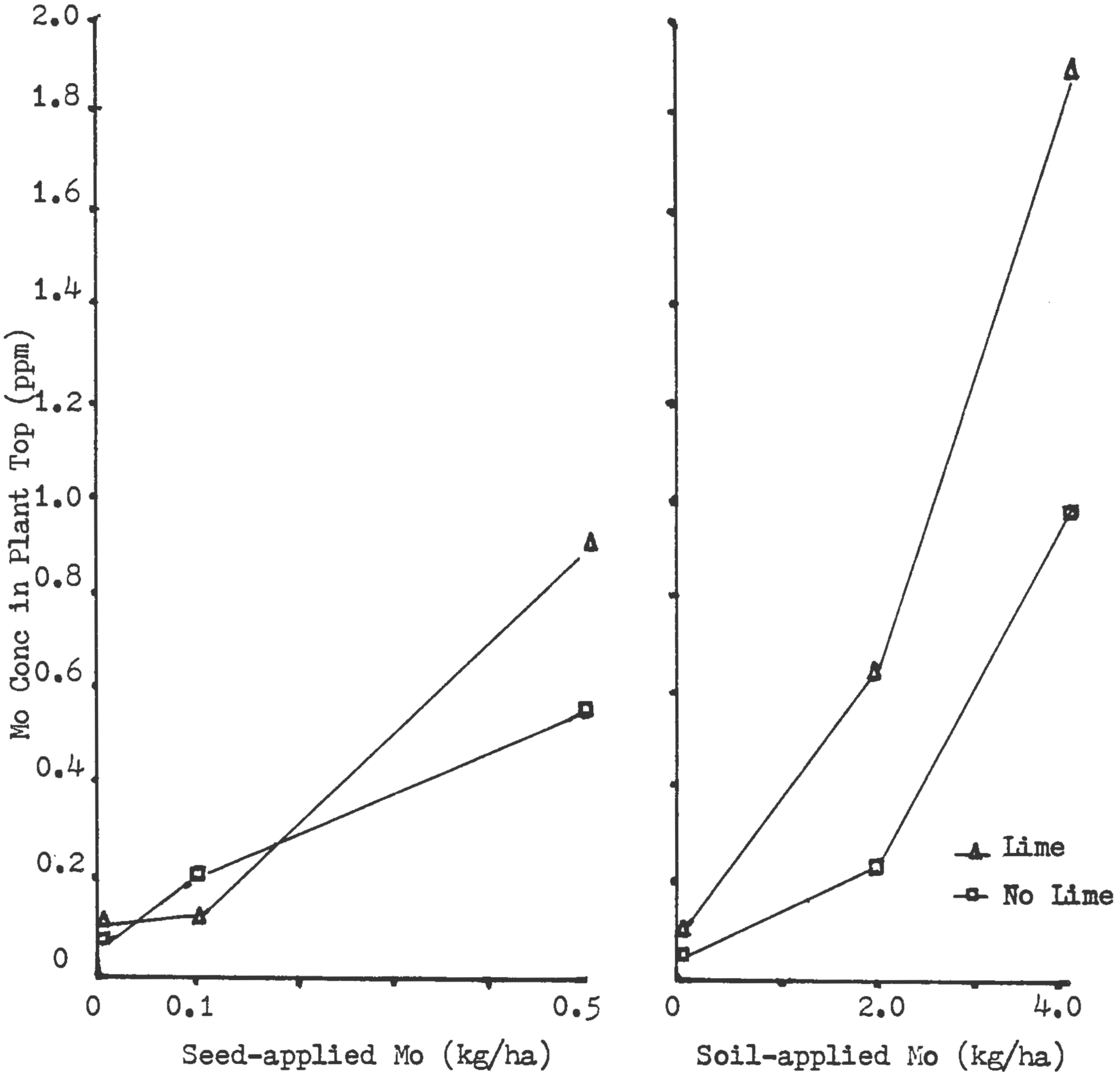


Fig. 1.20. Effects of seed-applied and soil-applied Mo, with or without lime, on Mo concentration in the top of Desmodium intortum grown in the Paaloa soil.

Table 1.41.

Effects of Mo, S and lime on extractable Mo in the Paaloa soil after harvesting *Desmodium intortum*.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	0.34	0.32	0.34	0.39	0.35 d*
0.1 kg Mo/ha (seed applied)	0.41	0.34	0.38	0.45	0.40 cd
0.5 kg Mo/ha (seed applied)	0.41	0.42	0.56	0.40	0.45 bc
2.0 kg Mo/ha (soil applied)	0.45	0.63	0.44	0.52	0.51 b
4.0 kg Mo/ha (soil applied)	0.84	0.79	0.90	0.81	0.84 a
	0.49	0.50	0.53	0.51	
Mean	0.50 a		0.52 a		

	No S	S
Mean	0.51 a	0.51 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Relationship between Mo Concentrations in Plant and Soil

Relationship between Mo Concentration in Plant Top and Mo Concentration in Nodule

There was a highly significant correlation between Mo concentration in plant top and Mo concentration in nodule of desmodium grown in Paaloa soil (Fig. 1.21). The shape of the curve drawn from a scatter diagram was similar to that drawn from a diagram of the relationship of desmodium grown in Wahiawa soil (Fig. 1.6). The results suggest that the relationship between Mo concentration in top and nodule of legume grown in different soil types was the same.

Relationship between Mo Concentration in Plant Top and Extractable Mo in Soil

There was a highly significant correlation between Mo concentration in desmodium top and extractable Mo in soil (Fig. 1.22). Molybdenum concentration increased as extractable Mo increased. The result suggested that although there were some factors (e.g. Al toxicity, S deficiency) limiting plant growth, Mo concentration in top was still dependent on extractable Mo in soil.

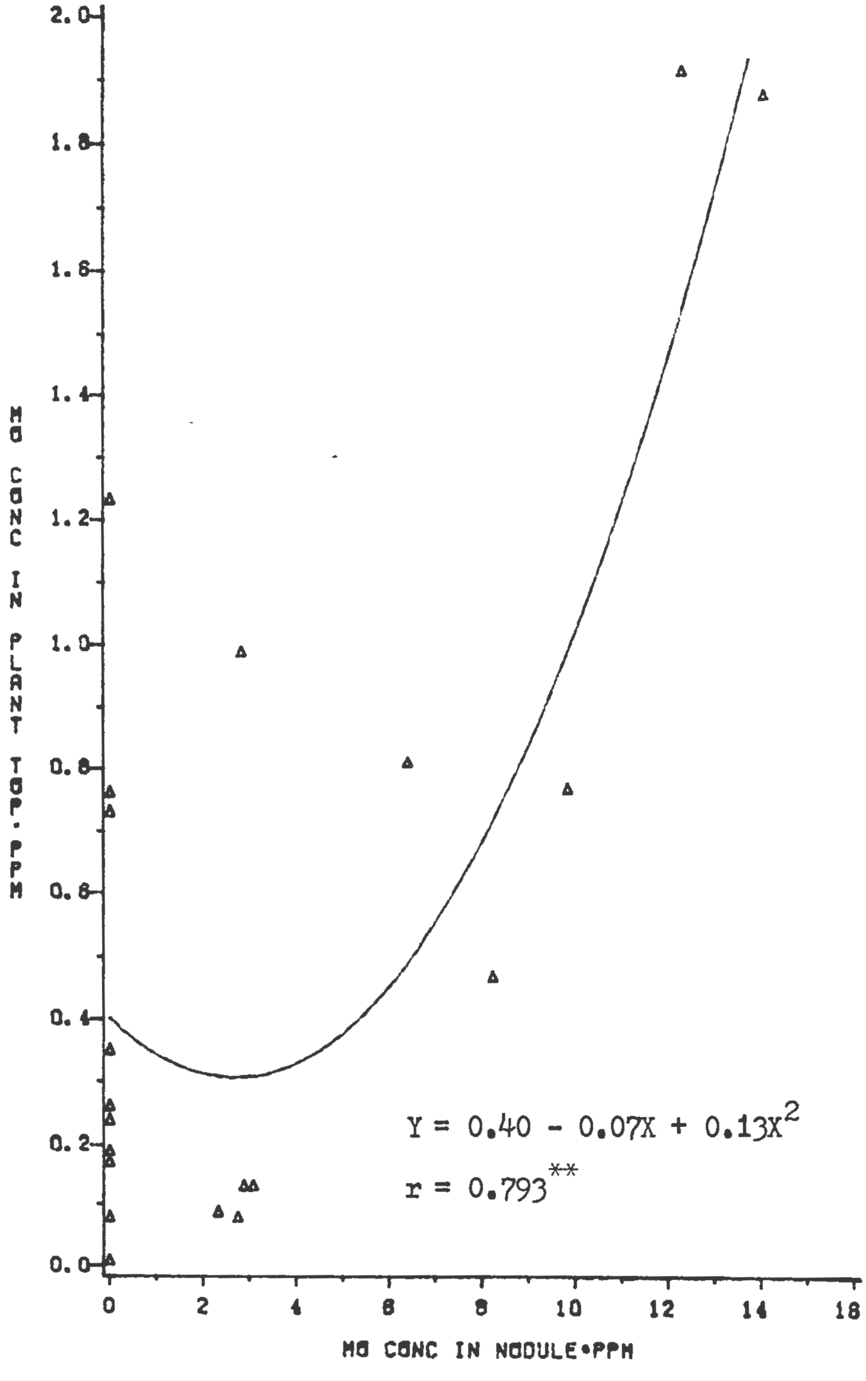


Fig. 1.21. Relationship between Mo concentration in plant top and Mo concentration in nodule of Desmodium intortum grown in the Paaloa soil.

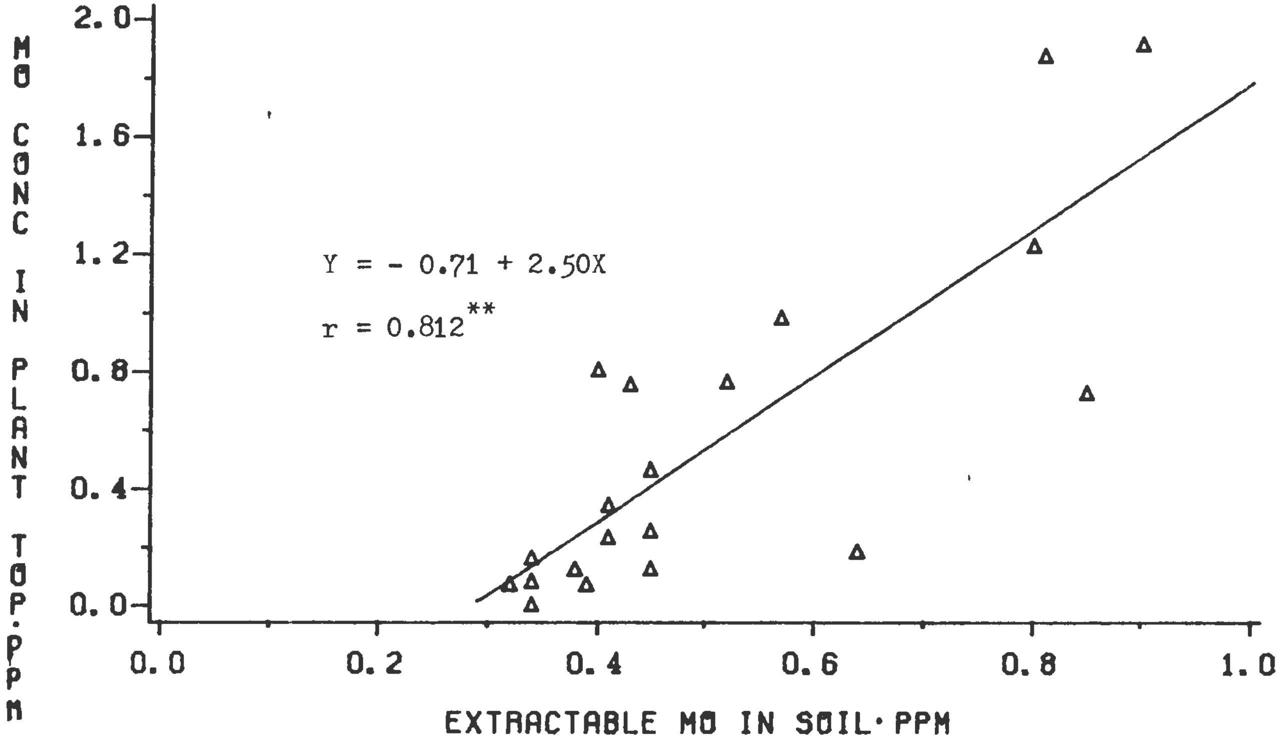


Fig. 1.22. Relationship between Mo concentration in the top of Desmodium intortum and extractable Mo in the Paaloa soil.

Relationship between Dry Matter Yield and Mo Concentrations

Relationship between Dry Matter Yield and Mo Concentration in Nodule

There was a highly significant correlation between dry matter yield and Mo concentration in nodule. The yield of desmodium increased as the Mo concentration in nodule increased (Fig. 1.23). There was a highly significant correlation between relative yield and Mo concentration(Fig. 1.24). The Mo critical level could not be determined because lime and S were limiting factors for plant growth. The magnitude of a response curve was low suggesting that the response of desmodium to Mo was low. The low response of Mo was due to the strong influences of Al toxicity. The dry matter yields obtained from desmodium receiving high rates of Mo without lime were low while the Mo concentration of plants receiving the same treatment were high (Table 1.23). The results suggested that the critical level of Mo concentration cannot be defined when there were factors other than Mo limiting plant growth.

Relationship between Dry Matter Yield and Mo Concentration in Plant Top

There was no significant correlation between dry matter yield and Mo concentration in plant top (Fig. 1.25). There was also no significant correlation between relative yield and Mo concentration (Fig. 1.26). Lack of a significant difference between dry matter yield or relative yield and Mo concentration was due to Al toxicity and S deficiency in the Paaloa soil.

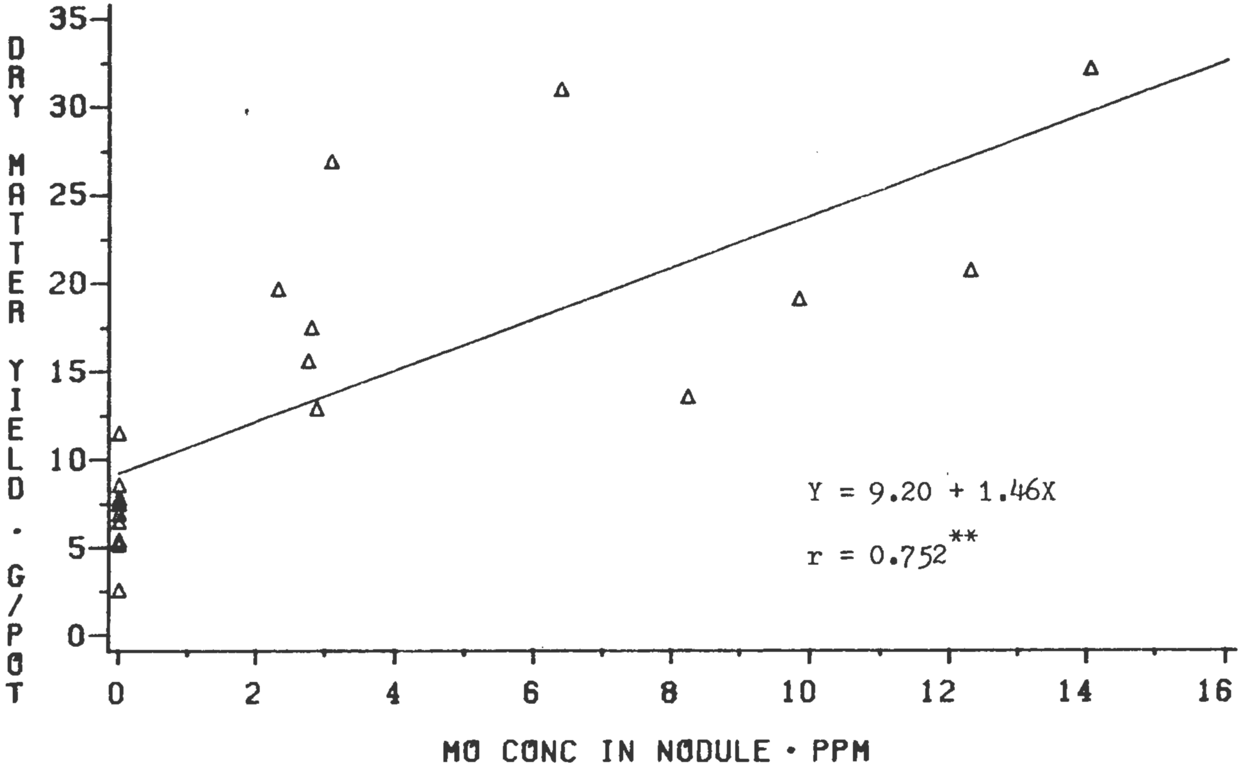


Fig. 1.23. Relationship between dry matter yield and Mo concentration in nodule of *Desmodium intortum* grown in the Paaloa soil.

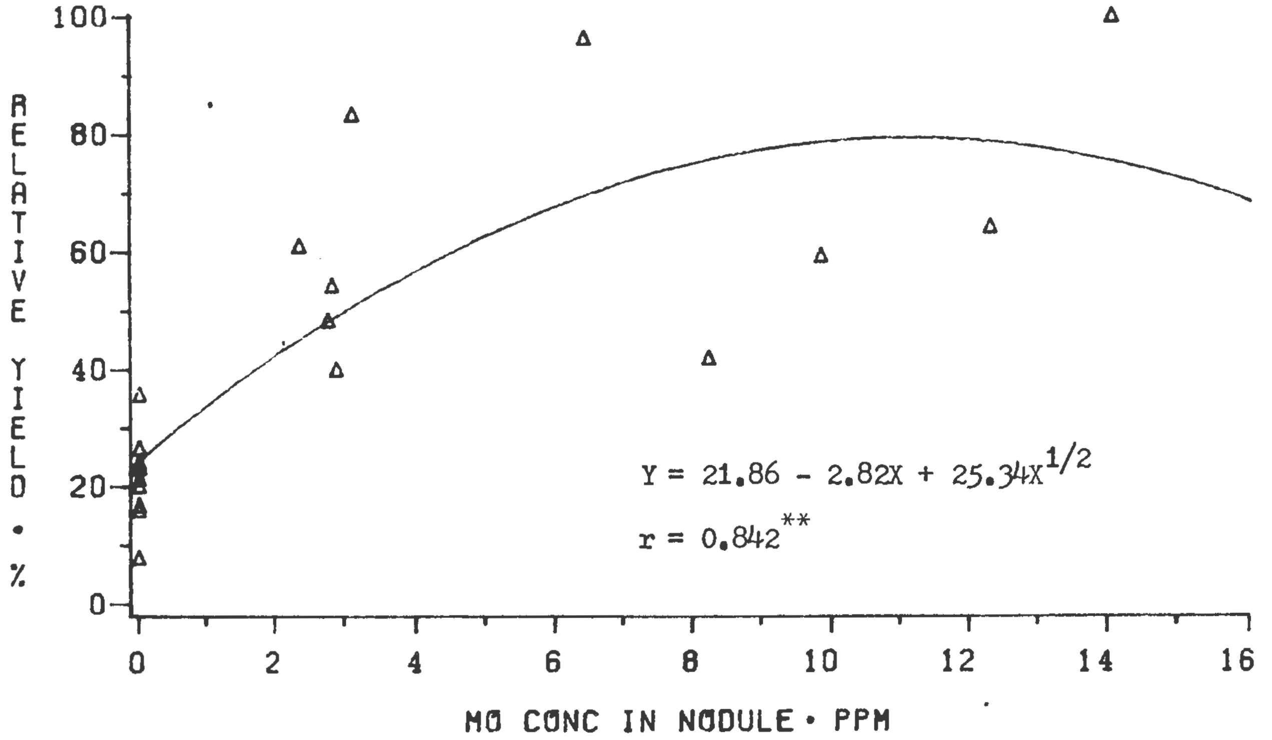


Fig. 1.24. Relationship between relative yield and Mo concentration in nodule of Desmodium intortum grown in the Paaloa soil.

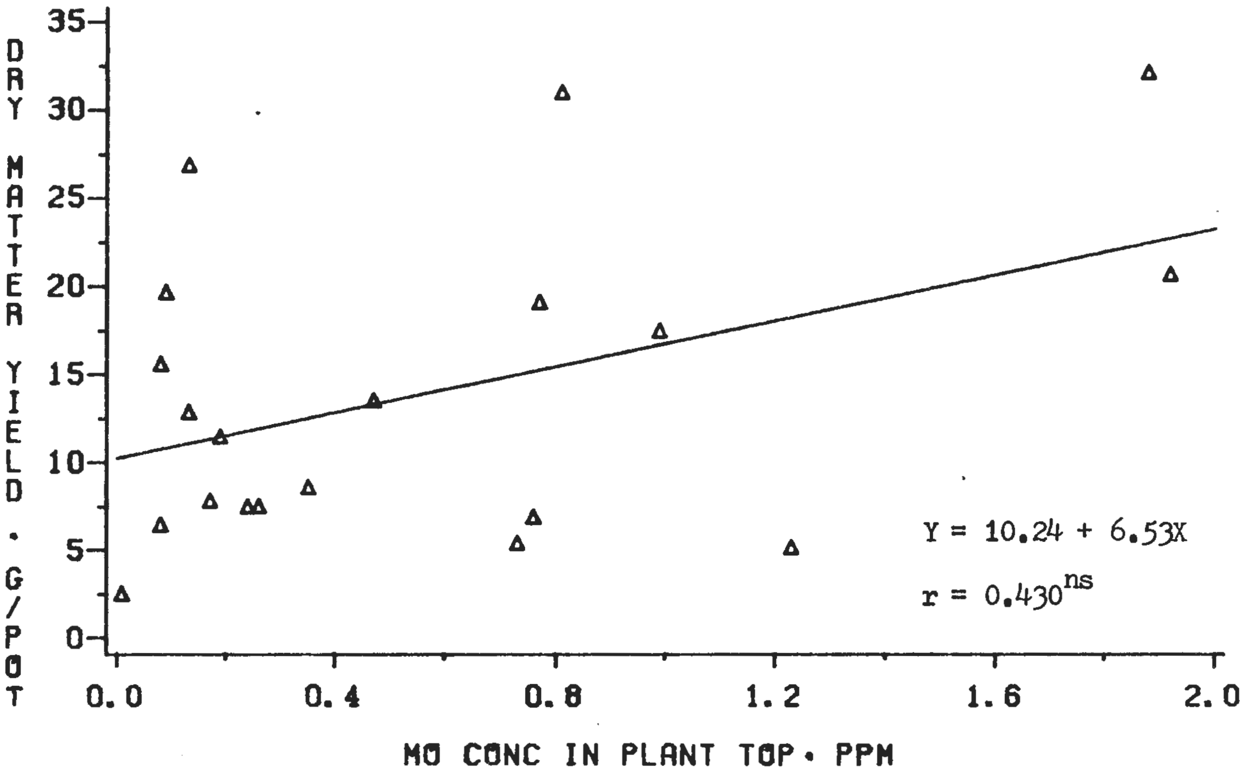


Fig. 1.25. Relationship between dry matter yield and Mo concentration in the top of Desmodium intortum grown in the Paaloa soil.

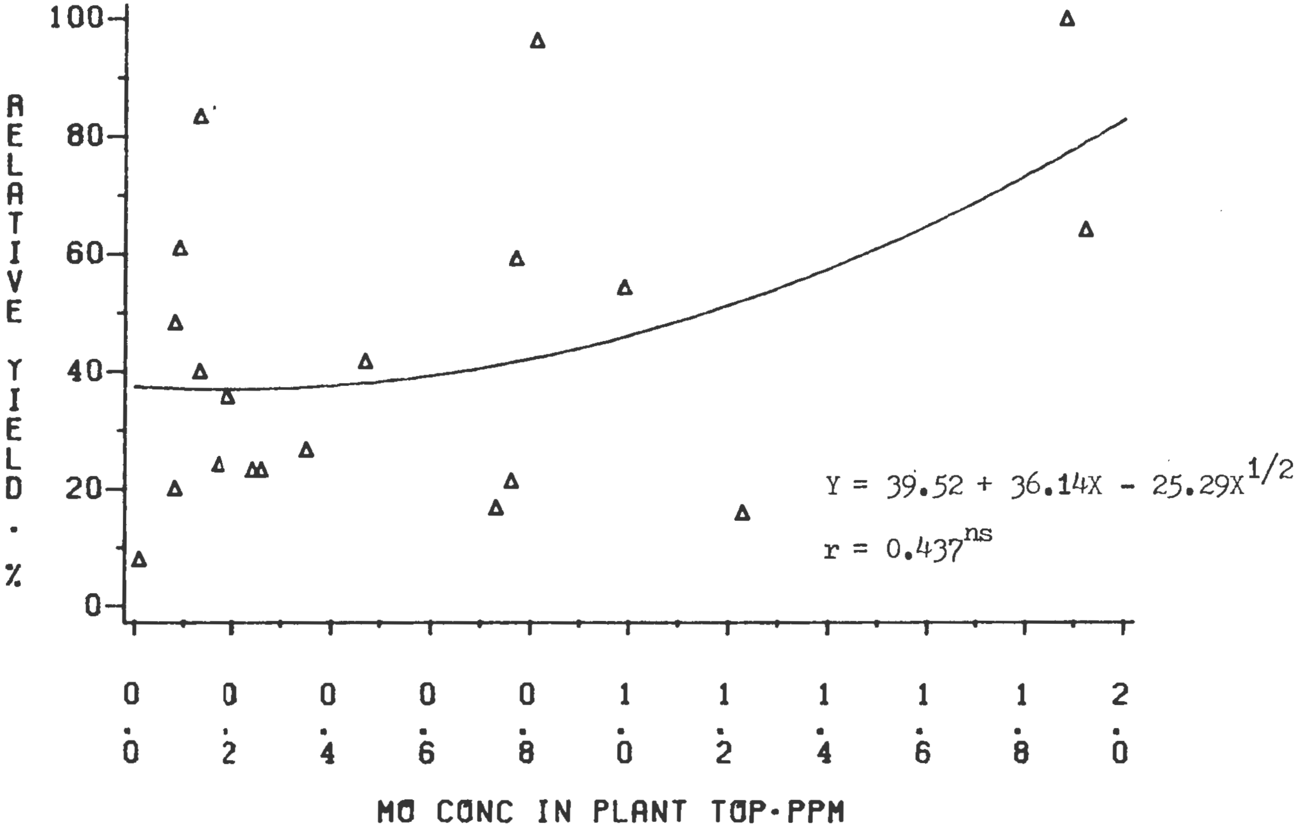


Fig. 1.26. Relationship between relative yield and Mo concentration in the top of <u>Desmodium intortum</u> grown in the Paaloa soil.

Relationship between Dry Matter Yield and Extractable Mo in Soil

There was no significant correlation between dry matter yield and extractable Mo as shown in the scatter diagram (Fig. 1.27). The correlation coefficient of the relationship was 0.176 (p = 0.46). There was also no significant correlation between relative yield and extractable Mo (Fig. 1.28). The correlation coefficient of the relationship was 0.176 (p = 0.46). Lack of the significant correlation between yield and extractable Mo was due to Al toxicity and/or S deficiency. Extractable Mo was significantly increased when rates of Mo application increased (Table 1.41) but yield did not significantly increase when Mo-applied rates were increased.

Relationship between N Concentration in Plant Top and Mo Concentrations

Relationship between N Concentration in Plant Top and Mo Concentration in Nodule

There was a highly significant correlation between N concentration in the top and Mo concentration in nodule of desmodium grown in Paaloa soil (Fig. 1.29). Nitrogen concentration in top increased as Mo concentration in nodule increased. The fact that Mo is required for nitrogenase enzyme responsible for nitrogen fixation is the reason for the increase of N concentration in plant top when Mo concentration in nodule increased.

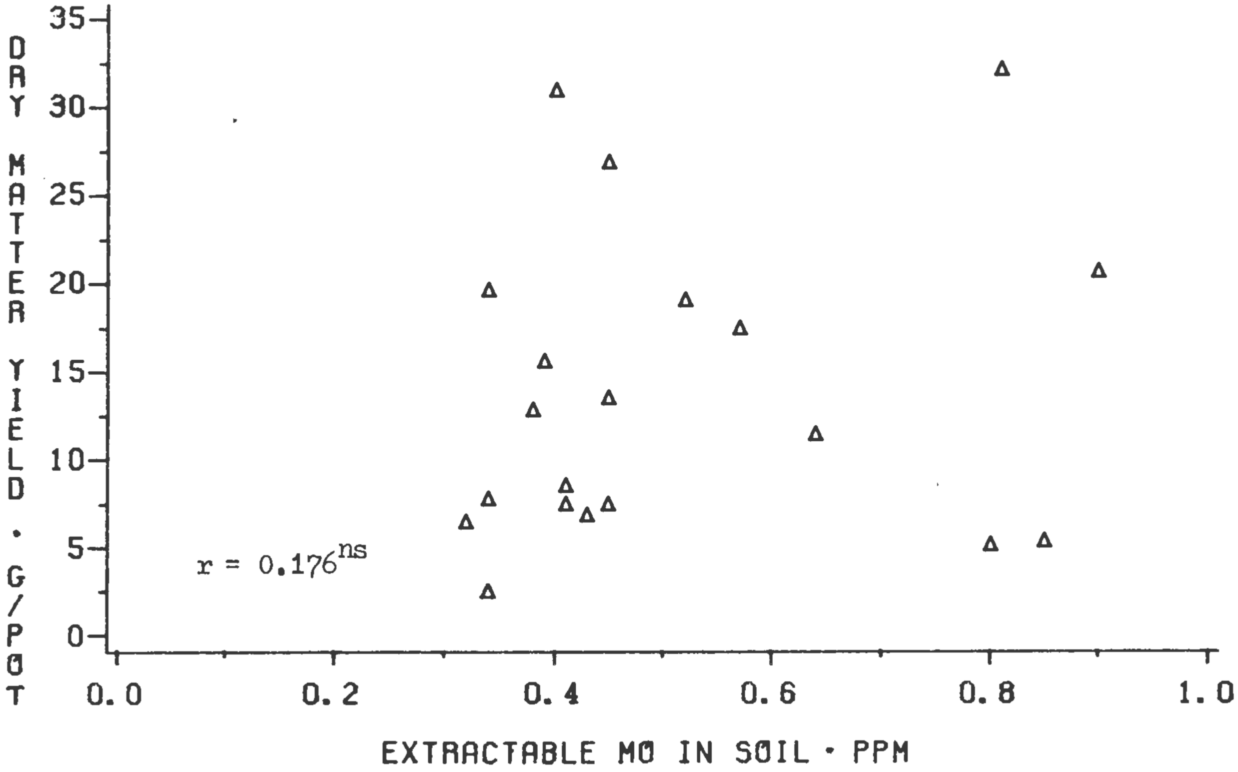


Fig. 1.27. Relationship between dry matter yield of <u>Desmodium intortum</u> and extractable Mo in the Paaloa soil.

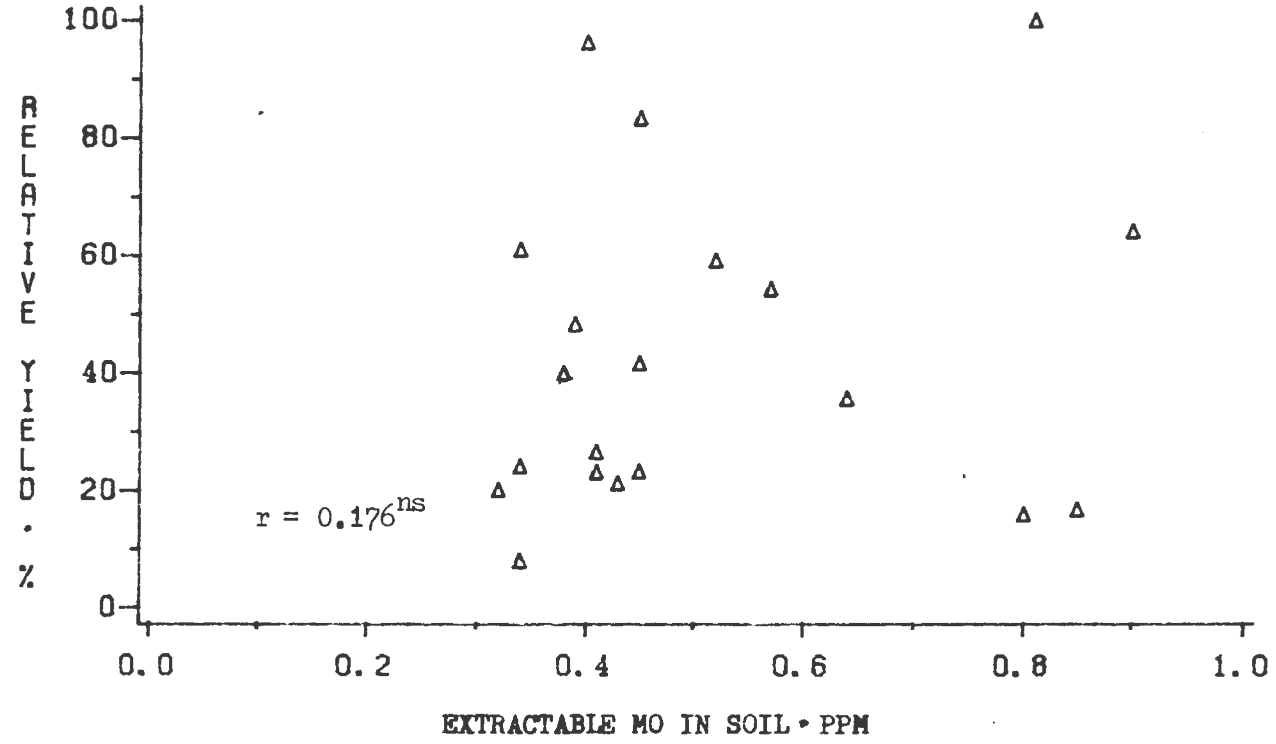


Fig. 1.28. Relationship between relative yield of <u>Desmodium intortum</u> and extractable Mo in the Paaloa soil.

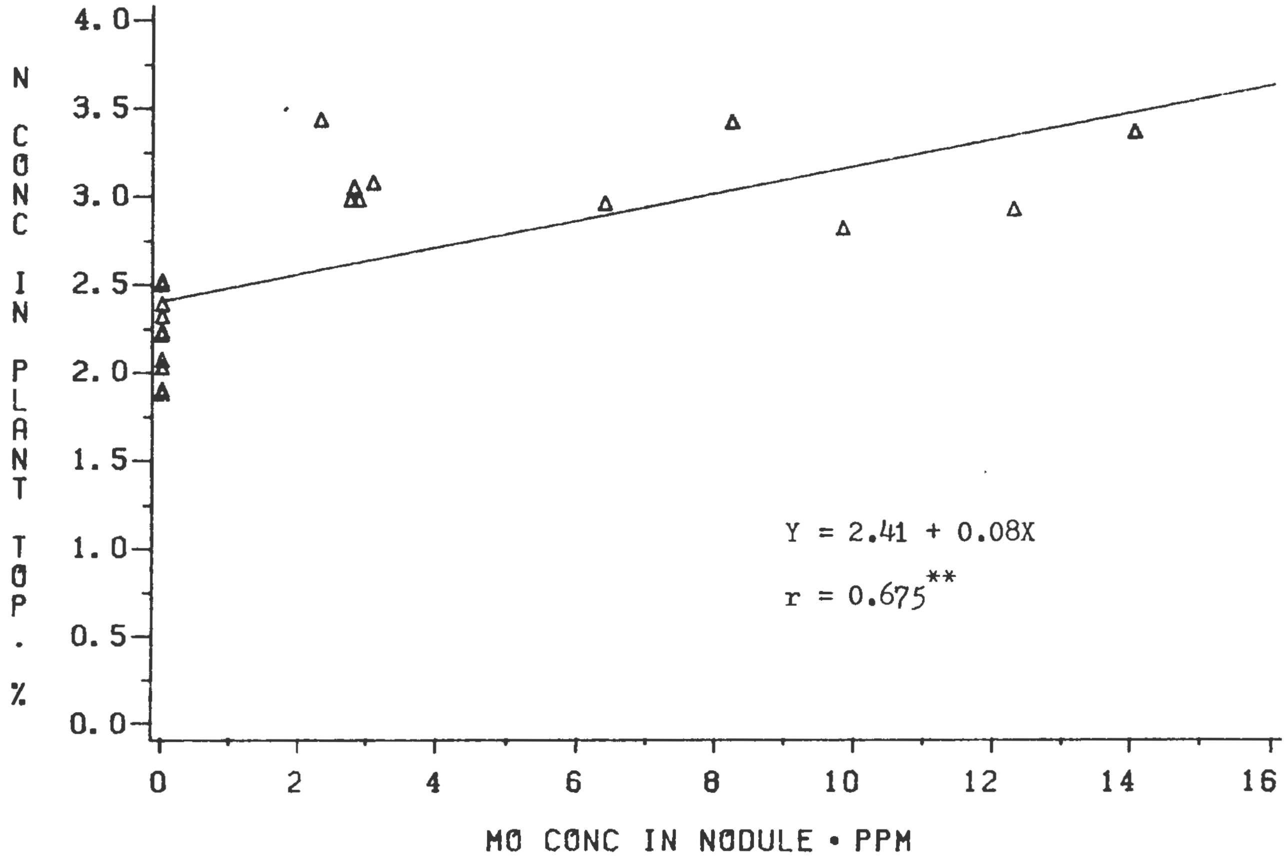


Fig. 1.29. Relationship between N concentration in plant top and Mo concentration in nodule of *Desmodium intortum* grown in the Paaloa soil.

Relationship between N and Mo Concentrations in Plant Top

There was no significant correlation between N and Mo concentration in the top of desmodium (Fig. 1.30). The correlation coefficient was 0.236 ($p = 0.32$). Nitrogen concentration did not increase as Mo concentration increased, suggesting that under condition in which factors other than Mo limit plant growth, N concentration as affected by Mo is likely not correlated with Mo concentration.

Relationship between N Concentration in Plant Top and Extractable Mo in Soil

There was no significant correlation between N concentration in plant top and extractable Mo (Fig. 1.31). The correlation coefficient of the relationship was 0.017 ($p = 0.94$). The lack of a significant correlation was not unexpected because extractable Mo significantly increased when Mo-applied rates increased but N concentration of desmodium grown in unlimed soil did not increase.

Relationship between S Concentration in Plant Top and Mo Concentrations

There was no significant correlation between S and Mo concentrations in plant top. The correlation coefficient of this relationship was 0.129 ($p = 0.32$). There was also no significant correlation between S concentration and extractable Mo in soil. The correlation coefficient of this relationship was 0.034 ($p = 0.80$). However, there was a highly significant correlation between S concentration in plant top and Mo

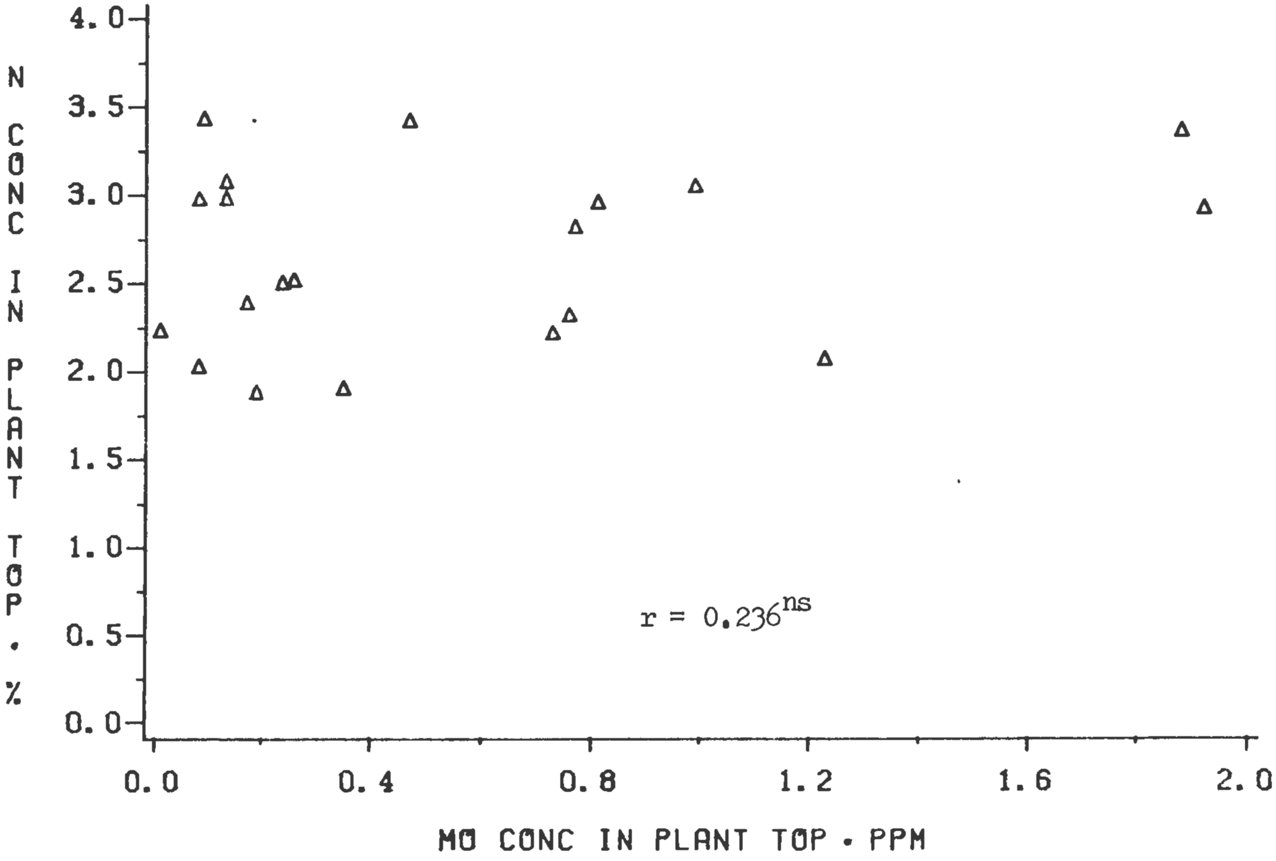


Fig. 1.30. Relationship between N and Mo concentration in the top of <u>Desmodium intortum</u> grown in the Paaloa soil.

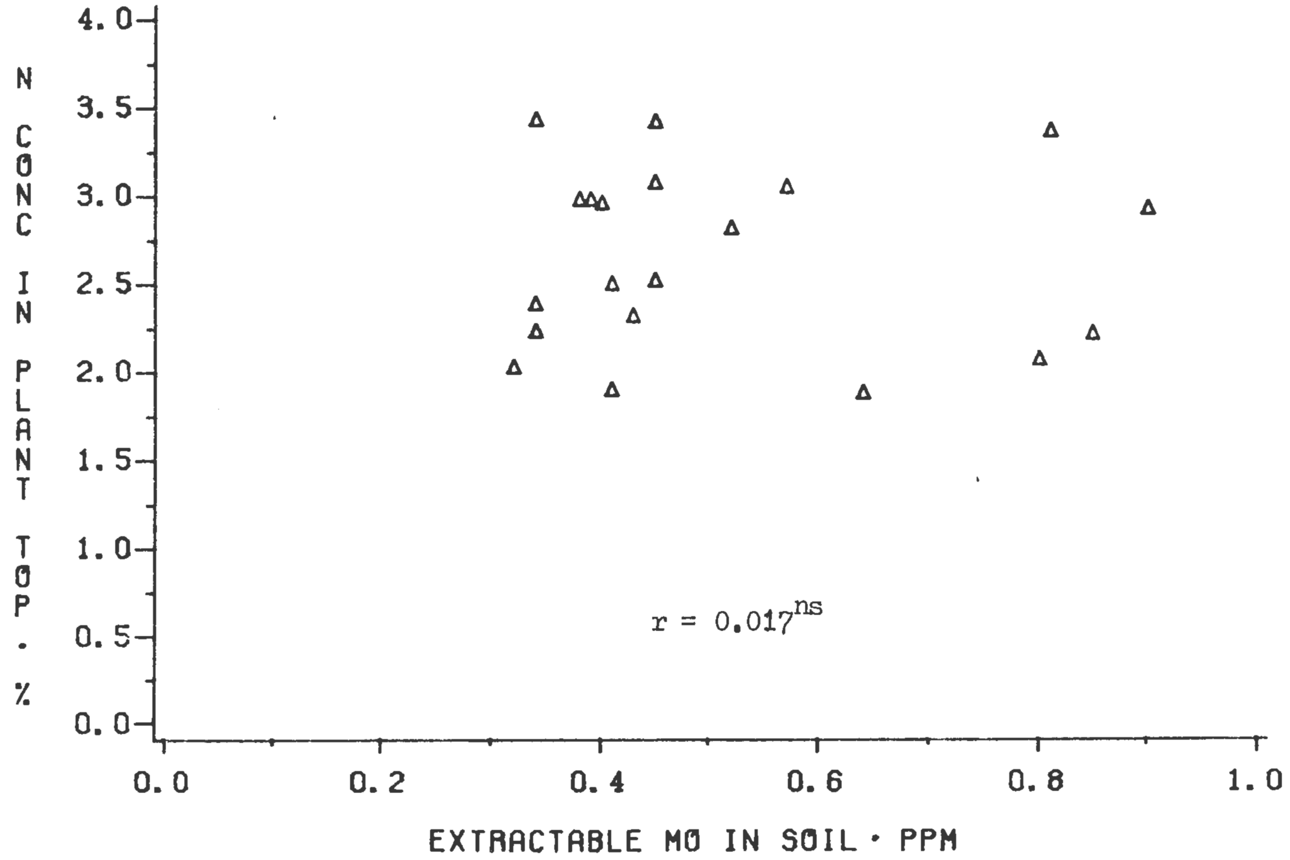


Fig. 1.31. Relationship between N concentration in the top of <u>Desmodium intortum</u> and extractable Mo in the Paaloa soil.

concentration in nodule with a correlation coefficient of 0.611. Sulfur concentration in plant top decreased as Mo concentration in nodule increased (Fig. 1.32). A similar relationship was observed in the earlier experiment on desmodium grown in the Wahiawa soil (Fig. 1.19).

Relationship between Cu Concentration in Plant Top and Mo Concentrations

There was no significant correlation between Cu concentration in plant top and Mo concentration in nodule of desmodium grown in the Paaloa soil. The correlation coefficient of the relationship was 0.441 (r = 0.06). There was also no significant correlation between Cu and Mo concentrations in plant top. The coefficient of the relationship was 0.270 (r = 0.25). There was no significant correlation between Cu concentration in plant top and extractable Mo in soil after harvesting desmodium. The coefficient of the relationship was 0.142 (r = 0.142). The results indicated that there were no relationships between Cu concentration in plant top and Mo concentrations in nodule, in plant top or extractable Mo in soil.

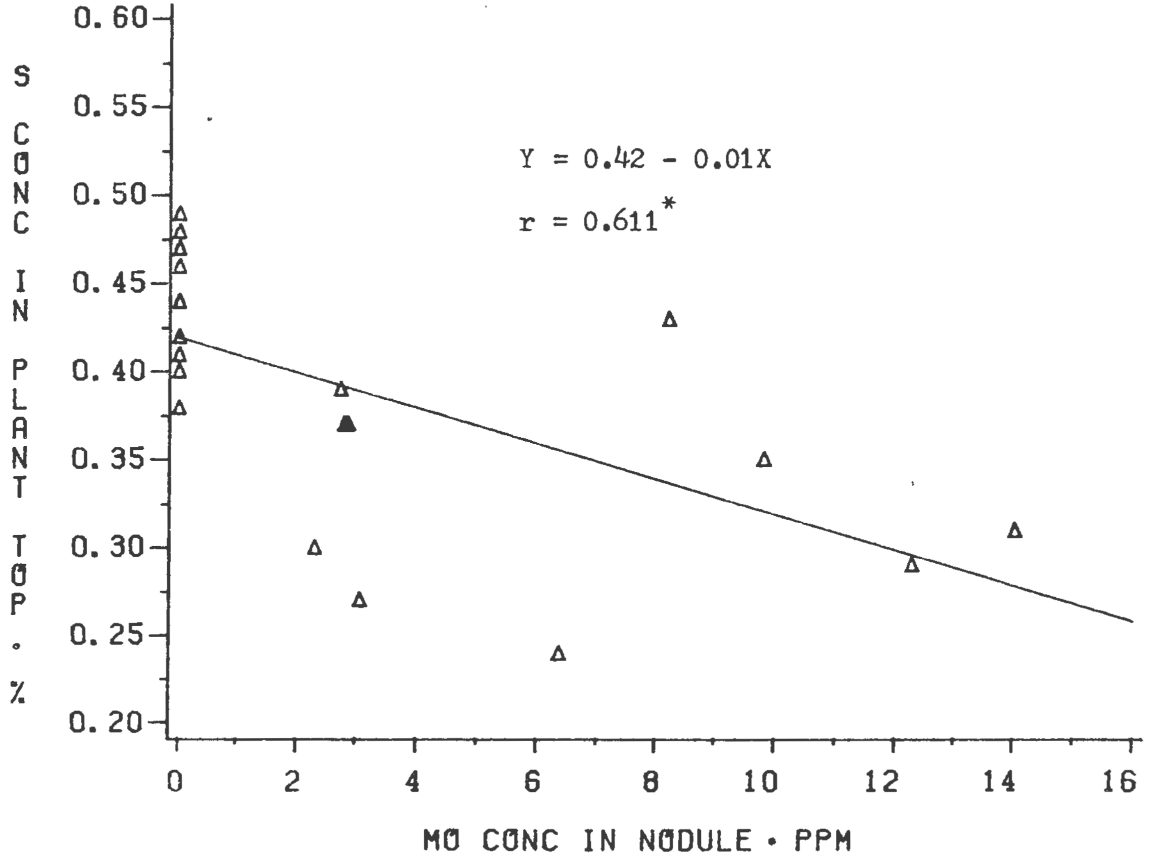


Fig. 1.32. Relationship between S concentration in plant top and Mo concentration in nodule of <u>Desmodium</u> <u>intortum</u> grown in the Paaloa soil.

a.3. Effects of Mo, S and Lime on Growth, Yield and Nutrient Concentration of *Centrosema pubescens* Grown in the Wahiawa Soil

Yield Response

Molybdenum application significantly increased *Centrosema pubescens* dry matter yield (Table 1.42). Centrosema plants receiving no Mo produced significantly lower yield than those receiving various rates of Mo, except the 0.1 kg Mo/ha seed-applied rate. Seed-applied Mo at 0.5 kg Mo/ha produced the highest yield. However, that yield was not significantly higher than yield obtained from other Mo treatments. Soil-applied Mo at both 2.0 and 4.0 kg Mo/ha rates also produced high yields. Seed-applied Mo at 0.5 kg Mo/ha increased yield by 16.5% above the no-Mo treatment whereas soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased yield by 13.2% and 12.9%, respectively. Although the yield obtained from the 0.1 kg Mo/ha seed-applied Mo treatment was not significantly different from that obtained from other Mo treatments, the yield obtained from this treatment was 10.8%, 7.3% and 7.1% lower from that obtained from 0.5 kg Mo/ha seed-applied Mo, 2.0 and 4.0 kg Mo/ha soil-applied Mo treatments, respectively. Dry matter yield obtained from 0.1 kg Mo/ha seed-applied Mo treatment also was not significantly different from that obtained from the no-Mo treatment, suggesting that seed-applied Mo at this rate may not be enough for optimum yield of centrosema grown in the Wahiawa soil. The results indicated that seed-applied Mo at 0.5 kg Mo/ha was as effective as soil-applied Mo at 2.0 and 4.0 kg Mo/ha. The same trend of Mo response has also been observed with *Desmodium intortum* grown in Wahiawa and Paaloa soils at the greenhouse experiments previously conducted.

Table 1.42.

Effects of Mo, S and lime on dry matter yield of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	g/pot				
No Mo added	7.17	9.77	10.07	8.40	8.85 b*
0.1 kg Mo/ha (seed applied)	10.70	9.53	8.00	9.73	9.49 ab
0.5 kg Mo/ha (seed applied)	12.07	10.90	9.63	9.93	10.63 a
2.0 kg Mo/ha (soil applied)	11.03	9.10	10.80	10.03	10.24 a
4.0 kg Mo/ha (soil applied)	12.00	9.20	10.07	9.53	10.20 a
	10.59	9.70	9.71	9.53	
Mean	10.15 a		9.62 a		

	No S	S
Mean	10.15 a	9.61 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Sulfur application did not significantly increase dry matter yield of centrosema (Table 1.42). One explanation for the lack of S response can be offered from the soil test data which indicated that this soil contained 49 ppm available S ($Ca(H_2PO_4)_2$-extractable SO_4-S). Correlation studies by Fox et al. (1964) showed that 6 ppm of available S in the soil was the critical level for S response. In the range of 6 to 10 ppm a response to S might occur and above 10 ppm a response to S was unlikely.

Lime application did not significantly increase dry matter yield of centrosema. However, comparison between lime and no-lime treatment for legumes receiving no Mo and no S revealed that liming increased yield by 28.7% above the no-lime treatment. The result indicated that lime had a tendency to increase yield. The positive effect of lime is likely due to the influence of lime on Mo availability to plant since Mo availability increases as soil pH increases and plant respond very well to Mo application. However, when Mo was applied along with lime, effect of lime on yield became less significant than when Mo was not applied.

It should be noted that the percentage yield increase of centrosema due to Mo was lower than that of desmodium grown in the same soil and receiving the same rates of Mo applied. Centrosema responded to Mo application less than did desmodium. The difference in Mo response of both legumes should be due to their seed size and Mo concentration in the seed. Although desmodium seed contains more Mo concentration (1.08 ppm) than centrosema seed (0.597 ppm), the amount of Mo per seed of desmodium (0.002 ug) is lower than that of centrosema (0.014 ug) since seed size of desmodium is smaller than that of centrosema. Desmodium, therefore, requires more Mo from external sources than centrosema which has a high Mo reserve in the seed.

Nodulation Response

Number of Nodule:

Molybdenum and S application did not significantly increase nodule number of centrosema but liming significantly increased nodule number (Table 1.43). The effect of lime should be due to the fact that lime provides good soil condition for rhizobia multiplication and infection since acidity of soil prevents nodulation by inhibiting the survival or multiplication of rhizobia in soil or the initial build-up of rhizobia populations in the rhizosphere (Mulder and van Veen, 1960; Robson and Loneragan, 1970).

There was a significant interaction between S and lime. The results indicated that when no lime was applied, S tended to increase nodule number but when lime was applied, S tended to decrease nodule number. Although S did not have a significant effect on number of nodules within lime or no-lime treatments, it did have a significant effect between lime and no-lime treatment. Number of nodules obtained from lime applied without S was significantly higher than that obtained from no lime applied with S treatment.

When S fertilizer was not applied it was observed that although there were no significant differences among nodule number of centrosema receiving various rates of Mo without lime, there were significant differences among those of centrosema receiving various rates of Mo with

Table 1.43.

Effects of Mo, S and lime on number of nodule of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	number				
No Mo added	111 e*	141 e	204 de	155 e	153
0.1 kg Mo/ha (seed applied)	85 e	174 de	395 ab	153 e	202
0.5 kg Mo/ha (seed applied)	81 e	96 e	147 e	286 abcd	152
2.0 kg Mo/ha (soil applied)	110 e	221 bcde	428 a	221 bcde	245
4.0 kg Mo/ha (soil applied)	113 e	191 de	265 abcde	391 ab	240
	100 c	164 bc	288 a	241 ab	
Mean	132 b		265 a		
	No S	S			
Mean	194 a	203 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

lime (Table 1.43). The number of nodules of plants grown in limed soil and receiving soil-applied Mo at 2.0 kg Mo/ha without S was higher than that of plants receiving 0.5 kg Mo/ha seed-applied Mo with lime but without S, no Mo without S but with lime, 0.1 kg Mo/ha seed-applied Mo with lime and S, or 2.0 kg Mo/ha soil-applied Mo with S but without lime. It should also be noted that nodule number of centrosema receiving seed-applied Mo without lime and S tended to be less than that of other treatments. The decrease in nodule number was due to the detrimantal effect of Mo salt to rhizobia in the inoculant as reported by many researchers (Giddens, 1964; Burton and Curley, 1966).

Fresh Weight of Nodule:

Molybdenum and S did not have significant effect on nodule fresh weight of centrosema (Table 1.44). However, soil-applied Mo tended to increase nodule fresh weight. Liming significantly increased nodule weight of centrosema. The influence of lime in increasing nodule weight can be considered as a consequence of the effect of lime in increasing nodule number.

Dry Weight of Nodule:

Molybdenum and lime significantly increased nodule dry weight of centrosema (Table 1.45). Sulfur did not significantly increase nodule weight. Nodule weight obtained from centrosema receiving no Mo, and seed-applied Mo at 0.1 and 0.5 kg Mo/ha were not significantly different from each other. Nodule weights of centrosema receiving soil-applied Mo at both rates were not significantly different from each other. However,

Table 1.44.

Effects of Mo, S and lime on nodule fresh weight of _Centrosema pubescens_ grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	g/pot				
No Mo added	1.08	1.25	2.28	1.46	1.52 a*
0.1 kg Mo/ha (seed applied)	0.85	1.23	1.58	0.88	1.14 a
0.5 kg Mo/ha (seed applied)	0.98	0.85	1.42	1.76	1.25 a
2.0 kg Mo/ha (soil applied)	0.81	2.54	2.71	1.82	1.97 a
4.0 kg Mo/ha (soil applied)	1.41	1.03	2.06	2.36	1.72 a
	1.03	1.38	2.47	1.66	
Mean	1.21 b		2.07 a		
	No S		S		
Mean	1.75 a		1.52 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.45.

Effects of Mo, S and lime on nodule dry weight of Centrosema pubescens grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	g/pot				
No Mo added	0.11	0.14	0.30	0.16	0.18 c*
0.1 kg Mo/ha (seed applied)	0.09	0.19	0.36	0.17	0.20 c
0.5 kg Mo/ha (seed applied)	0.15	0.15	0.23	0.41	0.23 bc
2.0 kg Mo/ha (soil applied)	0.16	0.37	0.49	0.30	0.33 ab
4.0 kg Mo/ha (soil applied)	0.26	0.26	0.35	0.32	0.37 a
	0.15	0.22	0.35	0.32	
Mean	0.19 b		0.34 a		

	No S	S
Mean	0.25 a	0.27 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

nodule weights of centrosema receiving both rates of soil-applied Mo were significantly higher than those of centrosema receiving no Mo or 0.1 kg Mo/ha. Nodule dry weight obtained from centrosema receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from that obtained from legume receiving soil-applied Mo at 2.0 kg Mo/ha but was significantly lower than that of legume reveiving 4.0 kg Mo/ha soil application. Influence of Mo in increasing nodule weight was likely because Mo increased plant growth; thus, photosynthates required for nodule growth increased. As a consequence, nodule weight increased.

When averaged over all Mo and S rates, lime significantly increased nodule dry weight by 78.9% above the no lime counterpart. The results suggested that lime provides favorable condition for nodule growth.

Nitrogenase Activity Response

Molybdenum application significantly increased nitrogenase activity of centrosema grown in Wahiawa soil (Table 1.46). Nitrogenase activity of legume receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than that of legume receiving no Mo. However, nitrogenase activity of legume receiving soil-applied Mo at 4.0 kg Mo/ha was not significantly higher than that of legume receiving soil-applied Mo at 2.0 kg Mo/ha and seed-applied Mo at 0.5 kg Mo/ha. Although nitrogenase activity of centrosema receiving no Mo was 34.0%, 166%, and 199% lower than those of centrosema receiving 0.1 kg Mo/ha and 0.5 kg Mo/ha seed-applied Mo and 2.0 kg Mo/ha soil-applied Mo, respectively, the differences were not statistically significant. Sulfur had no effect on nitrogenase activity. Liming tended to increase nitrogenase activity since nitrogenase

Table 1.46.

Effects of Mo, S and lime on nitrogenase activity of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	nmole C_2H_2/plant/hr				
No Mo added	0.19	0.24	91.47	112.70	26.15 b*
0.1 kg Mo/ha (seed applied)	0.23	0.31	102 20	37.38	35.03 b
0.5 kg Mo/ha (seed applied)	26.59	96.72	26.15	128.33	69.45 ab
2.0 kg Mo/ha (soil applied)	0.45	15.08	262 13	35.35	78.25 ab
4.0 kg Mo/ha (soil applied)	173.73	136.82	57.24	160.58	132.09 a
	40.24	49.84	107.84	74.88	
Mean	45.04 a		91.36 a		
	No S	S			
Mean	74.04 a	62.36 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

activity of centrosema grown in limed soil was 103% higher than that of legume grown in unlimed soil.

Influence of Mo, S and Lime on Some Chemical Compositions in Plant

Nitrogen:

Molybdenum application tended to increase N concentration in the top of centrosema although there was no significant difference among N concentrations of plants receiving various rates of Mo (Table 1.47). Lime had little effect on N concentration in plant top. Sulfur application significantly decreased N concentration in plant top. However, it should be noted that N concentration was low, suggesting that some factors other than Mo may have limited nitrogen fixation.

Phosphorus:

Phosphorus concentration in the top of centrosema receiving seed-applied Mo at 0.5 kg Mo/ha was significantly lower than that in plants receiving no Mo (Table 1.48). However, the decrease was only 8.1%. Phosphorus concentration in plants receiving seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at 2.0 and 4.0 kg Mo/ha rates as well as that in plants receiving no Mo was not significantly different. Phosphorus in the top of plants receiving seed-applied Mo at 0.5 kg Mo/ha was also not significantly different from that of plants receiving other rates of Mo application. The reason that P concentration of plants

Table 1.47.

Effects of Mo, S and lime on N concentration in the top of *Centrosema pubescens* grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	1.27	1.40	1.64	1.30	1.40 a*
0.1 kg Mo/ha (seed applied)	1.66	1.31	1.39	1.39	1.44 a
0.5 kg Mo/ha (seed applied)	1.58	1.49	1.58	1.33	1.50 a
2.0 kg Mo/ha (soil applied)	1.71	1.40	1.40	1.43	1.48 a
4.0 kg Mo/ha (soil applied)	1.51	1.39	1.41	1.63	1.49 a
	1.55	1.40	1.48	1.42	
Mean	1.48 a		1.45 a		

	No S	S
Mean	1.52 a	1.41 b

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.48.

Effects of Mo, S and lime on P concentration in the top of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	%				
No Mo added	0.23	0.24	0.25	0.24	0.24 a*
0.1 kg Mo/ha (seed applied)	0.21	0.22	0.26	0.25	0.24 ab
0.5 kg Mo/ha (seed applied)	0.21	0.22	0.23	0.23	0.22 b
2.0 kg Mo/ha (soil applied)	0.21	0.24	0.24	0.23	0.23 ab
4.0 kg Mo/ha (soil applied)	0.21	0.22	0.24	0.25	0.23 ab
	0.21	0.23	0.24	0.24	
Mean	0.22 b		0.24 a		

	No S	S
Mean	0.23 a	0.23 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

receiving seed-applied Mo at 0.5 kg Mo/ha was lower than that of plant receiving no Mo was due to a "dilution effect" since dry matter yield of plant receiving seed-applied Mo at this rate was significantly higher than that of plants receiving no Mo.

Sulfur had little effect on P concentration in plant top but liming significantly increased P concentration. Lime rendered P in soil to be more available to plant since P adsorption by soil decreased when lime was applied. Interaction among Mo, S and lime on P concentration was not observed in this experiment.

Potassium:

Molybdenum, S or lime had no significant effect on K concentration in the top of centrosema since K concentrations of plants receiving various rates of Mo, S and lime were not significantly different from each other (Table 1.49). No interaction among Mo, S and lime on K concentration was observed.

Calcium:

Molybdenum and S had no significant effect on Ca concentration in the top of centrosema (Table 1.50). The effect of Mo on Ca concentration in centrosema was slightly different from that in desmodium since Ca concentration in desmodium was decreased when Mo was applied. This difference must be due to the difference of legume species in Ca adsorption and relative growth rate regarding Mo response. Liming significantly increases Ca concentration in plant top. This result was expected since lime provides Ca for plants as well as its influence in soil pH.

Table 1.49.

Effects of Mo, S and lime on K concentration in the top of Centrosema pubescens grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	----------	----------	%	----------	----------
No Mo added	1.23	1.21	1.25	1.21	1.22 a*
0.1 kg Mo/ha (seed applied)	1.30	1.12	1.18	1.21	1.20 a
0.5 kg Mo/ha (seed applied)	1.17	1.21	1.32	1.20	1.23 a
2.0 kg Mo/ha (soil applied)	1.34	1.16	1.22	1.24	1.24 a
4.0 kg Mo/ha (soil applied)	1.19	1.17	1.23	1.26	1.21 a
	1.25	1.17	1.24	1.22	
Mean	1.21 a		1.23 a		

	No S	S
Mean	1.25 a	1.20 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.50.

Effects of Mo, S and lime on Ca concentration in the top of <u>Centrosema pubescens</u> grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	%				
No Mo added	1.07	1.18	1.39	1.17	1.20 a*
0.1 kg Mo/ha (seed applied)	1.01	1.17	1.47	1.36	1.25 a
0.5 kg Mo/ha (seed applied)	1.15	1.04	1.25	1.31	1.19 a
2.0 kg Mo/ha (soil applied)	1.07	1.13	1.29	1.31	1.20 a
4.0 kg Mo/ha (soil applied)	1.03	1.16	1.22	1.34	1.19 a
	1.07	1.14	1.32	1.30	
Mean	1.11 b		1.31 a		
	No S		S		
Mean	1.20 a		1.22 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Magnesium:

Magnesium concentration in the top of plants receiving soil-applied Mo at 2.0 kg Mo/ha was significantly higher than that of plants receiving no Mo, seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at 4.0 kg Mo/ha but was not significantly higher than that of plants receiving seed-applied Mo at 0.5 kg Mo/ha (Table 1.51). Magnesium concentrations in the top of plants receiving no Mo, seed-applied Mo at both rates, and soil-applied Mo at 4.0 kg Mo/ha were not significantly different from each other.

Sulfur had no significant effect on Mg concentration in plant top suggesting that there was no influence of S on Mg absorption by plant. Although the difference was small, liming significantly decreased Mg concentration in plant top (Table 1.51). This effect was due to the influence of lime on Mg absorption. This result was similar to that reported by Andrew and Johnson (1976) that an increase in Ca resulted in a marked decrease in Mg concentration in plant top of some tropical and temperate pasture legumes.

Sulfur:

Molybdenum had no significant effect on S concentration in plant top (Table 1.52). Sulfur application also did not increase S concentration in plant top. The reason that S application did not increase S concentration was due to an adequate supply of S in the soil for plant need. It has been indicated by Bouma (1975) that when S is present at levels higher than necessary for maximum growth, there is little or no

Table 1.51.

Effects of Mo, S and lime on Mg concentration in the top of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	%				
No Mo added	0.20	0.20	0.20	0.19	0.20 b*
0.1 kg Mo/ha (seed applied)	0.23	0.20	0.20	0.23	0.22 b
0.5 kg Mo/ha (seed applied)	0.24	0.24	0.22	0.19	0.23 ab
2.0 kg Mo/ha (soil applied)	0.28	0.25	0.23	0.24	0.25 a
4.0 kg Mo/ha (soil applied)	0.22	0.21	0.18	0.20	0.16 b
	0.24	0.22	0.21	0.21	
Mean	0.23 a		0.21 b		
	No S	S			
Mean	0.22 a	0.22 a			

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.52.

Effects of Mo, S and lime on S concentration in the top of *Centrosema pubescens* grown in the Wahiawa soil.

Mo treatment	No lime No S	No lime S	Lime No S	Lime S	Mean
	----	----	%	----	----
No Mo added	0.17	0.16	0.18	0.16	0.17 a*
0.1 kg Mo/ha (seed applied)	0.16	0.17	0.20	0.19	0.18 a
0.5 kg Mo/ha (seed applied)	0.18	0.16	0.18	0.18	0.18 a
2.0 kg Mo/ha (soil applied)	0.19	0.17	0.18	0.18	0.18 a
4.0 kg Mo/ha (soil applied)	0.16	0.17	0.17	0.18	0.17 a
	0.17	0.17	0.18	0.18	
Mean	0.17 b		0.18 a		

	No S	S
Mean	0.178 a	0.174 a

---

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

further uptake at high level. However, he also noted that some reports showed a continued uptake. So the different effects of S fertilizers on S uptake by plants suggest that the relationship between S applied and S concentration in plants varied with species and ontogenic changes. He also pointed out that although "luxury uptake" of sulfate, in the sense of uptake in excess of the plant requirements, does occur, most of the evidences indicate that this is not as pronounced as for other nutrient elements.

Liming significantly increased S concentration in plant top. The increase in S concentration with application of lime most likely resulted from increased S availability in soil due to a higher soil pH. Elskins and Ensminger (1971) have reported that sorbed sulfate not readily available to plants becomes increasingly available as the pH of the soil is increased.

Iron:

Molybdenum,S and lime had little effect on Fe concentration in plant top (Table 1.53). Although there was a tendency that lime and S applications decreased Fe concentration in plant top, such difference was not significant.

Manganese:

Manganese concentration in plants receiving soil-applied Mo at 4.0 kg Mo/ha was significantly lower than that in plants receiving soil-applied Mo at 2.0 kg Mo/ha and seed-applied Mo at 0.5 kg Mo/ha (Table 1.54). Manganese concentration in plants receiving soil-applied

Table 1.53.

Effects of Mo, S and lime on Fe concentration in the top of Centrosema pubescens grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	--------	--------	%	--------	--------
No Mo added	176	158	163	167	166 a*
0.1 kg Mo/ha (seed applied)	167	160	165	166	165 a
0.5 kg Mo/ha (seed applied)	237	171	169	159	184 a
2.0 kg Mo/ha (soil applied)	186	171	172	177	177 a
4.0 kg Mo/ha (soil applied)	160	160	165	164	162 a
	185	164	167	167	
Mean	175 a		167 a		

	No S	S
Mean	176 a	166 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.54.

Effects of Mo, S and lime on Mn concentration in the top of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	285	291	243	223	261 ab*
0.1 kg Mo/ha (seed applied)	305	265	235	219	256 ab
0.5 kg Mo/ha (seed applied)	357	307	232	237	283 a
2.0 kg Mo/ha (soil applied)	322	282	262	240	277 a
4.0 kg Mo/ha (soil applied)	289	251	227	214	245 b
	312	280	240	227	
Mean	296 a		233 b		

	No S	S
Mean	276 a	253 b

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Mo at 4.0 kg Mo/ha, however, was not significantly lower than that of plants receiving no Mo and seed-applied Mo at 0.1 kg Mo/ha.

Sulfur application significantly decreased Mn concentration in plant top. This result suggested that there might be an interaction between S and Mn regarding Mn absorption by centrosema. Liming significantly decreased Mn concentration in plant top since lime rendered soil Mn unavailable to plants. This is a beneficial effect of lime because it alleviates Mn toxicity which usually occurs in plants grown in acid soil containing high amount of Mn. However, Mn toxicity symptom was not observed in this experiment and Mn concentration in plant top was not high. The reason that Mn toxicity did not occur was due to the fact that pH of the Wahiawa soil used in this experiment (5.2) was high enough to decrease Mn availability in soil.

Zinc:

Molybdenum and S had no significant effect on Zn concentration in the top of centrosema (Table 1.55). Liming significantly decreased Zn concentration in plant top. The influence of lime in decreasing Zn concentration is due to the fact that Zn availability in soil was suppressed by liming. Interactive effect of Mo, S and lime on Zn concentration in plant top was observed in this experiment. When seed-applied Mo at 0.1 kg Mo/ha and no lime were applied, application of S tended to increase Zn concentration. However, when the same Mo treatment and lime were applied, application of S decreased Zn concentration. When seed-applied Mo at 0.5 kg Mo/ha and no lime were applied, S application decreased Zn concentration but when the same Mo rate and lime were applied,

Table 1.55.

Effects of Mo, S and lime on Zn concentration in the top of *Centrosema pubescens* grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	28.0	27.7	23.3	16.7	23.9 a*
0.1 kg Mo/ha (seed applied)	23.3	29.3	26.7	23.3	25.7 a
0.5 kg Mo/ha (seed applied)	32.0	23.0	20.3	22.0	24.3 a
2.0 kg Mo/ha (soil applied)	29.0	26.7	24.0	21.0	25.3 a
4.0 kg Mo/ha (soil applied)	24.7	24.0	21.0	24.0	23.4 a
	27.4	26.1	23.1	21.4	
Mean	26.8 a		22.3 b		

	No S	S
Mean	25.3 a	23.8 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

S application increased Zn concentration (Table 1.55). The Zn concentration in plants supplied with seed-applied and soil-applied Mo within S and lime treatments varied and the trend of variation was different from treatment to treatment.

Silicon:

Molybdenum application significantly increased Si concentration in plant top (Table 1.56). Silicon concentrations in plants receiving seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at 2.0 and 4.0 kg Mo/ha rates were significantly higher than those in plants receiving no Mo. However Si concentration in plants receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly higher than that in plants receiving no Mo. Sulfur had no significant effect on Si concentration in plant top. Liming significantly decreased Si concentration in plant top. The effect of lime in decreasing Si concentration in the top of centrosema was similar to that observed with desmodium grown in Wahiawa soil at the earlier experiment conducted previously (Table 1.17).

Copper:

There were no significant differences among Cu concentrations in plants receiving various rates of Mo (Table 1.57). Sulfur or lime also had no significant effect on Cu concentrations in the top of centrosema. However, when averaged over all Mo and S rates, lime tended to increase Cu concentration.

Table 1.56.

Effects of Mo, S and lime on Si concentration in the top of Centrosema pubescens grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	----------	----------	%	----------	----------
No Mo added	0.36	0.36	0.26	0.24	0.30 b*
0.1 kg Mo/ha (seed applied)	0.37	0.45	0.32	0.31	0.36 a
0.5 kg Mo/ha (seed applied)	0.43	0.40	0.27	0.27	0.34 ab
2.0 kg Mo/ha (soil applied)	0.40	0.44	0.34	0.29	0.37 a
4.0 kg Mo/ha (soil applied)	0.41	0.40	0.32	0.30	0.36 a
	0.39	0.41	0.30	0.28	
Mean	0.40 a		0.29 b		
	No S	S			
Mean	0.35 a	0.35 a			

---

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.57.

Effects of Mo, S and lime on Cu concentration in the top of Centrosema pubescens grown in the Wahiawa soil.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	ppm				
No Mo added	6.33	7.00	9.33	9.67	8.08 a*
0.1 kg Mo/ha (seed applied)	6.00	8.67	10.33	7.67	8.17 a
0.5 kg Mo/ha (seed applied)	4.33	4.33	9.00	8.00	6.42 a
2.0 kg Mo/ha (soil applied)	5.33	3.69	7.33	8.33	6.17 a
4.0 kg Mo/ha (soil applied)	11.67	6.00	6.00	8.33	8.00 a
	6.73	5.93	8.40	8.40	
Mean	6.33 a		8.40 a		
	No S		S		
Mean	7.57 a		7.17 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Aluminum:

Aluminum concentration in the top of desmodium receiving no Mo was significantly lower than that of plants receiving soil-applied Mo at 2.0 kg Mo/ha but was not significantly lower than in other Mo treatments (Table 1.58). Aluminum concentrations in centrosema receiving various rates of Mo were not significantly different from each other. The results suggested that legumes supplied with Mo tended to accumulate more Al than those supplied with no Mo. The reason for this effect may partly be due to the ability of legumes to absorb a large amount of Al when their growth was increased by Mo application.

Sulfur had no significant effect on Al concentration although Al concentration in the top of centrosema tended to decrease when S was applied. Although it has been established that lime alleviates Al toxicity of plant (Kamprath, 1970; Adams, 1981), liming did not significantly decrease Al concentration in plant top. However, there was no Al toxicity symptom was observed. Wahiawa soil used in this experiment contains low Al (Table 1.1) so Al toxicity might not be a problem for legumes grown in this soil.

Influences of Mo, S and Lime on Mo Concentrations in **Plant and Soil**

Molybdenum Concentration in Nodule

Application of Mo by both seed coating and soil treatment significantly increased Mo concentration in nodules of centrosema (Table 1.59). Molybdenum concentration in noduled of plants receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than those of plants receiving

Table 1.58.

Effects of Mo, S and lime on Al concentration in the top of *Centrosema pubescens* grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	221	218	176	220	209 b*
0.1 kg Mo/ha (seed applied)	400	226	267	245	285 ab
0.5 kg Mo/ha (seed applied)	247	219	215	209	223 ab
2.0 kg Mo/ha (soil applied)	245	266	377	338	306 a
4.0 kg Mo/ha (soil applied)	254	313	341	341	287 ab
	273	248	275	251	
Mean	261 a		263 a		

	No S	S
Mean	274 a	249 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.59.

Effects of Mo, S and lime on Mo concentration in nodule of *Centrosema pubescens* grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	2.27	2.10	3.02	3.82	2.80 d*
0.1 kg Mo/ha (seed applied)	4.10	4.79	4.40	5.77	4.77 c
0.5 kg Mo/ha (seed applied)	5.54	4.32	5.65	5.27	5.20 c
2.0 kg Mo/ha (soil applied)	5.67	5.99	6.84	6.90	6.35 b
4.0 kg Mo/ha (soil applied)	7.57	7.07	7.60	7.89	7.53 a
	5.03	4.86	5.50	5.93	
Mean	4.95 b		5.72 a		

	No S	S
Mean	5.27 a	5.40 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

no Mo or other rates of Mo. Molybdenum concentration in nodules of plants receiving soil-applied Mo at 2.0 kg Mo/ha was significantly higher than in those of plants receiving seed-applied Mo at both rates or no Mo. Molybdenum concentration in plants receiving seed-applied Mo at both rates were not significantly different from each other. Molybdenum concentration in nodules of plants receiving seed-applied Mo at both rates were significantly higher than in those of plant receiving no Mo. Molybdenum concentration in nodules of plant receiving soil-applied Mo was significantly higher than in those of plants receiving seed-applied Mo. The reason that Mo concentration in nodules of plant receiving soil-applied Mo was higher than in those of plants receiving seed-applied Mo was due to the high rate of soil-applied Mo.

Sulfur fertilizer had little effect on Mo concentration in nodules of centrosema but lime significantly increased Mo concentration. Molybdenum concentration in nodules of plants grown in limed soil was increased 15.5% above that in plants grown in unlimed soil. Liming increased soil pH and consequently increased Mo availability in soil. No interaction among Mo, S and lime on Mo concentration in nodule of centrosema was observed.

Molybdenum Concentration in Plant Top

Application of Mo fertilizer by soil treatment at both rates significantly increased Mo concentration in the top of centrosema grown in the Wahiawa soil (Table 1.60). Molybdenum concentration in the tops of plants receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than in those of plants receiving soil-applied Mo at 2.0 kg Mo/ha,

Table 1.60.

Effects of Mo, S and lime on Mo concentration in the top of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Mo treatment	No lime		Lime		Mean
	No S	S	No S	S	
	ppm				
No Mo added	0.06	0.07	0.19	0.50	0.21 c*
0.1 kg Mo/ha (seed applied)	0.21	0.23	0.38	0.33	0.29 c
0.5 kg Mo/ha (seed applied)	0.17	0.23	0.44	0.70	0.38 c
2.0 kg Mo/ha (soil applied)	0.18	0.27	0.96	1.45	0.71 b
4.0 kg Mo/ha (soil applied)	0.79	0.67	1.59	1.49	1.14 a
	0.28	0.29	0.71	0.89	
Mean	0.29 b		0.80 a		
	No S		S		
Mean	0.50 a		0.59 a		

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

seed-applied Mo at both rates, or no Mo. Mo concentration in the top of plants receiving soil-applied Mo at 2.0 kg Mo/ha was also higher than in those of plant receiving seed-applied Mo at both rates or no Mo. Molybdenum concentrations in plants receiving seed-applied Mo at both rates and no Mo were not significantly different from each other. It was observed that Mo concentration in the top of centrosema was relatively high as compared to that of desmodium grown in the same soil (Table 1.19). High Mo concentration in the top of centrosema may likely be due to the high Mo concentration in the seed (0.597 ppm). Molybdenum concentration in the seed has a significant effect on the Mo concentration in plant top since Mo in cotyledons are first translocated mainly to stems (Ishizuka, 1982). Molybdenum are then stored in stems before translocated to roots and nodules. It might be assumed that Mo concentration in the top of large-seed legumes may be higher than in that of small-seed legumes. Molybdenum reserved in seed could have an influence on the response of plant to Mo fertilizer.

Sulfur had little effect on Mo concentration in plant top. Lime significantly increased Mo concentration in the top. The influence of lime in increasing Mo concentration in plant top was due to the effect of lime in decreasing Mo adsorption in soil and, as a consequence, Mo availability increased.

Extractable Mo in Soil

Extractable Mo in soil with soil-applied Mo at both rates were significantly higher than those in soil with seed-applied Mo at both rates and no Mo (Table 1.61). Extractable Mo in soil with soil-applied Mo at

Table 1.61.

Effects of Mo, S and lime on extractable Mo in the Wahiawa soil after harvesting _Centrosema_ _pubescens_.

	No lime		Lime		
Mo treatment	No S	S	No S	S	Mean
	ppm				
No Mo added	0.37	0.58	0.53	0.58	0.52 c*
0.1 kg Mo/ha (seed applied)	0.49	0.61	0.61	0.60	0.58 c
0.5 kg Mo/ha (seed applied)	0.60	0.61	0.52	0.67	0.60 c
2.0 kg Mo/ha (soil applied)	0.79	1.19	0.62	0.98	0.90 b
4.0 kg Mo/ha (soil applied)	1.32	1.70	1.13	1.29	1.36 a
	0.71	0.94	0.68	0.82	
Mean	0.82 a		0.75 a		

	No S	S
Mean	0.70 b	0.88 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

4.0 kg Mo/ha was significantly higher than that in soil with soil-applied Mo at 2.0 kg Mo/ha. Extractable Mo in soil with seed-applied Mo at both rates and no Mo were not significantly different from each other.

Sulfur application significantly increased extractable Mo in soil after harvesting. This result suggested that there was likely an interaction between sulfate and molybdate in soil. However, Gonzalez et al. (1974) reported that sulfate ions did not compete with molybdate ions for the adsorption sites on clay surface. It should also be noted that S application did not increase extractable Mo in soil after harvesting desmodium grown in the same soil. Rhizosphere activity of different legume species may play a role in the nutrient status of soil.

When averaged over all Mo and S rates, there was no significant difference between extractable Mo in limed and unlimed soils. However, an interaction between Mo and lime on extractable Mo was observed in this experiment. For the no-Mo and seed-applied Mo at 0.1 kg Mo/ha treatments, liming increased extractable Mo (Table 1.61). For seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at both rates, liming decreased extractable Mo. It should be noted that Mo concentrations in nodule and in the tops of plants receiving high rates of Mo along with lime were much higher than in those of plants receiving no Mo and seed-applied Mo at 0.1 kg Mo/ha (Table 1.59, 1.60). Low extractable Mo observed after harvesting centrosema from 0.5 kg Mo/ha seed-applied, 2.0 and 4.0 kg Mo/ha soil-applied treatments, along with lime, might result from high absorption of Mo by the legume.

Relationship between Mo Concentrations in Plant and Soil

Relationship between Mo Concentration in Plant Top and Mo Concentration in Nodule

There was a highly significant correlation between Mo concentration in plant top and Mo concentration in nodule. The relationship fitted a quadratic curve with a correlation coefficient of 0.840 (Fig. 1.33). At low Mo concentration in nodule, Mo concentration in plant top was also low indicating that Mo did not accumulate in the plant top when nodule still need much Mo for nitrogenase activity. As the Mo concentration in nodules increased, the Mo concentration in plant tops increased exponentially, suggesting that when Mo concentration in nodules was adequate for nitrogenase activity the excess Mo was transferred to the plant top. So plant top was the sink of Mo when high rates of Mo were applied. The pattern of relationship between Mo concentration of nodule and plant top of centrosema was similar to that of desmodium grown in Wahiawa and Paaloa soils in the greenhouse experiments conducted earlier. This similarity of both legumes suggested that the distribution and translocation of Mo within the whole plant of different pasture legume species could be the same.

Relationship between Mo Concentration in Plant Top and Extractable Mo in soil

There was a highly significant correlation between Mo concentration in the top of centrosema and extractable Mo in soil after harvesting with a correlation coefficient of 0.569 (Fig. 1.34). Molybdenum

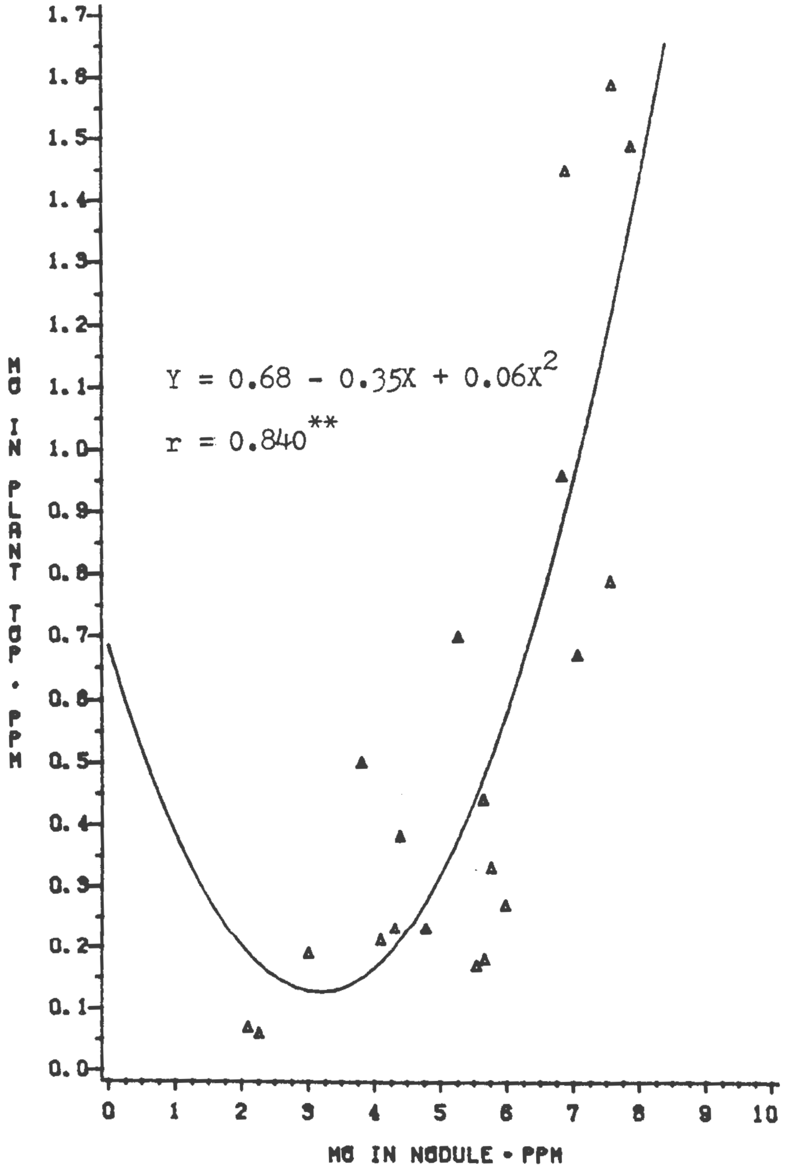


Fig. 1.33. Relationship between Mo concentration in plant top and Mo concentration in nodule of *Centrosema pubescens* grown in the Wahiawa soil.

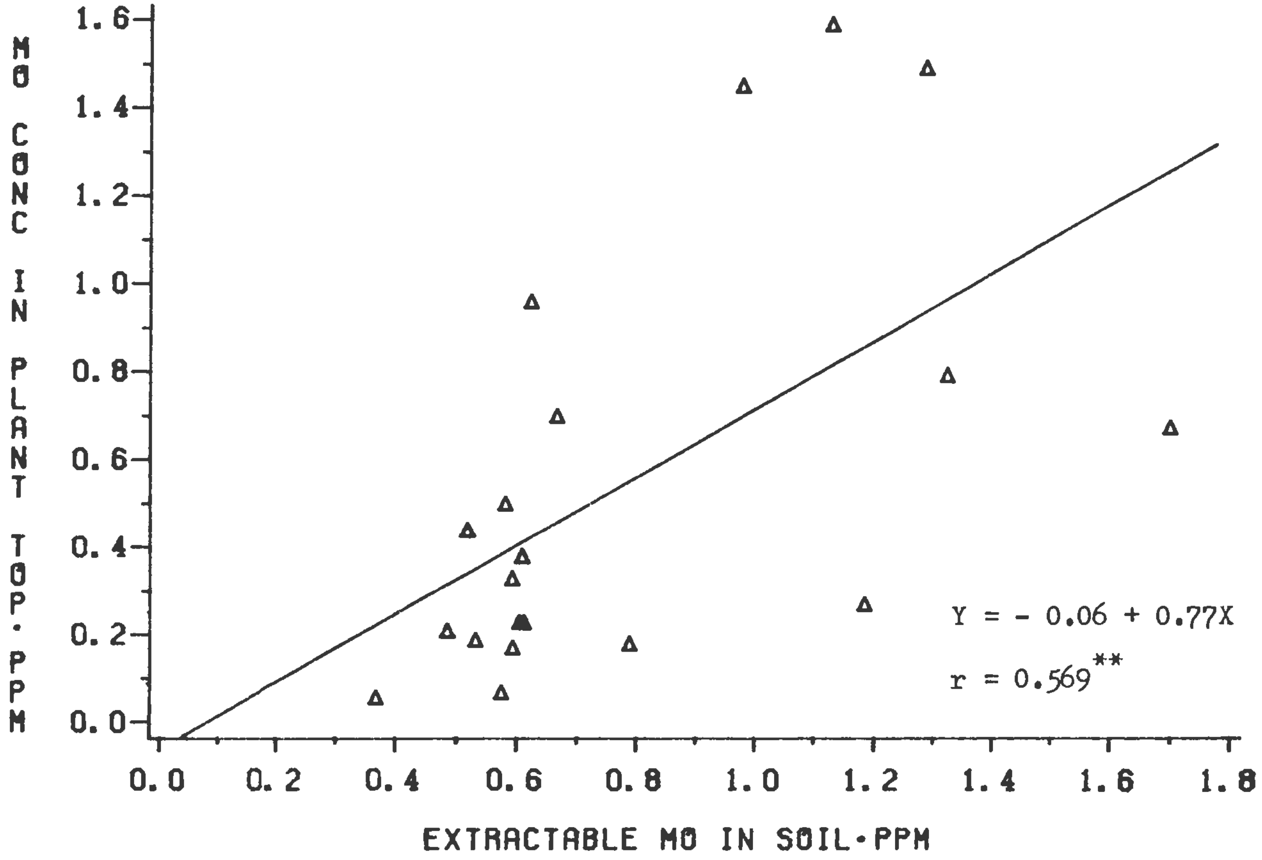


Fig. 1.34. Relationship between Mo concentration in the top of <u>Centrosema pubescens</u> and extractable Mo in the Wahiawa soil.

concentration in plant top increased as extractable Mo increased.

Relationship between Mo Concentration in Nodule and Extractable Mo in soil

Threr was a highly significant correlation between Mo concentration in nodule and extractable Mo in soil with a correlation coefficient of 0.817 (Fig. 1.35). Molybdenum concentration in nodule increased as extractable Mo increased. According to a scatter diagram, Mo concentration in nodule highly increased when extractable Mo increased from 0.2 to 0.8 ppm. The rate of increase in nodule Mo concentration declined as extractable Mo was higher than 0.8 ppm, suggesting that excess Mo concentration in nodule was translocated to plant top.

Relationship between Dry Matter Yield and Mo Concentrations

Relationship between Dry Matter Yield and Mo Concentration in Nodule

There was no significant correlation between dry matter yield and Mo concentration in nodules. (Fig. 1.36). Despite the lack of significance, some definite trends are evident. Dry matter yield tended to increase as the Mo concentration in nodule increased. The high yield was observed when Mo concentration in nodule was in the range of 5 to 8 ppm.

When nodule Mo concentrations were plotted against relative yields in a scatter diagram (Fig. 1.37), there was no significant correlation between relative yield and Mo concentration in nodules. Critical Mo concentration in nodule associated with 80% maximum yield was approximately 4.2 ppm.

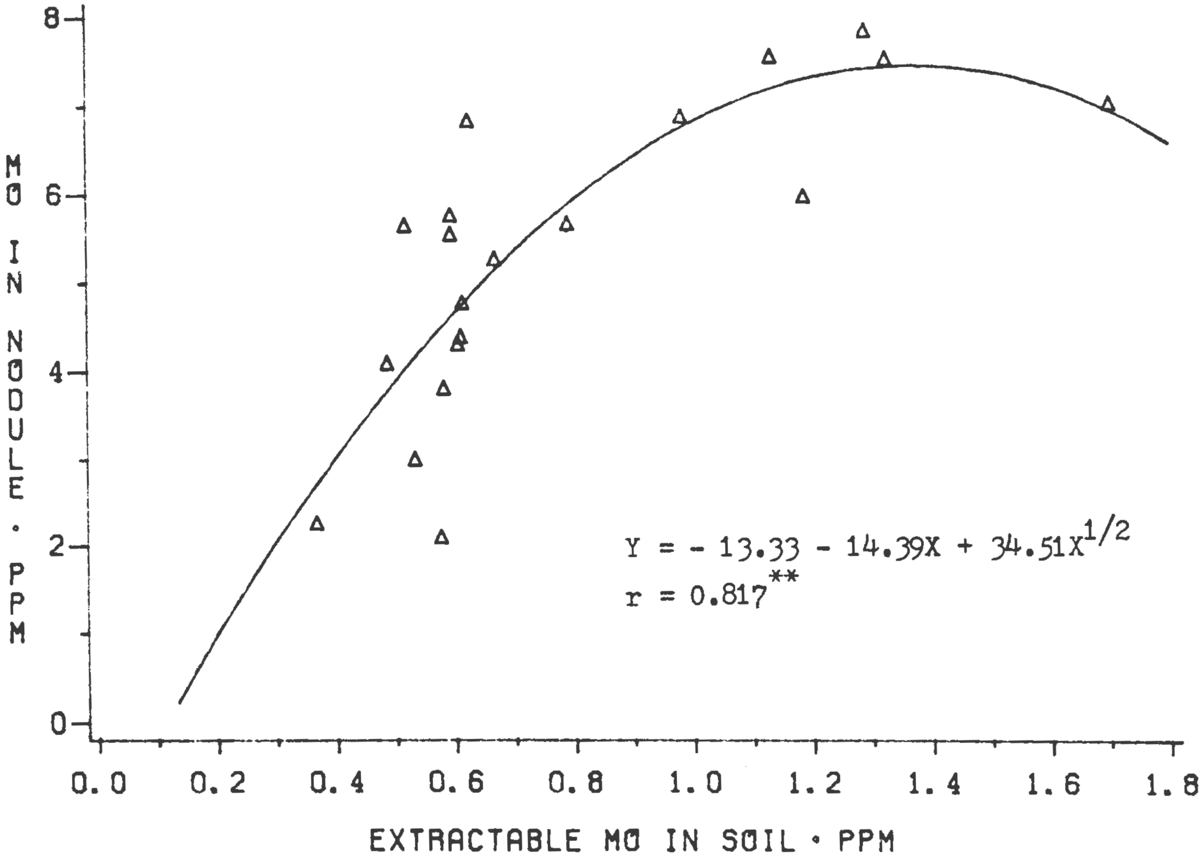


Fig. 1.35. Relationship between Mo concentration in nodule of *Centrosema pubescens* and extractable Mo in the Wahiawa soil.

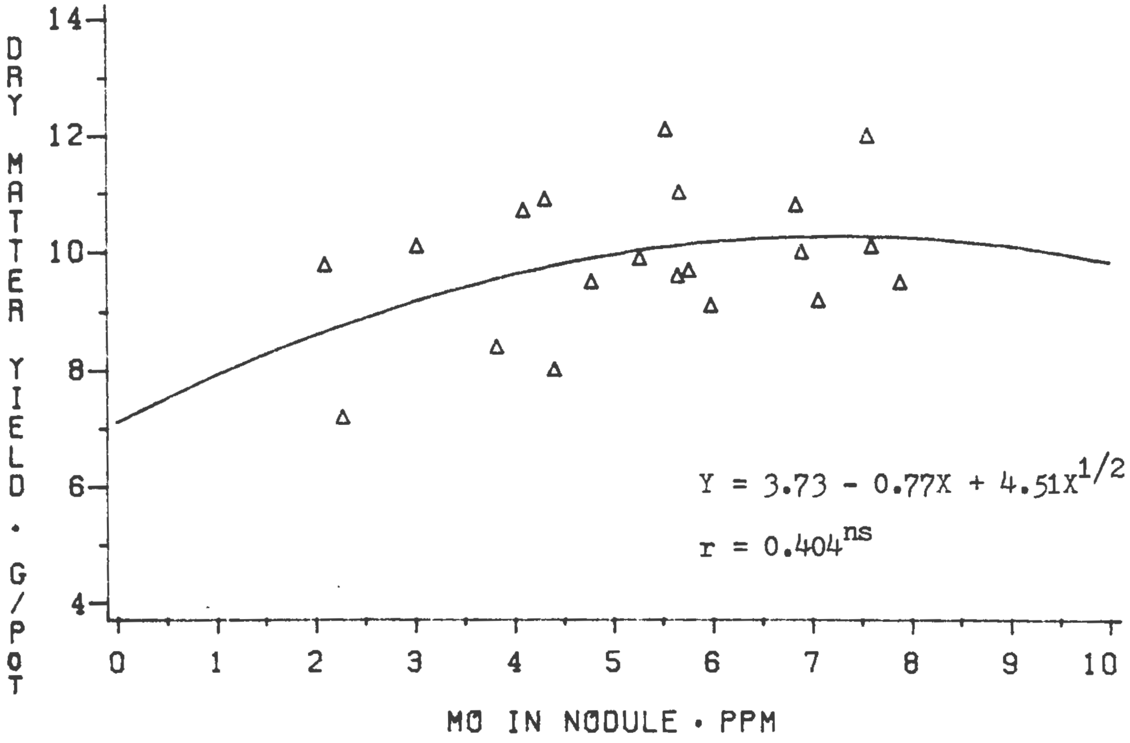


Fig. 1.36. Relationship between dry matter yield and Mo concentration in nodule of Centrosema pubescens grown in the Wahiawa soil.

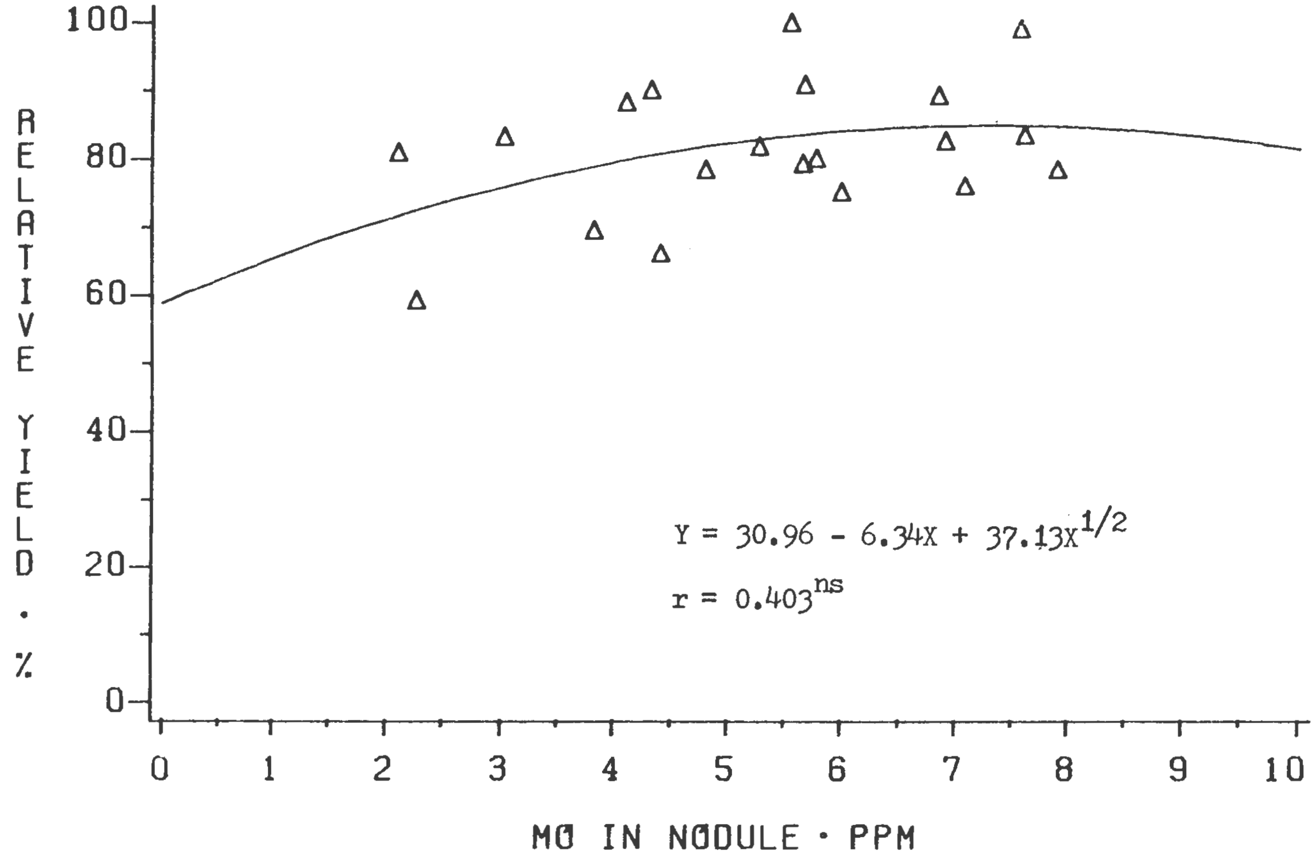


Fig. 1.37. Relationship between relative yield and Mo concentration in nodule of <u>Centrosema</u> <u>pubescens</u> grown in the Wahiawa soil.

Relationship between Dry Matter Yield and Mo Concentration in Plant Top

There was no significant correlation between dry matter yield and Mo concentration in plant top (Fig. 1.38). Lack of significant correlation between yield and Mo concentration suggest that centrosema is less responsive to Mo than desmodium. Although Mo concentration in plant top of centrosema was as high as desmodium, the increment of increase in yield of centrosema as affected by Mo application was less than that of desmodium (Table 1.2 and 1.42). Dry matter yield of legumes usually depends on nitrogen fixation of nodule which requires Mo. Molybdenum concentration in nodule should therefore be more correlated with dry matter yield than Mo concentration in plant top.

When Mo concentrations in plant tops were plotted against relative yields in a scatter diagram (Fig. 1.39), there was no significant correlation between relative yield and Mo concentration in plant top. However, it may be approximated that Mo concentration in the top be at least 0.2 ppm for optimum growth of centrosema.

Relationship between Dry Matter yield and Extractable Mo in soil

There was no significant correlation between dry matter yield and extractable Mo in soil after harvesting (Fig. 1.40). There was also no significant difference between relative yield and extractable Mo in soil (Fig. 1.41). However, it may be approximated that at least 0.4 ppm extractable Mo is required for optimum growth of centrosema.

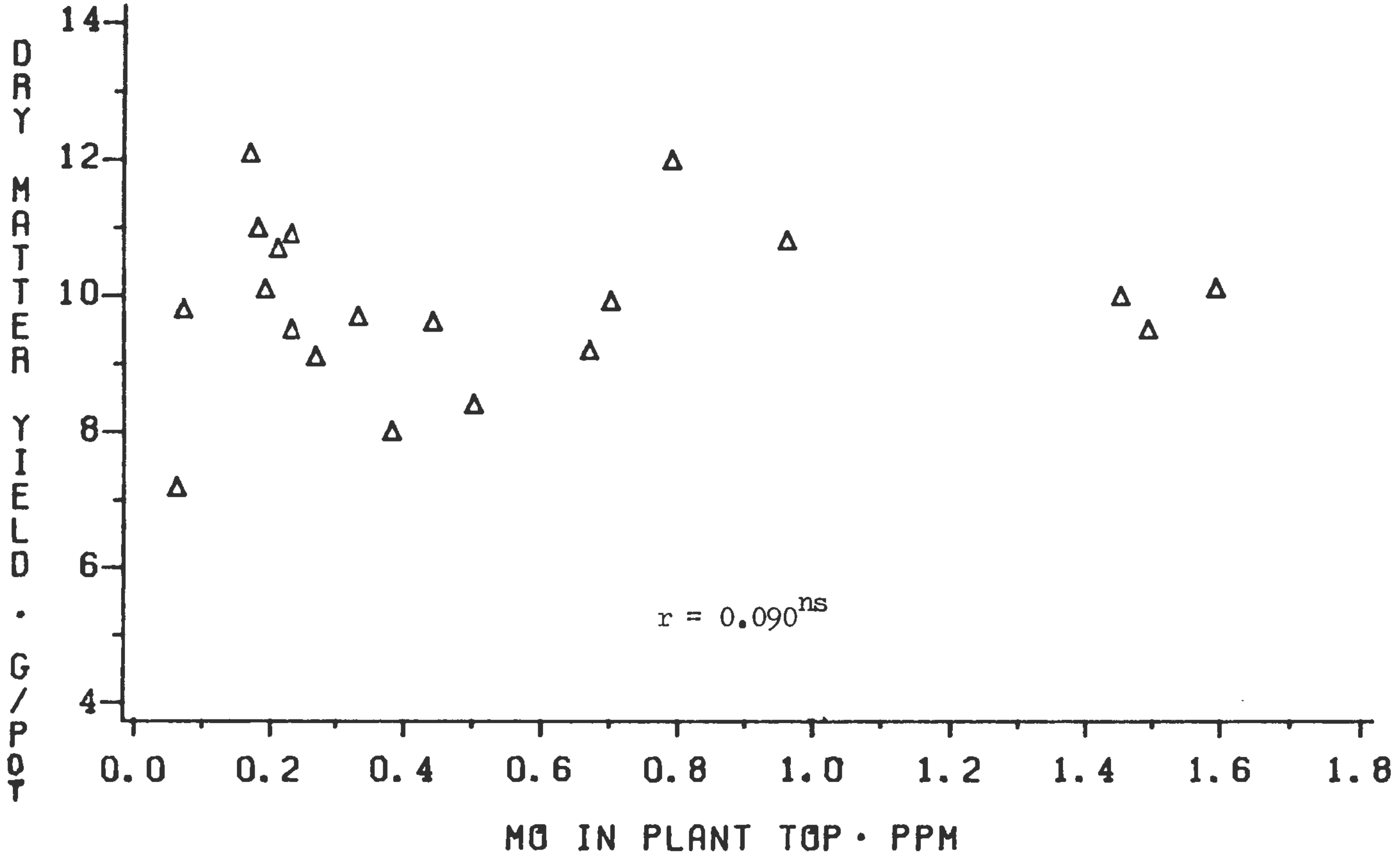


Fig. 1.38. Relationship between dry matter yield and Mo concentration in the top of *Centrosema pubescens* grown in the Wahiawa soil.

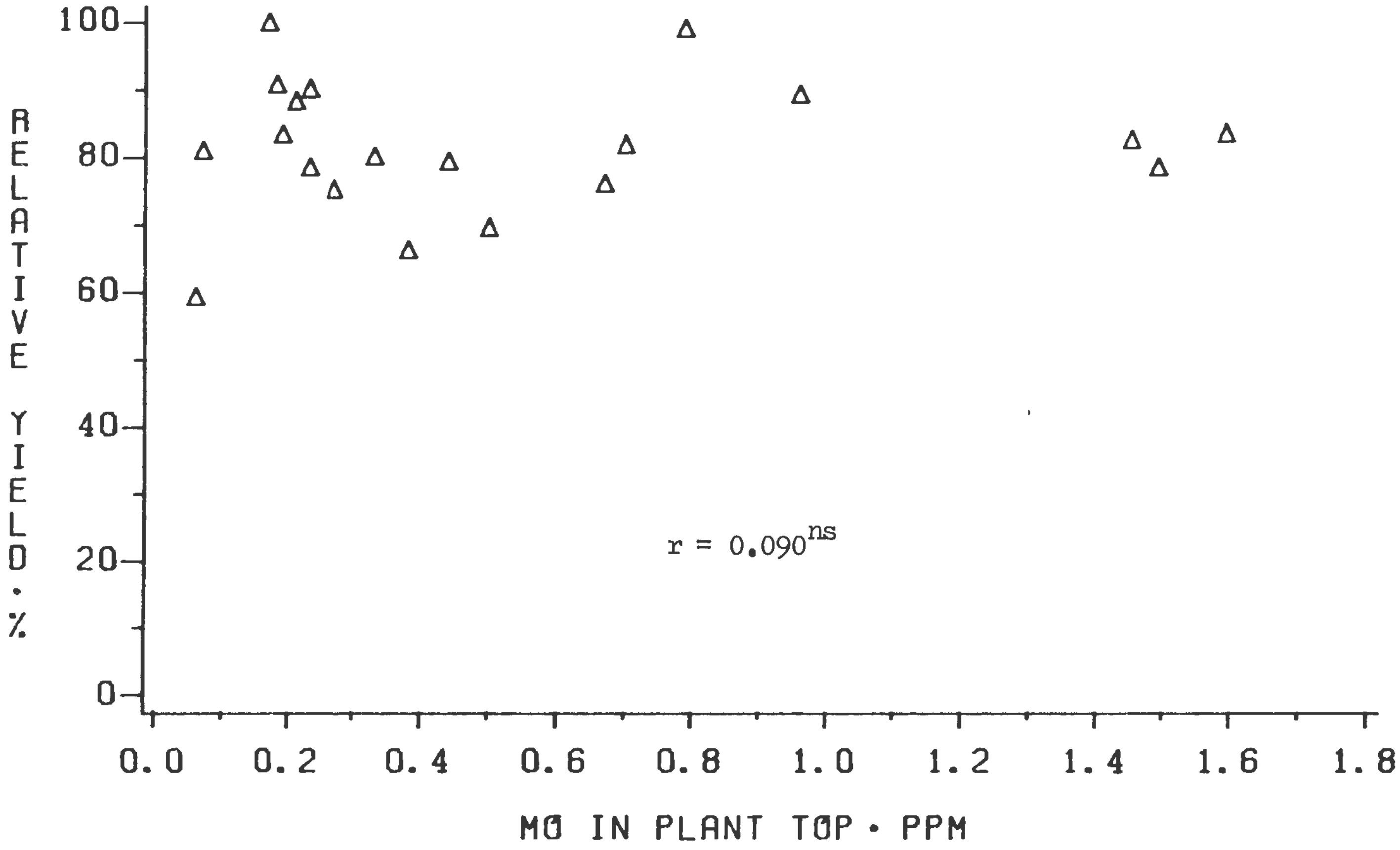


Fig. 1.39. Relationship between relative yield and Mo concentration in the top of Centrosema pubescens grown in the Wahiawa soil.

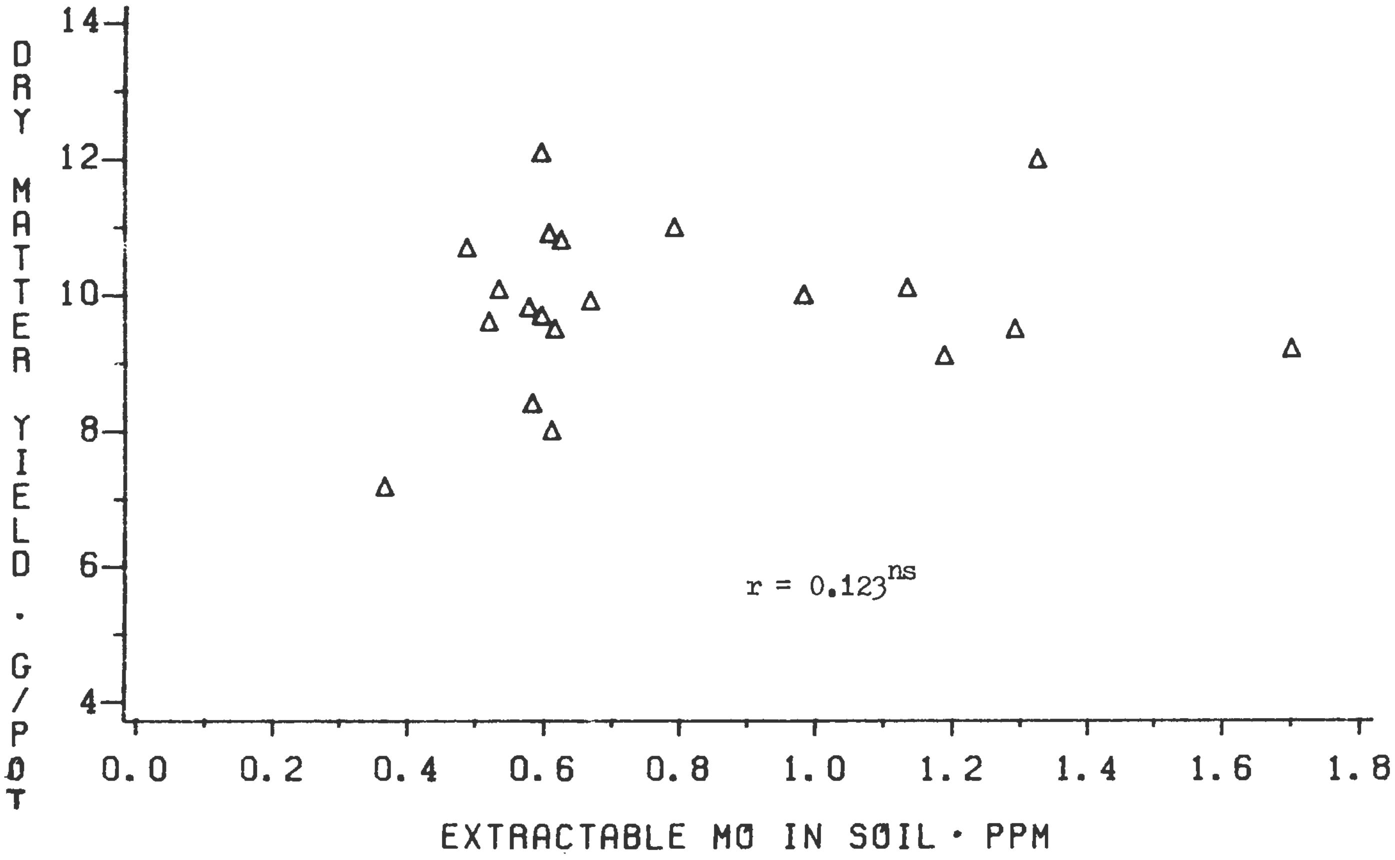


Fig. 1.40. Relationship between dry matter yield of Centrosema pubescens and extractable Mo in the Wahiawa soil.

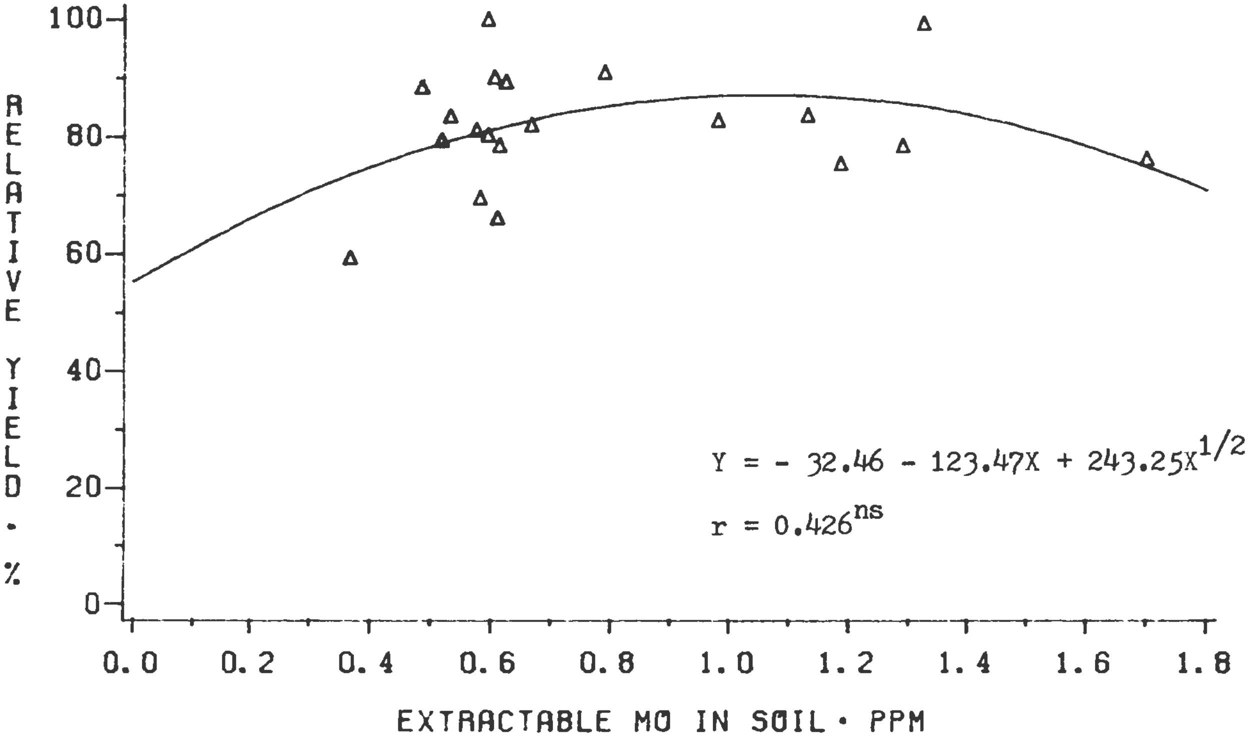


Fig. 1.41. Relationship between relative yield of Centrosema pubescens and extractable Mo in the Wahiawa soil.

Relationship between N Concentration in Plant Top and Mo Concentrations

Relationship between N Concentration in Plant Top and Mo Concentration in Nodule

There was no significant correlation between N concentration in plant top and Mo concentration in nodule of centrosema grown in Wahiawa soil (Fig. 1.42). Nitrogen concentration in plant top was slightly increased as Mo concentration increased . However, N concentration in plant was quite low due to low nitrogenase activity. There was also no significant difference between N concentration in plant receiving different rates of Mo (Table 1.47). These results likely may have been the reason for no significant correlation between Mo concentration in nodule and N concentration in plant top. Some factors other than Mo may have limited nitrogenase activity, such that N concentration was low and did not correlate well with Mo concentration in nodule.

Relationship between N and Mo Concentrations in Plant Top

There was also no significant correlation between N and Mo concentrations in the top of centrosema. Nitrogen concentration in the top did not change as Mo concentration increased (Fig. 1.43). Although Mo concentration in the top was less concerned with nitrogen fixation than Mo concentration in nodule, it was expected to correlate with N concentration in the top because it correlated with Mo concentration in nodule. Lack of significant correlation between N and Mo concentration in tops was, partly, due to the lack of significant correlation between N

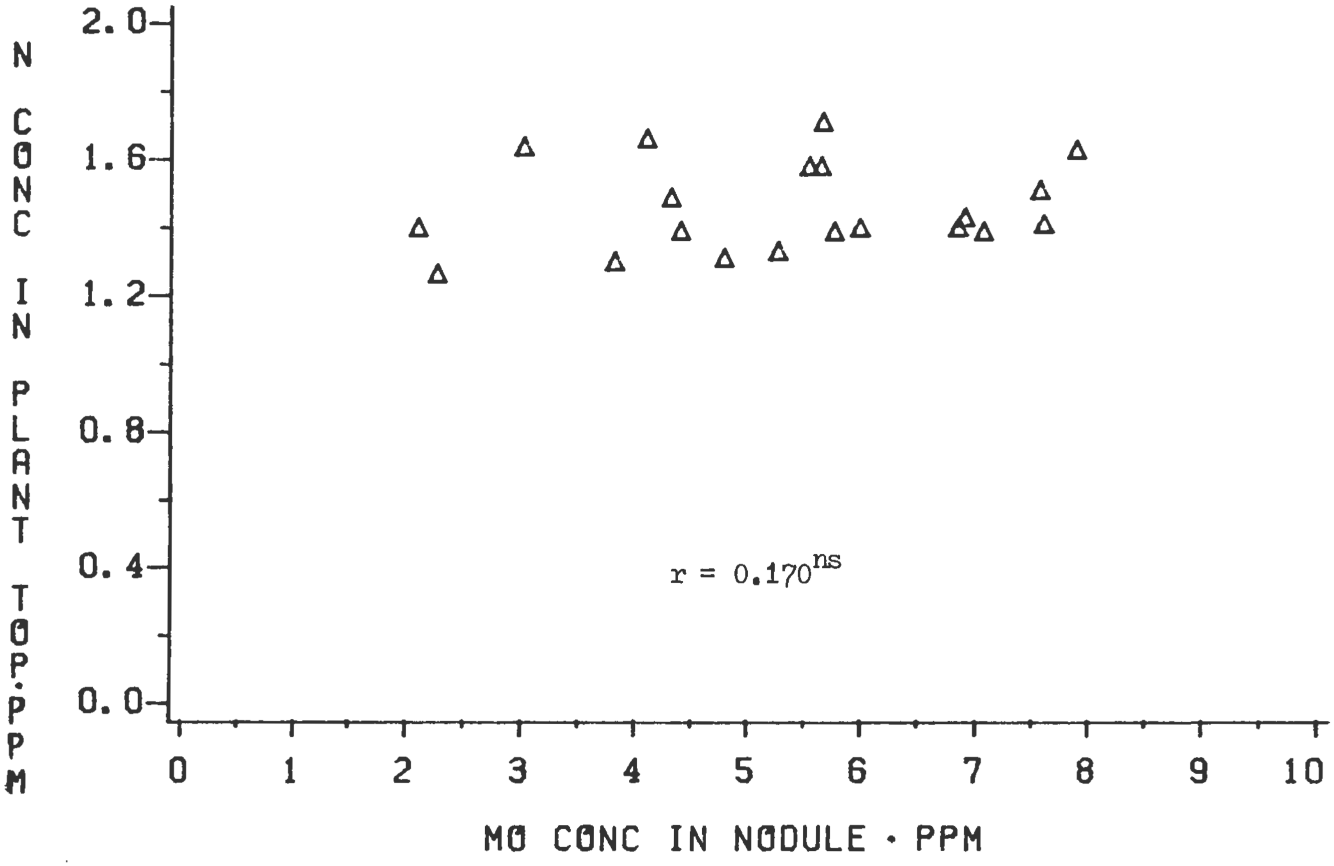


Fig. 1.42. Relationship between N concentration in plant top and Mo concentration in nodule of Centrosema pubescens grown in the Wahiawa soil.

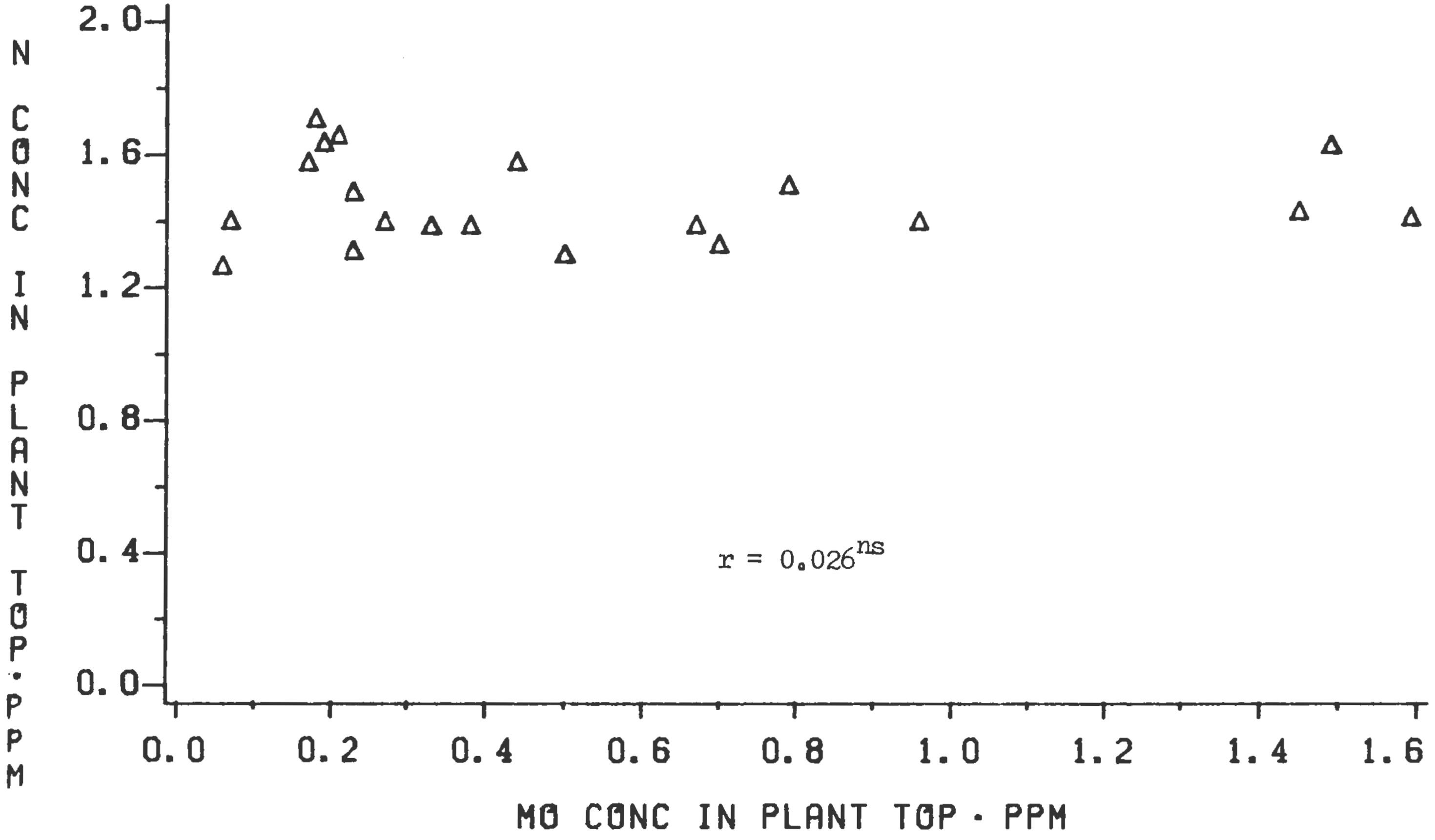


Fig. 1.43. Relationship between N and Mo concentration in the top of <u>Centrosema pubescens</u> grown in the Wahiawa soil.

concentration in plant top and Mo concentration in nodule.

Relationship between N Concentration in Plant top and Extractable Mo in soil

There was no significant correlation between N concentration in plant top and extractable Mo in soil after harvesting centrosema (Fig. 1.44). Nitrogen concentration in plant top did not significantly increase as extractable Mo increased. However, it may be approximate from this scatter diagram that at least 0.4 ppm extractable Mo is requires to increase N concentration in plant top.

Relationship between Nitrogenase Activity and Mo Concentration in Nodule

There was a significant correlation between nitrogenase activity and Mo concentration in nodules with a correlation coefficient of 0.546. Nitrogenase activity increased as Mo concentration in nodules increased (Fig. 1.45). Molybdenum concentration in nodule, therefore, can be used as one of the criteria to assess nitrogenase activity of legumes.

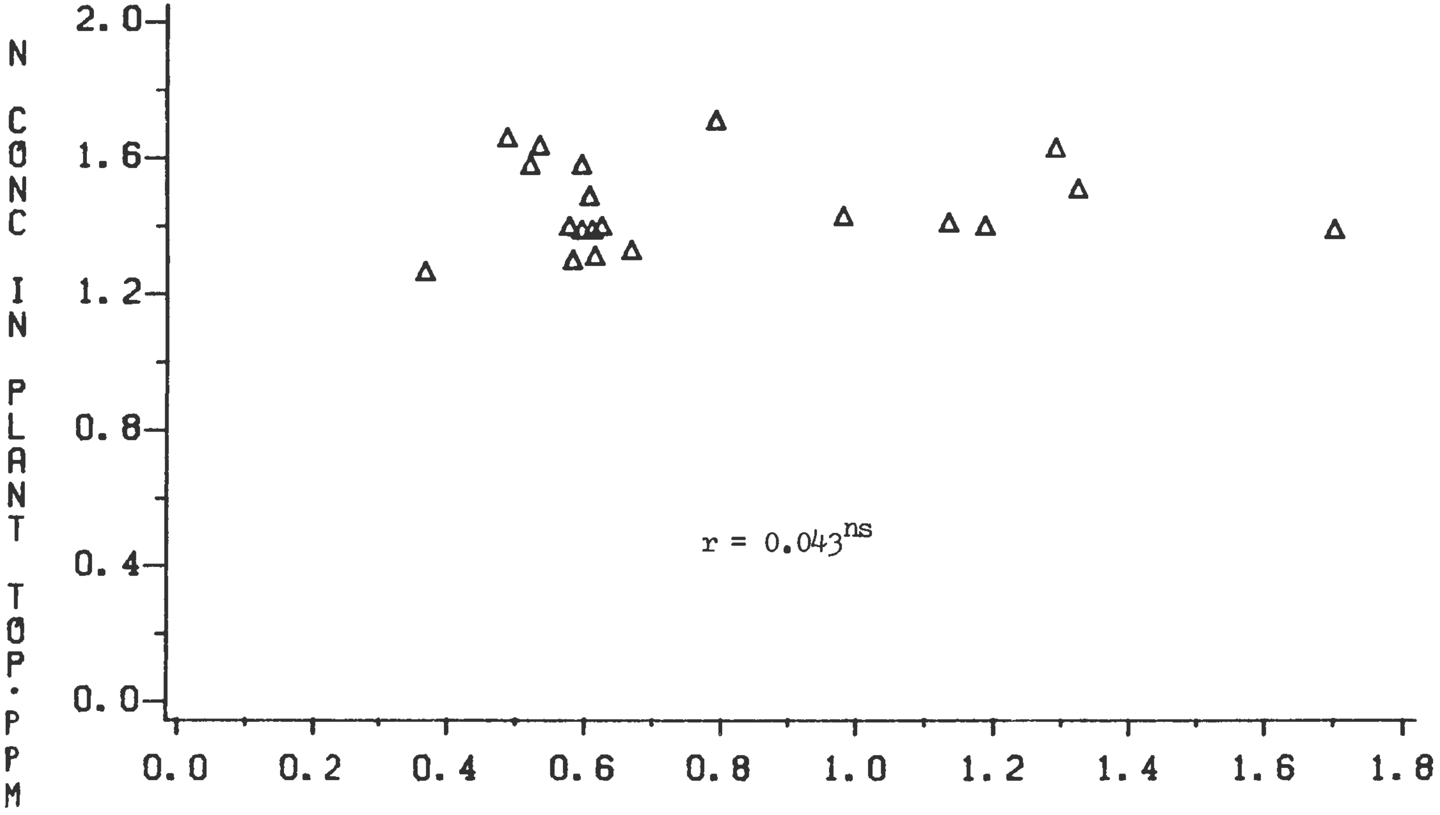


Fig. 1.44. Relationship between N concentration in the top of Centrosema pubescens and extractable Mo in the Wahiawa soil.

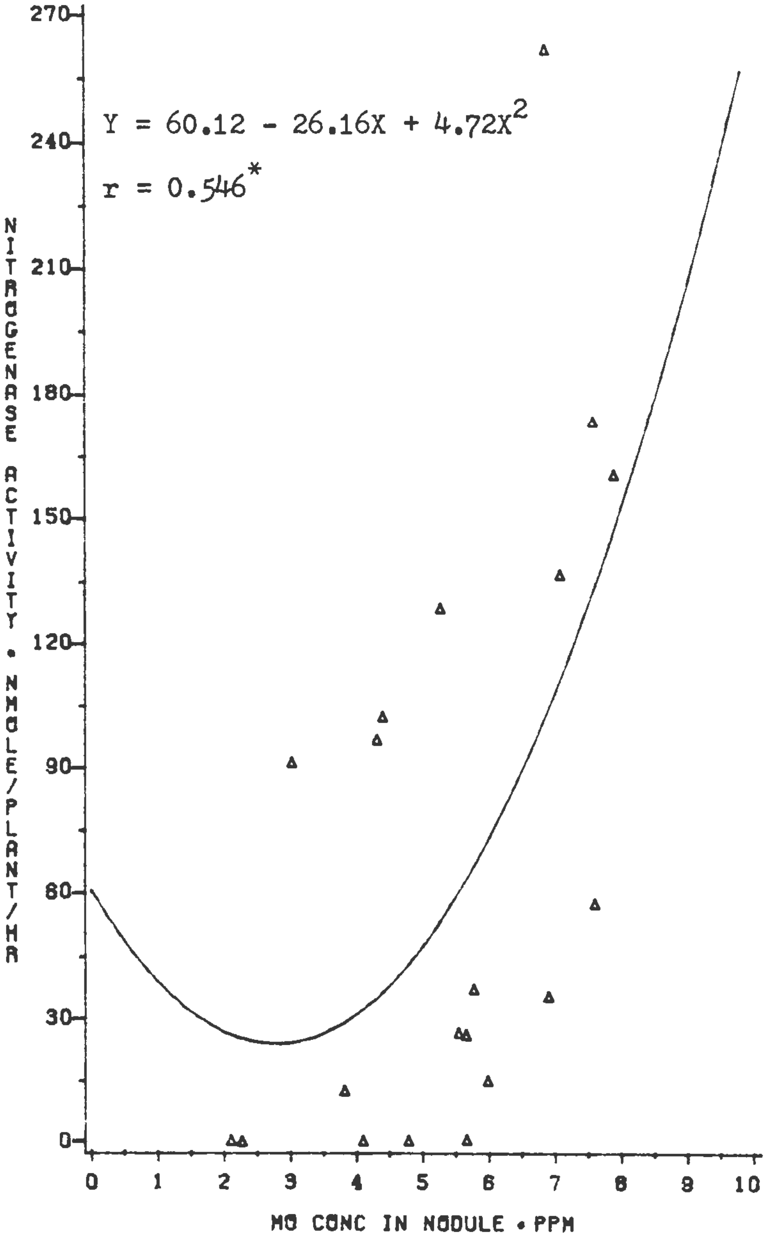


Fig. 1.45. Relationship between nitrogenase activity and Mo concentration in nodule of Centrosema pubescens grown in the Wahiawa soil.

Relationship between S Concentration in Plant Top and Mo Concentration

Relationship between S Concentration in Plant Top and Mo Concentration in Nodule

There was no significant correlation between S concentration in plant top and Mo concentration in nodule of centrosema. The correlation coefficient of the relationship was 0.072 ($p = 0.58$). This result indicated that there was no relationship between S concentration in plant top and Mo concentration in nodule of centrosema.

Relationship between S and Mo Concentrations in Plant Top

There was no significant correlation between S and Mo concentrations in the top of centrosema grown in Wahiawa soil. The correlation of the relationship was 0.031 ($p = 0.81$). This result indicated that there was no relationship between Mo and S concentrations in the top of centrosema.

Relationship between S concentration in Plant Top and Extractable Mo in Soil

There was no significant correlation between S concentration in plant top and extractable Mo in soil after harvesting centrosema. The correlation coefficient of the relationship was 0.037 ($p = 0.78$). the result indicated that there was no relationship between S concentration in the top of centrosema and extractable Mo in the Wahiawa soil.

Relationship between Cu Concentration in Plant Top and Mo Concentrations

Relationship between Cu Concentration in Plant Top and Mo Concentration in Nodule

There was no significant correlation between Cu concentration in plant top and Mo concentration in nodule of centrosema. The correlation coefficient of the relationship was 0.034 ($p = 0.82$). The result indicated that there was no relationship between Cu concentration in plant top and Mo concentration in nodule of centrosema.

Relationship between Cu and Mo Concentrations in Plant Top

There was no significant correlation between Cu and Mo concentrations in the top of centrosema. The correlation coefficient of the relationship was 0.140 ($p = 0.29$). The result indicated that there was no relationship between Cu and Mo concentrations in plant top.

Relationship between Cu Concentration in Plant Top and Extractable Mo in Soil

There was also no significant correlation between Cu concentration in plant top and extractable Mo in soil after harvesting centrosema. The correlation coefficient of the relationship was 0.082 ($p = 0.53$). The result indicated that there was no relationship between Cu concentration in plant top and extractable Mo in soil.

b. Field Experiment

Effects of Mo and Lime on Growth, Yield and Nutrient Concentration of *Desmodium intortum* and *Centrosema pubescens* Grown in the Wahiawa Soil

Yield Response

First Cutting:

Molybdenum application by seed coating and soil treatment significantly increased desmodium and centrosema dry matter yield (Table 1.62). When averaged over the two legume species, yield of legumes receiving soil-applied Mo at 4.0 kg Mo/ha plus lime was the highest. Yield obtained from this treatment was 144.0% above that obtained from the no-Mo treatment. Yield of legumes receiving soil-applied Mo at 4.0 kg Mo/ha plus lime was significantly higher than those of legumes receiving seed-applied Mo at 0.1 kg Mo/ha and soil-applied Mo at 2.0 kg Mo/ha but was not significantly higher than those of legumes receiving lime, seed-applied Mo at 0.3 and 0.5 kg Mo/ha, and soil-applied Mo at 1.0 and 4.0 kg Mo/ha. Legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha significantly produced higher yield than those receiving no Mo. Plants which received no Mo were light green, slightly stunted, and appeared N deficient in comparison to treated plants. Legumes which received seed-applied Mo at 0.1 kg Mo/ha did not significantly produce higher yield than those which received no Mo, suggesting that seed-applied Mo at this rate was not enough for optimum yield of pasture legumes grown in Wahiawa soil. This result was similar to that obtained from the greenhouse experiments previously conducted with desmodium and centrosema grown in the same soil.

Table 1.62.

Effects of Mo and lime on dry matter yields of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (First Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	ton/ha		
No Mo added	1.64	2.12	1.88 d*
Lime	4.54	3.95	4.24 ab
0.1 kg Mo/ha (seed applied)	2.98	2.86	2.92 cd
0.3 kg Mo/ha (seed applied)	5.55	3.09	4.32 ab
0.5 kg Mo/ha (seed applied)	5.33	2.39	3.86 abc
0.5 kg Mo/ha (seed applied) plus lime	4.11	2.88	3.49 abc
1.0 kg Mo/ha (soil applied)	3.43	2.74	3.09 bcd
2.0 kg Mo/ha (soil applied)	3.61	2.19	2.90 cd
4.0 kg Mo/ha (soil applied)	4.82	3.25	4.04 abc
4.0 kg Mo/ha (soil applied) plus lime	5.94	3.24	4.59 a
Mean	4.20 a	2.87 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Legumes receiving soil-applied Mo at 4.0 kg Mo/ha significantly produced higher yield than those receiving no Mo. Yield obtained from this treatment was 114.9% above that obtained from the no-Mo treatment. Although yields obtained from the 1.0 and 2.0 kg Mo/ha soil-applied treatments were also higher than those obtained from the no-Mo treatment, these differences were not significant. Yields of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha were not significantly different from those of legumes receiving soil-applied Mo at 4.0 kg Mo/ha, suggesting that seed-applied Mo at those rates were as effective as the high rate of soil-applied Mo.

Lime applied alone significantly increased yield (Table 1.62). Yield obtained from the lime treatment was 125.5% above that obtained from the no-Mo treatment without lime. Yield obtained from the lime treatment was not significantly different from that obtained from the 4.0 kg Mo/ha soil-applied treatment suggesting that lime also plays an important role in pasture production. When lime was applied along with the high rate of Mo (e.g. 0.5 kg Mo/ha seed-applied and 4.0 kg Mo/ha soil-applied), yield response due to lime was not evident. Yields obtained from the 0.5 kg Mo/ha seed-applied treatment, with or without lime, were not significantly different from each other and so were those obtained from the 4.0 kg Mo/ha soil-applied treatment, with or without lime. These results suggested that a response to lime by pasture legumes was due to an influence of lime in increasing Mo availability in soil.

When averaged over all Mo and lime rates, yield of desmodium was significantly higher than that of centrosema. It should be noted that desmodium grew faster than centrosema. Desmodium was more responsive to Mo than centrosema since yield increase by the same rate of Mo application was higher for desmodium than for centrosema. However, the trend of response of both species was similar, i.e. yield increased as Mo rate increased.

Second Cutting:

Legumes were harvested at 60 days after the first cutting. There were no significant differences among dry matter yields obtained from treatments receiving various rates of Mo when averaged over the two legume species (Table 1.63). However, the trend of yield increase was similar to that of the first cutting (Table 1.62) which showed that yield increased as Mo rate increased. It should be noted that while yield obtained from the no-Mo treatment at the second cutting did not decrease from that obtained from the same treatment at the first cutting, yield obtained from the Mo treatment decreased markedly, especially for desmodium. This result was due to the effect of cutting on a regrowth of plant. Before the first cutting pasture legumes were allowed to grow 112 days for establishment. At harvest time of first cutting, desmodium stands from the Mo treatment were quite large and stems were widespread. Such great growth may have drained out the reserved carbohydrate from the stem close to the soil so that the regrowth of plant after harvesting was affected. However, desmodium receiving various rate of Mo grew vigorously after one month from the first cutting.

Third Cutting:

Legumes were harvested at 60 days from the second cutting. Dry matter yields of desmodium and centrosema were increased when Mo was

Table 1.63.

Effects of Mo and lime on dry matter yields of <u>Desmodium intortum</u> and <u>Centrosema pubescens</u> grown in the Wahiawa soil (Second Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	ton/ha		
No Mo added	1.64	1.87	1.76 a*
Lime	2.40	2.02	2.21 a
0.1 kg Mo/ha (seed applied)	2.19	1.69	1.94 a
0.3 kg Mo/ha (seed applied)	2.42	1.93	2.17 a
0.5 kg Mo/ha (seed applied)	2.79	1.65	2.22 a
0.5 kg Mo/ha (seed applied) plus lime	2.37	1.61	1.99 a
1.0 kg Mo/ha (soil applied)	2.39	1.64	2.01 a
2.0 kg Mo/ha (soil applied)	2.28	1.99	2.13 a
4.0 kg Mo/ha (soil applied)	2.52	1.66	2.10 a
4.0 kg Mo/ha (soil applied) plus lime	2.46	1.63	2.05 a
Mean	2.35 a	1.77 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

applied (Table 1.64). When averaged over the two legume species, yield of legumes receiving no Mo was significantly lower than those of legumes receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 2.0 kg Mo/ha. Yield of legumes receiving no Mo, however, was not significantly lower than those of legumes receiving other rates of Mo. Yields obtained from legumes receiving various rates of seed-applied or soil-applied Mo were not significantly different from each other. Yield of legumes receiving seed-applied Mo at 0.5 kg Mo/ha with lime or soil-applied Mo at 4.0 kg Mo/ha with lime was not significantly different from that of legumes receiving the same rate of Mo without lime. Although yield of legumes grown in limed soil without Mo was 23% higher than that of legume grown in unlimed soil without Mo, it was not statistically significant.

Yield of desmodium was significantly higher than that of centrosema indicating that desmodium regrowth was more vigorous and faster than centrosema. There was no interaction between Mo treatments and legume species.

Fourth Cutting:

Legumes were harvested at 60 days after the third cutting. At this cutting the differences among dry matter yields obtained from Mo treatments became evident (Table 1.65). When averaged over the two legume species, yield of legumes receiving soil-applied Mo at 4.0 kg Mo/ha with lime was significantly higher than those of legumes receiving other rates of soil-applied Mo. Yield obtained from this soil-applied Mo treatment was 109% above that obtained from the no-Mo treatment. Yield of legume receiving soil-applied Mo at 4.0 kg Mo/ha with lime was

Table 1.64

Effects of Mo and lime on dry matter yields of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Third Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	----------	ton/ha ----------	----------
No Mo added	1.52	1.18	1.35 b*
Lime	2.00	1.33	1.67 ab
0.1 kg Mo/ha (seed applied)	1.79	1.20	1.50 ab
0.3 kg Mo/ha (seed applied)	2.00	1.13	1.57 ab
0.5 kg Mo/ha (seed applied)	2.35	1.38	1.87 a
0.5 kg Mo/ha (seed applied) plus lime	2.07	1.00	1.53 ab
1.0 kg Mo/ha (soil applied)	2.06	1.08	1.57 ab
2.0 kg Mo/ha (soil applied)	2.63	1.13	1.88 a
4.0 kg Mo/ha (soil applied)	2.10	1.21	1.66 ab
4.0 kg Mo/ha (soil applied) plus lime	2.30	1.13	1.72 ab
Mean	2.08 a	1.18 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.65

Effects of Mo and lime on dry matter yields of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fourth Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	----------	ton/ha	----------
No Mo added	1.31	0.70	1.00 d*
Lime	2.37	1.15	1.76 b
0.1 kg Mo/ha (seed applied)	1.83	1.04	1.44 c
0.3 kg Mo/ha (seed applied)	2.35	1.06	1.70 bc
0.5 kg Mo/ha (seed applied)	2.71	1.15	1.93 ab
0.5 kg Mo/ha (seed applied) plus lime	2.49	1.11	1.80 b
1.0 kg Mo/ha (soil applied)	2.39	0.97	1.68 bc
2.0 kg Mo/ha (soil applied)	2.26	1.04	1.65 bc
4.0 kg Mo/ha (soil applied)	2.53	0.97	1.75 b
4.0 kg Mo/ha (soil applied) plus lime	3.01	1.18	2.10 a
Mean	2.33 a	1.04 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

significantly higher than that of legumes receiving the same rate of Mo without lime. Yields of legumes receiving soil-applied Mo at 1.0, 2.0 or 4.0 kg Mo/ha were not significantly different from each other. Although yield of legumes receiving soil-applied Mo at 4.0 kg Mo/ha plus lime was significantly higher than those of legumes receiving other rates of Mo, it was not significantly higher than that of legumes receiving seed-applied Mo at 0.5 kg Mo/ha. These results suggested that seed-applied Mo at this rate also has a residual effect on growth of pasture legumes that are harvested many times a year. Yield of legumes receiving seed-applied Mo at 0.3, 0.5 and 0.5 kg Mo/ha plus lime were not significantly different from each other. Yield of legumes receiving seed-applied Mo at 0.5 kg Mo/ha was significantly higher than those of legumes receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo. Yield of legumes receiving only lime was significantly higher than those of legumes receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo but was not significantly different from those of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha or soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha.

It should be mentioned that at this fourth cutting which performed on January 3, 1984 desmodium and centrosema had flowers and pods through the experimental plots.

Fifth Cutting:

When averaged over all legume species, dry matter yield of legumes receiving soil-applied Mo at 4.0 kg Mo/ha plus lime was significantly higher than those of legumes receiving soil-applied Mo at 1.0 kg Mo/ha, seed-applied Mo at 0.1 and 0.3 kg Mo/ha, and no Mo (Table 1.66).

Table 1.66

Effects of Mo and lime on dry matter yields of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fifth Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	ton/ha		
No Mo added	1.63	1.15	1.39 e*
Lime	3.53	1.49	2.51 abc
0.1 kg Mo/ha (seed applied)	2.19	1.47	1.83 d
0.3 kg Mo/ha (seed applied)	3.40	1.40	2.40 bc
0.5 kg Mo/ha (seed applied)	4.09	1.30	2.70 abc
0.5 kg Mo/ha (seed applied) plus lime	4.38	1.30	2.84 ab
1.0 kg Mo/ha (soil applied)	3.40	1.20	2.30 c
2.0 kg Mo/ha (soil applied)	3.51	1.51	2.51 abc
4.0 kg Mo/ha (soil applied)	3.58	1.38	2.48 abc
4.0 kg Mo/ha (soil applied) plus lime	4.34	1.38	2.86 a
Mean	3.41 a	1.36 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Yields of legumes receiving soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha were not significantly different from each other. Yields of legumes receiving those soil-applied Mo rates were also not significantly different from those of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha but were significantly higher than those of legumes receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo. Yield of legumes receiving seed-applied Mo at 0.5 kg Mo/ha plus lime was also significantly higher than those of legumes receiving soil-applied Mo at 1.0 kg Mo/ha, seed-applied Mo at 0.1 kg Mo/ha and no Mo. Yield of legumes receiving only lime was not significantly different from those of legumes receiving high rates of Mo but significantly higher than those receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo. It should be noted that when averaged over all Mo and lime rates, yield of desmodium was significantly higher than that of centrosema.

Consideration of the differences in dry matter yields of desmodium and centrosema receiving various rates of Mo and/or lime from cuttings 1 to 5 indicated that seed-applied Mo at 0.3 and 0.5 kg Mo/ha were as effective as soil- applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha in supplying Mo to desmodium and centrosema. Seed-applied Mo at 0.1 kg Mo/ha, although increasing yield above the no-Mo treatment, was not considered adequate for optimum yields of the two legume species. Lime also increased pasture legume yield at the same magnitude as Mo. Seed-applied Mo at 0.3 and 0.5 kg Mo/ha also had residual effect similar to that of soil- applied Mo.

Influences of Mo and Lime on Some Chemical Compositions in Plant

Nitrogen

First Cutting:

When average over all legume species, N concentration in the top of legumes receiving no Mo was significantly lower than those of legumes receiving various rates of seed-applied and soil-applied Mo (Table 1.67). However, N concentration in the top of legumes receiving no Mo was not significantly lower than that of legumes receiving seed-applied Mo at 0.1 kg Mo/ha, suggesting that seed-applied Mo at this rate was not enough for plant need to produce optimum N concentration which contributed to high yield. Nitrogen concentrations of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha, and soil-applied Mo at various rates were not significantly different from each other. Nitrogen concentration of legumes receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than that of legumes receiving seed-applied Mo at 0.1 kg Mo/ha. Nitrogen concentration of legumes receiving only lime was significantly higher than that of legumes receiving no Mo but was not significantly different from those of legumes receiving various rates of seed- or soil-applied Mo. Nitrogen concentration of legumes receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 4.0 kg Mo/ha with lime were not significantly different from those of legumes receiving the same rates of Mo without lime.

When average over all Mo and lime rates, N concentration of desmodium was significantly higher than that of centrosema, suggesting that nitrogen fixation as affected by Mo or lime of desmodium was higher

Table 1.67.

Effects of Mo and lime on N concentration in the top of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (First Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	----------------	% ----------------	
No Mo added	1.85	1.92	1.88 c*
Lime	2.74	2.58	2.66 ab
0.1 kg Mo/ha (seed applied)	2.28	2.27	2.27 bc
0.3 kg Mo/ha (seed applied)	2.82	2.66	2.74 ab
0.5 kg Mo/ha (seed applied)	3.15	2.37	2.76 ab
0.5 kg Mo/ha (seed applied) plus lime	3.05	2.63	2.84 ab
1.0 kg Mo/ha (soil applied)	2.65	2.40	2.53 ab
2.0 kg Mo/ha (soil applied)	2.91	2.41	2.66 ab
4.0 kg Mo/ha (soil applied)	3.13	2.67	2.90 a
4.0 kg Mo/ha (soil applied) plus lime	3.11	2.66	2.89 a
Mean	2.77 a	2.46 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

than that of centrosema. There was no interaction between Mo rates and legume species on N concentration in plant top. It should be noted that N deficiency symptoms occurred with desmodium receiving no Mo. Nitrogen concentration in the top of desmodium receiving no Mo (1.85%) was lower than the critical N concentration of 2.7% to 3.0% suggested by Johansen and Kerridge (1981).

Second Cutting:

When averaged over the two legume species, N concentrations of legumes receiving various rates of soil-applied Mo were not significantly different from each other (Table 1.68). Nitrogen concentrations of legumes receiving soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha were also not significantly different from those of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha but were significantly higher than those of legumes receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo. Nitrogen concentrations of legumes receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha were not significantly different from that of legumes receiving seed-applied Mo at 0.1 kg Mo/ha but were significantly different from that of legumes receiving no Mo. Nitrogen concentration of legumes receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly different from that of legumes receiving no Mo. Nitrogen concentrations of legumes receiving lime alone and lime along with seed-applied Mo at 0.5 kg Mo/ha or soil-applied Mo at 4.0 kg Mo/ha were not significantly different from those of legumes receiving other rates of seed-applied and soil-applied Mo but were significantly higher than that of legumes receiving no Mo. When averaged over all Mo and lime rates, N concentration in the top of desmodium was significantly lower than that of centrosema. At this cutting

Table 1.68.

Effects of Mo and lime on N concentration in the top of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Second Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	------------------	% ------------------	
No Mo added	1.96	2.57	2.26 c*
Lime	2.77	3.50	3.14 ab
0.1 kg Mo/ha (seed applied)	2.45	2.99	2.72 bc
0.3 kg Mo/ha (seed applied)	2.96	3.10	3.03 ab
0.5 kg Mo/ha (seed applied)	3.27	3.13	3.20 ab
0.5 kg Mo/ha (seed applied) plus lime	3.46	3.46	3.46 a
1.0 kg Mo/ha (soil applied)	3.11	3.51	3.31 a
2.0 kg Mo/ha (soil applied)	3.43	3.26	3.35 a
4.0 kg Mo/ha (soil applied)	3.43	3.58	3.50 a
4.0 kg Mo/ha (soil applied) plus lime	3.50	3.53	3.52 a
Mean	3.03 b	3.26 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

nitrogen fixation of centrosema was likely to be more effective than that of centrosema in the first cutting. Moreover, N concentration in the top of centrosema receiving no Mo was higher than that of desmodium receiving the same Mo treatment.

Third Cutting:

Molybdenum application significantly increased N concentration in the top of desmodium and tended to increase N concentration in the top of centrosema (Table 1.69). Nitrogen concentrations of desmodium receiving various rates of seed-applied Mo and soil-applied Mo, except seed-applied Mo at 0.1 kg Mo/ha, were significantly higher than that of legume receiving no Mo. Nitrogen concentration of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly higher than that of legume receiving no Mo. Nitrogen concentrations of desmodium receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha were not significantly different from each other. However, N concentrations of desmodium receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha were significantly higher than that of legume receiving seed-applied Mo at 0.1 kg Mo/ha. Nitrogen concentrations of desmodium receiving various rates of soil-applied Mo were not significantly different from each other. Nitrogen concentrations of desmodium receiving soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha were also not significantly different from those of legume receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha. Nitrogen concentrations of desmodium receiving lime alone, lime along with seed-applied Mo at 0.5 kg Mo/ha, and lime along with soil-applied Mo at 4.0 kg Mo/ha were also significantly higher than those of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha

Table 1.69

Effects of Mo and lime on N concentration in the top of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Third Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	----------	% ----------	----------
No Mo added	1.88 e*	3.01 abcd	2.45
Lime	2.59 d	3.43 a	3.01
0.1 kg Mo/ha (seed applied)	2.16 e	3.16 ab	2.66
0.3 kg Mo/ha (seed applied)	2.68 cd	3.30 a	2.99
0.5 kg Mo/ha (seed applied)	2.83 bcd	3.10 abc	2.96
0.5 kg Mo/ha (seed applied) plus lime	2.83 bcd	3.16 ab	3.00
1.0 kg Mo/ha (soil applied)	2.79 bcd	3.33 a	3.06
2.0 kg Mo/ha (soil applied)	2.67 cd	3.00 abcd	2.83
4.0 kg Mo/ha (soil applied)	2.86 bcd	3.42 a	3.14
4.0 kg Mo/ha (soil applied) plus lime	2.66 d	3.39 a	3.02
Mean	2.59 a	3.23 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

and no Mo.

Nitrogen concentration in the tops of centrosema receiving various rates of seed-applied and soil-applied Mo and/or lime were not significantly different from each other. However, Mo application tended to increase N concentration of this legume. When averaged over all Mo and lime rates, N concentration in the top of desmodium was significantly lower than that of centrosema.

Fourth Cutting:

Nitrogen concentrations in the top of desmodium receiving various rates of soil-applied Mo were not significantly different from those of desmodium receiving seed-applied Mo at 0.3 and 0.5 kg Mo/ha but were significantly higher than those of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo (Table 1.70). Nitrogen concentration of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly higher than that of no Mo. It should be mentioned that at this cutting desmodium receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo showed N deficiency symptoms. Nitrogen concentrations of desmodium receiving lime alone or lime with high rates of seed-applied and soil-applied Mo were not significantly different from those of desmodium receiving various rates of soil-applied Mo and seed-applied Mo at 0.3 and 0.5 kg Mo/ha but were also significantly higher than those of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo.

There were no significant differences among N concentrations of centrosema receiving various rates of seed-applied and soil-applied Mo and/or lime. Nitrogen concentration of centrosema receiving

Table 1. 70.

Effects of Mo and lime on N concentration in the top of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fourth Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	----------	% ----------	----------
No Mo added	2.16 c*	3.72 a	2.94
Lime	2.70 b	3.84 a	3.27
0.1 kg Mo/ha (seed applied)	2.32 c	3.73 a	3.03
0.3 kg Mo/ha (seed applied)	2.78 b	3.81 a	3.30
0.5 kg Mo/ha (seed applied)	2.84 b	3.63 a	3.24
0.5 kg Mo/ha (seed applied) plus lime	2.83 b	3.88 a	3.35
1.0 kg Mo/ha (soil applied)	2.84 b	3.72 a	3.29
2.0 kg Mo/ha (soil applied)	2.96 b	3.94 a	3.45
4.0 kg Mo/ha (soil applied)	3.04 b	3.70 a	3.37
4.0 kg Mo/ha (soil applied) plus lime	2.90 b	3.87 a	3.39
Mean	2.74 b	3.78 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

no Mo was not significantly lower than in other Mo treatments. When averaged over all Mo and lime rates, N concentration in the top of desmodium was significantly lower than that of centrosema.

Fifth Cutting:

Nitrogen concentration in the top of desmodium receiving no Mo was not significantly lower than in that of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha but was significantly lower than those of desmodium receiving other rates of seed-applied and soil-applied Mo (Table 1.71). Nitrogen concentration of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha was also significantly lower than those of desmodium receiving other rates of Mo. Nitrogen concentration of desmodium receiving seed-applied Mo at 0.3 kg Mo/ha was significantly lower than those of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at the various rates, suggesting that seed-applied Mo at 0.3 kg Mo/ha likely may not adequately supply Mo to plant need after 4 to 5 cuttings. Nitrogen concentration of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from those of desmodium receiving various rates of soil-applied Mo, indicating that seed-applied Mo at this rate was still as effective as soil-applied Mo for desmodium which had been cut five times. Nitrogen concentrations of desmodium receiving lime alone or lime with high rates of Mo were not significantly different from those of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at rates of 1.0 and 2.0 kg Mo/ha but were significantly lower than that of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha.

Nitrogen concentration in the top of centrosema receiving

Table 1.71.

Effects of Mo and lime on N concentration in the top of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fifth Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	------------------	% ------------------	------------------
No Mo added	1.45 h	3.06 bcd	2.25
Lime	2.60 ef	3.36 ab	2.98
0.1 kg Mo/ha (seed applied)	1.69 h	3.30 ab	2.49
0.3 kg Mo/ha (seed applied)	2.23 g	3.33 ab	2.78
0.5 kg Mo/ha (seed applied)	2.73 def	3.13 abc	2.93
0.5 kg Mo/ha (seed applied) plus lime	2.51 efg	3.29 ab	2.90
1.0 kg Mo/ha (soil applied)	2.37 ef	3.46 a	2.92
2.0 kg Mo/ha (soil applied)	2.72 def	3.29 ab	3.01
4.0 kg Mo/ha (soil applied)	3.07 abcd	3.20 ab	3.14
4.0 kg Mo/ha (soil applied) plus lime	2.80 cde	3.25 ab	3.03
Mean	2.42 b	3.27 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

no Mo was significantly lower than that of centrosema receiving soil-applied Mo at 1.0 kg Mo/ha but was not significantly lower than those of centrosema receiving other rates of Mo and/or lime (Table 1.71). Nitrogen concentrations of centrosema receiving various rates of seed-applied and soil-applied Mo were not significantly different from each other. When averaged over all Mo and lime rates, N concentration in the top of desmodium was significantly lower than that of centrosema. Nitrogen concentration in the top of desmodium receiving no Mo was significantly lower than that in the top of centrosema, indicating that the supply of Mo was not enough for desmodium whereas that of Mo was enough for centrosema. These results suggested that desmodium is more responsive to Mo than centrosema.

Molybdenum

First Cutting:

Molybdenum application significantly increased Mo concentrations in the tops of desmodium and centrosema (Table 1.72). When averaged over the two legume species, Mo concentration of legumes receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than those of legumes receiving other rates of Mo. Molybdenum concentration of legumes receiving soil-applied Mo at 2.0 kg Mo/ha was also significantly higher than those of legumes receiving soil-applied Mo at 1.0 kg Mo/ha and seed-applied Mo at various rates. Molybdenum concentrations of legumes receiving various rates of seed-applied Mo were not significantly different from each other. Molybdenum concentrations of legumes receiving lime alone

Table 1.72.

Effects of Mo and lime on Mo concentration in the tops of <u>Desmodium intortum</u> and <u>Centrosema pubescens</u> grown in the Wahiawa soil (First Cutting)

Mo treatment	<u>Desmodium intortum</u>	<u>Centrosema pubescens</u>	Mean
	---------- ppm	----------	----------
No Mo added	0.11	0.11	0.11 d*
Lime	0.19	0.08	0.14 d
0.1 kg Mo/ha (seed applied)	0.14	0.10	0.12 d
0.3 kg Mo/ha (seed applied)	0.16	0.12	0.14 d
0.5 kg Mo/ha (seed applied)	0.16	0.13	0.15 d
0.5 kg Mo/ha (seed applied) plus lime	0.16	0.13	0.15 d
1.0 kg Mo/ha (soil applied)	0.17	0.15	0.16 d
2.0 kg Mo/ha (soil applied)	0.35	0.27	0.31 c
4.0 kg Mo/ha (soil applied)	0.52	0.41	0.46 b
4.0 kg Mo/ha (soil applied) plus lime	0.51	0.68	0.60 a
Mean	0.25 a	0.22 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

or lime with seed-applied Mo at 0.5 kg Mo/ha were not significantly different from those of legume receiving no Mo, soil-applied Mo at 1.0 kg Mo/ha, and seed-applied Mo at 0.1, 0.3 and 0.5 kg Mo/ha. However, Mo concentration of legumes receiving soil-applied Mo at 4.0 kg Mo/ha with lime was significantly higher than those of legumes receiving other rates of Mo. When averaged over all Mo and lime rates, Mo concentration in the tops of desmodium and centrosema were not significantly different from each other.

Second Cutting:

When averaged over all legume species, Mo concentration in the tops of legumes receiving soil-applied Mo at 4.0 kg Mo/ha was significantly higher than those of legumes receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha, seed-applied Mo at various rates and no Mo (Table 1.73). Although Mo concentration of legumes receiving soil-applied Mo at 2.0 kg Mo/ha was not significantly higher than those of legume receiving soil-applied Mo at 1.0 kg Mo/ha and seed-applied Mo at different rates, it was significantly higher than that of legumes receiving no Mo. Molybdenum concentrations of legumes receiving lime alone or lime with seed-applied Mo at 0.5 kg Mo/ha was not significantly different from those of legumes receiving no Mo and seed-applied Mo at other rates. However, Mo concentration of legumes receiving soil applied Mo at 4.0 kg Mo/ha with lime was significantly higher than those of legumes receiving other rates of Mo. When averaged over all Mo and lime rates, Mo concentration in the top of desmodium was not significantly different from that in the top of centrosema. There was no interaction between legume species and Mo or lime treatments.

Table 1.73

Effects of Mo and lime on Mo concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Second Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	---------------	ppm ---------------	
No Mo added	0.08	0.08	0.08 d*
Lime	0.13	0.12	0.12 cd
0.1 kg Mo/ha (seed applied)	0.10	0.13	0.12 cd
0.3 kg Mo/ha (seed applied)	0.14	0.09	0.11 cd
0.5 kg Mo/ha (seed applied)	0.20	0.11	0.16 cd
0.5 kg Mo/ha (seed applied) plus lime	0.18	0.13	0.16 cd
1.0 kg Mo/ha (soil applied)	0.20	0.14	0.17 cd
2.0 kg Mo/ha (soil applied)	0.28	0.24	0.27 c
4.0 kg Mo/ha (soil applied)	0.51	0.35	0.43 b
4.0 kg Mo/ha (soil applied) plus lime	0.85	0.56	0.70 a
Mean	0.27 a	0.20 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Third Cutting:

Molybdenum concentration in the tops of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha with or without lime were significantly higher than those of desmodium receiving other rates of Mo (Table 1.74). Molybdenum concentration in the top of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha with lime was significantly higher than that of desmodium receiving the same rate of Mo without lime, indicating that lime had a significant effect in increasing Mo concentration in plant top at this cutting. Molybdenum concentration of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from that of desmodium receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha but was significantly higher than those of legume receiving seed-applied Mo at 0.1 kg Mo/ha and no Mo.

Molybdenum concentration in the top of centrosema receiving soil-applied Mo at 4.0 kg Mo/ha with or without lime were also significantly higher than those of centrosema receiving other rates of Mo (Table 1.74). Molybdenum concentration in the top of centrosema receiving soil-applied Mo at 4.0 kg Mo/ha with lime was significantly higher than that of centrosema receiving the same rate of Mo without lime. Molybdenum concentration of centrosema receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from those of centrosema receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha. Molybdenum concentration of centrosema receiving no Mo was not significantly different from those of centrosema receiving seed-applied Mo at various rates or lime.

When averaged over all Mo and lime rates, mean Mo concentration in the top of desmodium was not significantly different from

Table 1.74.

Effects of Mo and lime on Mo concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Third Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	------------------	ppm ----------------------------	
No Mo added	0.03 g*	0.05 fg	0.04
Lime	0.06 fg	0.10 efg	0.08
0.1 kg Mo/ha (seed applied)	0.03 g	0.08 fg	0.06
0.3 kg Mo/ha (seed applied)	0.08 fg	0.12 efg	0.10
0.5 kg Mo/ha (seed applied)	0.15 defg	0.13 defg	0.14
0.5 kg Mo/ha (seed applied) plus lime	0.11 fg	0.15 defg	0.13
1.0 kg Mo/ha (soil applied)	0.16 def	0.15 defg	0.16
2.0 kg Mo/ha (soil applied)	0.22 de	0.24 d	0.23
4.0 kg Mo/ha (soil applied)	0.61 b	0.37 c	0.49
4.0 kg Mo/ha (soil applied) plus lime	0.77 a	0.53 b	0.65
Mean	0.22 a	0.19 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

that of centrosema. However, while there there was no significant difference among Mo concentrations of desmodium and centrosema receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha and other rates of seed-applied Mo or no Mo, Mo concentration of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha, with or without lime, were significantly higher than those of centrosema receiving the same soil-applied Mo rates.

Fourth Cutting:

Molybdenum concentrations in the top of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha with or without lime were significantly higher than those of desmodium receiving other rates of Mo (Table 1.75). Molybdenum concentration of desmodium receiving soil-applied Mo at 2.0 kg Mo/ha was also significantly higher than those of desmodium receiving seed-applied Mo at various rates, no Mo, and lime. Molybdenum concentrations of desmodium receiving seed-applied Mo at 0.1, 0.3 and 0.5 kg Mo/ha were not significantly different from each other. Molybdenum concentration of desmodium receiving no Mo was not significantly different from those of desmodium receiving lime, seed-applied Mo at all rates, and soil-applied Mo at 1.0 kg Mo/ha.

Molybdenum concentrations of centrosema receiving soil-applied Mo at 4.0 kg Mo/ha, with or without lime, were significantly higher than those of centrosema receiving other rates of Mo or lime (Table 1.75). Molybdenum concentrations of centrosema receiving seed-applied Mo at 0.1, 0.3 and 0.5 kg Mo/ha were not significantly different from each other. Molybdenum concentrations of centrosema receiving those rates of seed-applied Mo were also not significantly different from those of centrosema receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha. Molybdenum concentrations

Table 1.75

Effects of Mo and lime on Mo concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fourth Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	ppm		
No Mo added	0.05 d*	0.07 d	0.06
Lime	0.07 d	0.11 d	0.09
0.1 kg Mo/ha (seed applied)	0.03 d	0.14 cd	0.09
0.3 kg Mo/ha (seed applied)	0.08 d	0.11 d	0.11
0.5 kg Mo/ha (seed applied)	0.16 d	0.13 cd	0.13
0.5 kg Mo/ha (seed applied) plus lime	0.11 d	0.14 cd	0.13
1.0 kg Mo/ha (soil applied)	0.17 cd	0.14 cd	0.16
2.0 kg Mo/ha (soil applied)	0.29 bc	0.19 cd	0.24
4.0 kg Mo/ha (soil applied)	0.66 a	0.38 b	0.52
4.0 kg Mo/ha (soil applied) plus lime	0.65 a	0.40 b	0.53
Mean	0.22 a	0.18 b	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

of centrosema receiving no Mo was not significantly different from those of centrosema receiving lime, seed-applied Mo at all rates, and soil-applied Mo at 1.0 and 2.0 kg Mo/ha. While there were no significant differences among Mo concentrations of desmodium and centrosema receiving the same rates of seed-applied Mo and soil-applied Mo at 1.0 and 2.0 kg Mo/ha, Mo concentrations of desmodium receiving soil-applied Mo at 4.0 kg Mo/ha, with or without lime, were significantly higher than those of centrosema receiving the same rates of Mo.

When averaged over all Mo and lime rates, mean Mo concentration of desmodium was significantly higher than that of centrosema. This result was different from the result obtained from the previous third cuttings where mean Mo concentrations of desmodium and centrosema were not significantly different from each other. The difference in results was due to the difference in the growth stages of legumes at harvesting. While the legumes were in a vegetative growth stage at the previous three cuttings, they were in a reproductive stage with flowers and pods appearing at the fourth cutting. At this reproductive stage, desmodium and centrosema were quite different from each other. Desmodium bore much more flowers and pods than centrosema. This difference should account for the difference in Mo concentration in plant tops.

Fifth Cutting:

When averaged over all legume species, Mo concentrations in the tops of legumes receiving soil-applied Mo at 4.0 kg Mo/ha, with or without lime, were significantly higher than those of legumes receiving other rates of Mo (Table 1.76). Molybdenum concentrations of legumes receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha were not significantly

Table 1.76

Effects of Mo and lime on Mo concentration in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa Soil (Fifth Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	------------------	ppm ------------------	
No Mo added	0.02	0.04	0.03 b*
Lime	0.04	0.05	0.05 b
0.1 kg Mo/ha (seed applied)	0.03	0.06	0.05 b
0.3 kg Mo/ha (seed applied)	0.04	0.07	0.06 b
0.5 kg Mo/ha (seed applied)	0.09	0.08	0.08 b
0.5 kg Mo/ha (seed applied) plus lime	0.06	0.10	0.08 b
1.0 kg Mo/ha (soil applied)	0.05	0.05	0.05 b
2.0 kg Mo/ha (soil applied)	0.16	0.07	0.12 b
4.0 kg Mo/ha (soil applied)	0.37	0.29	0.33 a
4.0 kg Mo/ha (soil applied) plus lime	0.40	0.22	0.31 a
Mean	0.13 a	0.10 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

different from those of legumes receiving seed-applied Mo, lime, or no Mo. When averaged over all Mo and lime rates, Mo concentrations of desmodium and centrosema were not significantly different from each other. It should be noted that Mo concentrations of plant tops obtained from the different treatments were relatively low as compared to those from the previous cuttings, suggesting that Mo in soil was being depleted.

Sulfur

First Cutting:

Sulfur concentration in the top of desmodium receiving no Mo was significantly higher than those of desmodium receiving other rates of Mo and/or lime (Table 1.77). Sulfur concentrations of desmodium receiving various rates of seed-applied Mo, soil-applied Mo, and/or lime were not significantly different from each other. Sulfur concentrations of centrosema receiving no Mo, seed-applied Mo, soil-applied Mo and/or lime were not significantly from each other (Table 1.77). There was an interaction between legume species and Mo fertilizer treatments since Mo fertilizer decreased S concentration in the top of desmodium whereas it tended to increase S concentration of centrosema. When averaged over all Mo and lime rates, S concentration of desmodium was significantly lower than that of centrosema.

Second Cutting:

Sulfur concentration in the top of desmodium receiving no Mo was significantly higher than those of desmodium receiving various rates of seed-applied Mo, soil-applied Mo, and lime (Table 1.78). There

Table 1.77

Effects of Mo and lime on S concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (First Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	----------	% ----------	----------
No Mo added	0.23 a*	0.22 a	0.23
Lime	0.16 b	0.25 a	0.21
0.1 kg Mo/ha (seed applied)	0.17 b	0.24 a	0.21
0.3 kg Mo/ha (seed applied)	0.14 b	0.25 a	0.20
0.5 kg Mo/ha (seed applied)	0.16 b	0.23 a	0.19
0.5 kg Mo/ha (seed applied) plus lime	0.15 b	0.24 a	0.20
1.0 kg Mo/ha (soil applied)	0.14 b	0.24 a	0.19
2.0 kg Mo/ha (soil applied)	0.15 b	0.23 a	0.19
4.0 kg Mo/ha (soil applied)	0.15 b	0.24 a	0.20
4.0 kg Mo/ha (soil applied) plus lime	0.15 b	0.24 a	0.20
Mean	0.16 b	0.24 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.78

Effects of Mo and lime on S concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Second Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	---------------	% ---------------	---------------
No Mo added	0.21 c*	0.24 abc	0.22
Lime	0.16 d	0.25 a	0.21
0.1 kg Mo/ha (seed applied)	0.18 d	0.22 bc	0.20
0.3 kg Mo/ha (seed applied)	0.16 d	0.25 a	0.21
0.5 kg Mo/ha (seed applied)	0.16 d	0.24 ab	0.20
0.5 kg Mo/ha (seed applied) plus lime	0.16 d	0.26 a	0.21
1.0 kg Mo/ha (soil applied)	0.16 d	0.26 a	0.21
2.0 kg Mo/ha (soil applied)	0.17 d	0.23 abc	0.20
4.0 kg Mo/ha (soil applied)	0.17 d	0.25 a	0.21
4.0 kg Mo/ha (soil applied) plus lime	0.16 d	0.26 a	0.21
Mean	0.17 b	0.25 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

were no significant differences among S concentrations of desmodium receiving various rates of seed-applied and soil-applied Mo. The trend of S concentration of desmodium was similar to that obtained in the first cutting. Sulfur concentration in the top of centrosema receiving no Mo was not significantly different from those of centrosema receiving other rates of Mo and lime (Table 1. 78). Sulfur concentration of centrosema receiving seed-applied Mo at 0.1 kg Mo/ha was not significantly lower than those of centrosema receiving no Mo, seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 2.0 kg Mo/ha but was significantly lower than those of centrosema receiving other treatments. When averaged over all Mo and lime rates, S concentration in the top of desmodium was significantly lower than that of centrosema.

Third Cutting:

Sulfur concentration in the top of desmodium receiving no Mo was significantly higher than those of desmodium receiving various rates of seed-applied and soil-applied Mo and lime (Table 1.79). However, that concentration was not significantly higher than the concentration of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha. Sulfur concentrations in the top of centrosema receiving various rates of Mo and/or lime were not significantly different from each other (Table 1.79). It should be noted that except for 0.1 kg Mo/ha seed-applied and no-Mo treatments, S concentrations in the top of centrosema receiving other rates of Mo or lime were significantly higher than those of desmodium receiving comparable treatments. When averaged over all Mo and lime rates, S concentration of centrosema was significantly higher than that of desmodium.

Table 1.79.

Effects of Mo and lime on S concentration in the tops of <u>Desmodium intortum</u> and <u>Centrosema pubescens</u> grown in the Wahiawa soil (Third Cutting).

Mo treatment	<u>Desmodium intortum</u>	<u>Centrosema pubescens</u>	Mean
	%		
No Mo added	0.20 ab*	0.21 ab	0.21
Lime	0.16 c	0.21 ab	0.19
0.1 kg Mo/ha (seed applied)	0.19 b	0.19 ab	0.19
0.3 kg Mo/ha (seed applied)	0.16 c	0.20 ab	0.18
0.5 kg Mo/ha (seed applied)	0.16 c	0.19 ab	0.18
0.5 kg Mo/ha (seed applied) plus lime	0.16 c	0.21 ab	0.18
1.0 kg Mo/ha (soil applied)	0.16 c	0.22 a	0.19
2.0 kg Mo/ha (soil applied)	0.16 c	0.19 ab	0.18
4.0 kg Mo/ha (soil applied)	0.16 c	0.22 a	0.19
4.0 kg Mo/ha (soil applied) plus lime	0.15 c	0.20 ab	0.18
Mean	0.17 b	0.21 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Fourth Cutting

Sulfur concentration in the top of desmodium receiving no Mo was significantly higher than those of desmodium receiving different rates of seed-applied and soil-applied Mo (Table 1.80). Sulfur concentration of desmodium receiving various rates of Mo were not significantly different from each other. There were no significant differences among S concentration in the top of centrosema receiving different rates of seed-applied and soil-applied Mo (Table 1.80). When averaged over all Mo and lime rates, Mo concentration in the top of centrosema was significantly higher than that of desmodium.

Fifth Cutting:

Sulfur concentration in the top of desmodium receiving no Mo was significantly higher than those of desmodium receiving other rates of seed-applied and soil-applied Mo, and lime (Table 1.81). Sulfur concentration of desmodium receiving seed-applied Mo at 0.1 kg Mo/ha was significantly higher than those of desmodium receiving other rates of Mo. Sulfur concentrations of desmodium receiving various rates of seed-applied and soil-applied Mo were not significantly different from each other. Sulfur concentration in the top of centrosema receiving soil-applied Mo at 1.0 kg Mo/ha was significantly higher than that of centrosema receiving seed-applied Mo at 0.5 kg Mo/ha but was not significantly higher than those of centrosema receiving other rates of Mo (Table 1.81). When averaged over all Mo and lime rates, S concentration in the top of centrosema was significantly higher than that of desmodium.

Table 1.80.

Effects of Mo and lime on S concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fourth Cutting).

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	%		
No Mo added	0.25 a*	0.25 a	0.25
Lime	0.18 b	0.24 a	0.21
0.1 kg Mo/ha (seed applied)	0.23 b	0.24 a	0.23
0.3 kg Mo/ha (seed applied)	0.18 b	0.25 a	0.22
0.5 kg Mo/ha (seed applied)	0.17 b	0.24 a	0.21
0.5 kg Mo/ha (seed applied) plus lime	0.17 b	0.25 a	0.21
1.0 kg Mo/ha (soil applied)	0.17 b	0.24 a	0.21
2.0 kg Mo/ha (soil applied)	0.18 b	0.25 a	0.22
4.0 kg Mo/ha (soil applied)	0.18 b	0.24 a	0.21
4.0 kg Mo/ha (soil applied) plus lime	0.17 b	0.24 a	0.21
Mean	0.19 b	0.24 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.81.

Effects of Mo and lime on S concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fifth Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	%		
No Mo added	0.27 a*	0.23 bc	0.25
Lime	0.17 d	0.23 bc	0.20
0.1 kg Mo/ha (seed applied)	0.23 bc	0.23 bc	0.23
0.3 kg Mo/ha (seed applied)	0.18 d	0.23 bc	0.21
0.5 kg Mo/ha (seed applied)	0.17 d	0.21 c	0.19
0.5 kg Mo/ha (seed applied) plus lime	0.17 d	0.24 bc	0.21
1.0 kg Mo/ha (soil applied)	0.19 d	0.24 b	0.22
2.0 kg Mo/ha (soil applied)	0.18 d	0.23 bc	0.21
4.0 kg Mo/ha (soil applied)	0.18 d	0.23 bc	0.21
4.0 kg Mo/ha (soil applied) plus lime	0.17 d	0.23 bc	0.20
Mean	0.19 b	0.23 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Copper

First Cutting:

Molybdenum fertilizer and lime had no effect on Cu concentration in the tops of desmodium and centrosema (Table 1.82). Since there was no interaction between legume species and Mo and lime treatment, it was possible to average all legume species, Mo and lime rates. When averaged over all legume species, there were no significant differences among Cu concentrations of legumes receiving no Mo, seed-applied Mo, soil-applied Mo and lime. When averaged over all Mo and lime rates, Cu concentration in the top of centrosema was significantly higher than that of desmodium.

Second Cutting:

At this cutting Mo and lime applications significantly increased Cu concentrations in the top of desmodium and centrosema (Table 1.83). When averaged over all legume species, Cu concentrations of legumes receiving seed-applied Mo, soil-applied Mo and lime were significantly higher than those of legumes receiving no Mo. Copper concentrations of legumes receiving seed-applied Mo, soil-applied Mo and lime were not significantly different from each other. When averaged over all Mo and lime rates, Cu concentration of desmodium was significantly lower than that of centrosema.

Third Cutting:

When averaged over all legume species, Cu concentrations in the tops of legumes receiving soil-applied Mo at 4.0 kg Mo/ha, with or

Table 1.82.

Effects of Mo and lime on Cu concentration in the top of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (First Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	ppm		
No Mo added	12.0	14.3	13.2 a*
Lime	12.0	16.3	14.2 a
0.1 kg Mo/ha (seed applied)	12.3	15.3	13.8 a
0.3 kg Mo/ha (seed applied)	11.7	16.3	14.0 a
0.5 kg Mo/ha (seed applied)	11.3	15.7	13.5 a
0.5 kg Mo/ha (seed applied) plus lime	12.3	16.7	14.5 a
1.0 kg Mo/ha (soil applied)	11.3	15.7	13.5 a
2.0 kg Mo/ha (soil applied)	12.7	15.3	14.0 a
4.0 kg Mo/ha (soil applied)	12.0	17.0	14.5 a
4.0 kg Mo/ha (soil applied) plus lime	11.0	16.3	13.7 a
Mean	11.9 b	15.9 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.83.

Effects of Mo and lime on Cu concentration in the top of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Second Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
		ppm	
No Mo added	13.3	14.0	13.7 b*
Lime	15.3	22.3	18.8 a
0.1 kg Mo/ha (seed applied)	15.0	20.0	17.5 a
0.3 kg Mo/ha (seed applied)	16.7	19.3	18.0 a
0.5 kg Mo/ha (seed applied)	17.0	18.7	17.8 a
0.5 kg Mo/ha (seed applied) plus lime	16.7	20.3	18.5 a
1.0 kg Mo/ha (soil applied)	15.0	20.0	17.5 a
2.0 kg Mo/ha (soil applied)	18.3	19.0	18.7 a
4.0 kg Mo/ha (soil applied)	17.3	21.3	19.3 a
4.0 kg Mo/ha (soil applied) plus lime	17.0	21.3	19.2 a
Mean	16.2 b	19.6 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

without lime, were not significantly different from each other (Table 1.84). Copper concentrations of legumes receiving the two treatments were significantly higher than those of legumes receiving soil-applied Mo at 2.0 kg Mo/ha and no Mo. Copper concentrations of legumes receiving various rates of seed-applied Mo were not significantly different from each other. Copper concentrations of legumes receiving seed-applied Mo were also not significantly different from those of legumes receiving soil-applied Mo. When averaged over all Mo and lime rates, Cu concentration in the top of desmodium was significantly lower than that of centrosema.

Fourth Cutting:

When averaged over all legume species, Cu concentration in the top of legumes receiving no Mo was significantly lower than those of legumes receiving lime and soil-applied Mo at 2.0 kg Mo/ha (Table 1.85). However, Cu concentration of legumes receiving no Mo was not significantly different from those of legumes receiving various rates of seed-applied Mo and soil-applied Mo other than the 2.0 kg Mo/ha rate. Copper concentration of legumes receiving seed-applied Mo, soil-applied Mo, and lime were not significantly different from each other. When averaged over all Mo and lime rates, Cu concentration in the top of desmodium was not significantly different from that of centrosema. It should be noted that Cu concentration in the top of legumes was low as compared to the Cu concentration obtained from the previous cuttings. This was probably due to the different growth stage of legumes since both legumes had flowers and pods at this cutting.

Table 1.84.

Effects of Mo and lime on Cu concentration in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Third Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	ppm		
No Mo added	10.3	12.3	11.3 bc*
Lime	11.7	15.0	13.3 ab
0.1 kg Mo/ha (seed applied)	12.0	13.0	12.5 abc
0.3 kg Mo/ha (seed applied)	12.0	13.0	12.5 abc
0.5 kg Mo/ha (seed applied)	12.7	12.0	12.3 abc
0.5 kg Mo/ha (seed applied) plus lime	12.3	12.7	12.5 abc
1.0 kg Mo/ha (soil applied)	11.7	12.0	11.8 abc
2.0 kg Mo/ha (soil applied)	11.0	10.7	10.8 c
4.0 kg Mo/ha (soil applied)	12.7	14.7	13.7 a
4.0 kg Mo/ha (soil applied) plus lime	12.7	15.3	14.0 a
Mean	11.9 b	13.1 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.85.

Effects of Mo and lime on Cu concentration in the tops of <u>Desmodium intortum</u> and <u>Centrosema pubescens</u> grown in the Wahiawa soil (Fourth Cutting)

Mo treatment	<u>Desmodium intortum</u>	<u>Centrosema pubescens</u>	Mean
	----------------	ppm	----------------
No Mo added	2.3	4.3	3.3 b*
Lime	5.3	5.7	5.5 a
0.1 kg Mo/ha (seed applied)	3.3	5.3	4.3 ab
0.3 kg Mo/ha (seed applied)	4.7	6.0	5.3 ab
0.5 kg Mo/ha (seed applied)	5.0	4.0	4.5 ab
0.5 kg Mo/ha (seed applied) plus lime	6.0	4.7	5.3 ab
1.0 kg Mo/ha (soil applied)	4.0	5.7	4.8 ab
2.0 kg Mo/ha (soil applied)	4.7	6.7	5.7 a
4.0 kg Mo/ha (soil applied)	5.0	4.7	4.8 ab
4.0 kg Mo/ha (soil applied) plus lime	5.7	4.0	4.8 ab
Mean	4.6 a	5.1 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Fifth Cutting:

When averaged over all legume species, there was no significance difference among Cu concentrations of legumes receiving no Mo, seed-applied Mo, soil-applied Mo and lime (Table 1.86). When averaged over all Mo and lime rates, Cu concentration in the top of desmodium was significantly lower than that of centrosema.

Copper to Molybdenum Ratio

First Cutting:

Molybdenum application at high rates significantly decreased Cu/Mo ratio in the tops of desmodium and centrosema (Table 1.87). When averaged over all Mo and lime rates, Cu/Mo ratios of legumes receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were significantly lower than those of legumes receiving no Mo and lime. Cu/Mo ratios of legumes receiving various rates of soil-applied and seed-applied were not significantly different from each other. When averaged over all Mo and lime rates, Cu/Mo ratio in the top of desmodium was significantly lower than that of centrosema.

Second Cutting:

At this cutting, Mo application at high rates still had an influence on Cu/Mo ratios in the tops of desmodium and centrosema (Table 1.88). When averaged over all legume species, Cu/Mo ratio in the top of legumes receiving soil-applied Mo at 4.0 kg Mo/ha was significantly lower than those of legumes receiving seed-applied Mo, lime and no Mo. Cu/Mo ratios of legumes receiving various rates of soil-applied Mo were

Table 1.86.

Effects of Mo and lime on Cu concentration in the tops of _Desmodium intortum_ and _Centrosema pubescens_ grown in the Wahiawa soil (Fifth Cutting)

Mo treatment	_Desmodium intortum_	_Centrosema pubescens_	Mean
	ppm		
No Mo added	12.0	13.3	12.7 a*
Lime	11.0	14.0	12.5 a
0.1 kg Mo/ha (seed applied)	11.0	14.0	12.5 a
0.3 kg Mo/ha (seed applied)	10.3	14.0	12.2 a
0.5 kg Mo/ha (seed applied)	10.7	13.0	11.8 a
0.5 kg Mo/ha (seed applied) plus lime	10.3	14.0	12.2 a
1.0 kg Mo/ha (soil applied)	11.0	13.0	12.0 a
2.0 kg Mo/ha (soil applied)	11.7	14.0	12.8 a
4.0 kg Mo/ha (soil applied)	12.3	13.3	12.8 a
4.0 kg Mo/ha (soil applied) plus lime	10.7	13.3	12.0 a
Mean	11.1 b	13.6 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.87

Effects of Mo and lime on Cu/Mo ratio in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (First Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	Cu/Mo		
No Mo added	116	331	223 a*
Lime	70	250	160 a
0.1 kg Mo/ha (seed applied)	103	165	134 abc
0.3 kg Mo/ha (seed applied)	91	164	127 abc
0.5 kg Mo/ha (seed applied)	102	145	123 abc
0.5 kg Mo/ha (seed applied) plus lime	140	142	141 abc
1.0 kg Mo/ha (soil applied)	85	130	107 abc
2.0 kg Mo/ha (soil applied)	41	66	53 bc
4.0 kg Mo/ha (soil applied)	24	48	36 c
4.0 kg Mo/ha (soil applied) plus lime	22	25	23 c
Mean	79 b	146 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.88.

Effects of Mo and lime on Cu/Mo ratio in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Second Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
		Cu/Mo	
No Mo added	213	200	207 a*
Lime	127	189	158 ab
0.1 kg Mo/ha (seed applied)	165	201	183 a
0.3 kg Mo/ha (seed applied)	123	253	188 a
0.5 kg Mo/ha (seed applied)	102	167	135 ab
0.5 kg Mo/ha (seed applied) plus lime	102	168	135 ab
1.0 kg Mo/ha (soil applied)	101	148	125 abc
2.0 kg Mo/ha (soil applied)	68	112	90 abc
4.0 kg Mo/ha (soil applied)	38	62	50 cd
4.0 kg Mo/ha (soil applied) plus lime	32	46	39 d
Mean	107 b	155 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

not significantly different from each other. Cu/Mo ratios of legumes receiving various rates of seed-applied Mo were also not significantly different from each other. When averaged over all Mo and lime rates, Cu/Mo ratio in the top of desmodium was significantly lower than that of centrosema.

Third Cutting:

When averaged over all legume species, although Cu/Mo ratio in the top of legumes receiving no Mo was not significantly different from those of legumes receiving lime and seed-applied Mo at 0.1 kg Mo/ha, it was significantly lower than those of legumes receiving other rates of Mo (Table 1.89). Cu/Mo ratios of legumes receiving seed-applied Mo at various rates were not significantly different from each other. When averaged over all Mo and lime rates, Cu/Mo ratios of desmodium and centrosema were not significantly different from each other.

Fourth Cutting:

When averaged over all legume species, Cu/Mo ratio in the top of legumes receiving no Mo was not significantly different from those of legumes receiving other rates of Mo (Table 1.90). Cu/Mo ratios of legumes receiving various rates of seed-applied Mo and soil-applied Mo were not significantly different from each other. Cu/Mo ratio of legumes receiving lime was significantly higher than those of legumes receiving soil-applied Mo at various rates. However, Cu/Mo ratio of legumes receiving lime was not significantly different from those of legumes receiving seed-applied Mo. When averaged over all Mo and lime rates, Cu/Mo ratios of desmodium and centrosema were not significantly different from each other.

Table 1.89.

Effects of Mo and lime on Cu/Mo ratio in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Third Cutting)

Mo treatment	Desmodium intortum	Centrosema pubescens	Mean
	Cu/Mo		
No Mo added	542	267	404 a*
Lime	210	150	180 ab
0.1 kg Mo/ha (seed applied)	625	169	397 a
0.3 kg Mo/ha (seed applied)	187	115	151 b
0.5 kg Mo/ha (seed applied)	119	93	106 b
0.5 kg Mo/ha (seed applied) plus lime	116	86	101 b
1.0 kg Mo/ha (soil applied)	91	82	86 b
2.0 kg Mo/ha (soil applied)	48	44	46 b
4.0 kg Mo/ha (soil applied)	21	40	31 b
4.0 kg Mo/ha (soil applied) plus lime	17	32	24 b
Mean	197 a	108 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 1.90.

Effects of Mo and lime on the Cu/Mo ratio in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa Soil (Fourth Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	Cu/Mo		
No Mo added	54	59	57 ab*
Lime	265	53	159 a
0.1 kg Mo/ha (seed applied)	160	39	100 ab
0.3 kg Mo/ha (seed applied)	80	55	67 ab
0.5 kg Mo/ha (seed applied)	73	30	52 ab
0.5 kg Mo/ha (seed applied) plus lime	84	35	59 ab
1.0 kg Mo/ha (soil applied)	25	44	34 b
2.0 kg Mo/ha (soil applied)	20	37	29 b
4.0 kg Mo/ha (soil applied)	8	12	10 b
4.0 kg Mo/ha (soil applied) plus lime	9	10	10 b
Mean	78 a	37 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

It should be noted that Cu/Mo ratios of legumes at this cutting were markedly lower than those of legumes at the previous cuttings. This difference was likely due to the difference in the growth stage at harvest. Legumes at this cutting had flowers and pods whereas legumes at the previous cuttings did not reach this stage of development.

Fifth Cutting:

When averaged over all legume species, Cu/Mo ratio in the top of legumes receiving no Mo was significantly higher than those of legumes receiving other rates of Mo and lime (Table 1.91). Cu/Mo ratios of legumes receiving various rates of soil-applied Mo were not significantly different from each other. Cu/Mo ratio of legumes receiving seed-applied Mo at 0.1 kg Mo/ha was significantly higher than that of legumes receiving seed-applied Mo at 0.5 kg Mo/ha but was not significantly higher than that of legumes receiving seed-applied Mo at 0.3 kg Mo/ha. Cu/Mo ratio of legumes receiving lime was significantly lower than that of legumes receiving no Mo but was not significantly different from those of legumes receiving other treatments. When averaged over all Mo and lime rates, Cu/Mo ratios of desmodium and centrosema were not significantly different from each other. It should be noted that Cu/Mo ratio of this cutting was relatively higher than those of the previous cuttings. This effect was likely due to the fact that Mo concentration in the top of legumes from this cutting was lower than those of legumes from the previous cuttings.

Table 1.91.

Effects of Mo and lime on the Cu/Mo ratio in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fifth Cutting)

Mo treatment	*Desmodium intortum*	*Centrosema pubescens*	Mean
	Cu/Mo		
No Mo added	925	369	647 a*
Lime	274	276	275 bcd
0.1 kg Mo/ha (seed applied)	586	356	471 ab
0.3 kg Mo/ha (seed applied)	496	227	361 bc
0.5 kg Mo/ha (seed applied)	155	184	170 cd
0.5 kg Mo/ha (seed applied) plus lime	179	192	186 cd
1.0 kg Mo/ha (soil applied)	243	260	252 cd
2.0 kg Mo/ha (soil applied)	84	231	157 cd
4.0 kg Mo/ha (soil applied)	38	47	42 d
4.0 kg Mo/ha (soil applied) plus lime	35	64	50 d
Mean	302 a	221 a	

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Relationship between Dry Matter Yield and Mo Concentration in Plant Top

First Cutting:

There was no significant correlation between dry matter yield and Mo concentration in the top of desmodium. The low coefficient of determination (r^2) of 0.311 indicated that only 31% of the dry matter yield data correlated with Mo concentration in plant top. However, according to a scatter diagram, in the Mo concentration range of 0 to 0.3 ppm dry matter yield increased as the Mo concentration increased (Fig. 1.46). There was also no significant correlation between yield and Mo concentration in the the top of centrosema. The correlation coefficient of the relationship was 0.310 ($p = 0.70$). The r^2 of this relationship was 0.096. The fact that r^2 of the relationship of desmodium was higher than that of centrosema suggested that yield of desmodium was more correlated to Mo concentration than in centrosema.

There was no significant correlation between relative yield and Mo concentration in the top of desmodium. The correlation coefficient of the relationship was 0.558 ($p = 0.27$). According to a scatter diagram, in the Mo concentration range of 0 to 0.3 ppm, relative yield increased as Mo concentration increased (Fig. 1.47). There was also no significant correlation between relative yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.309 ($p = 0.70$).

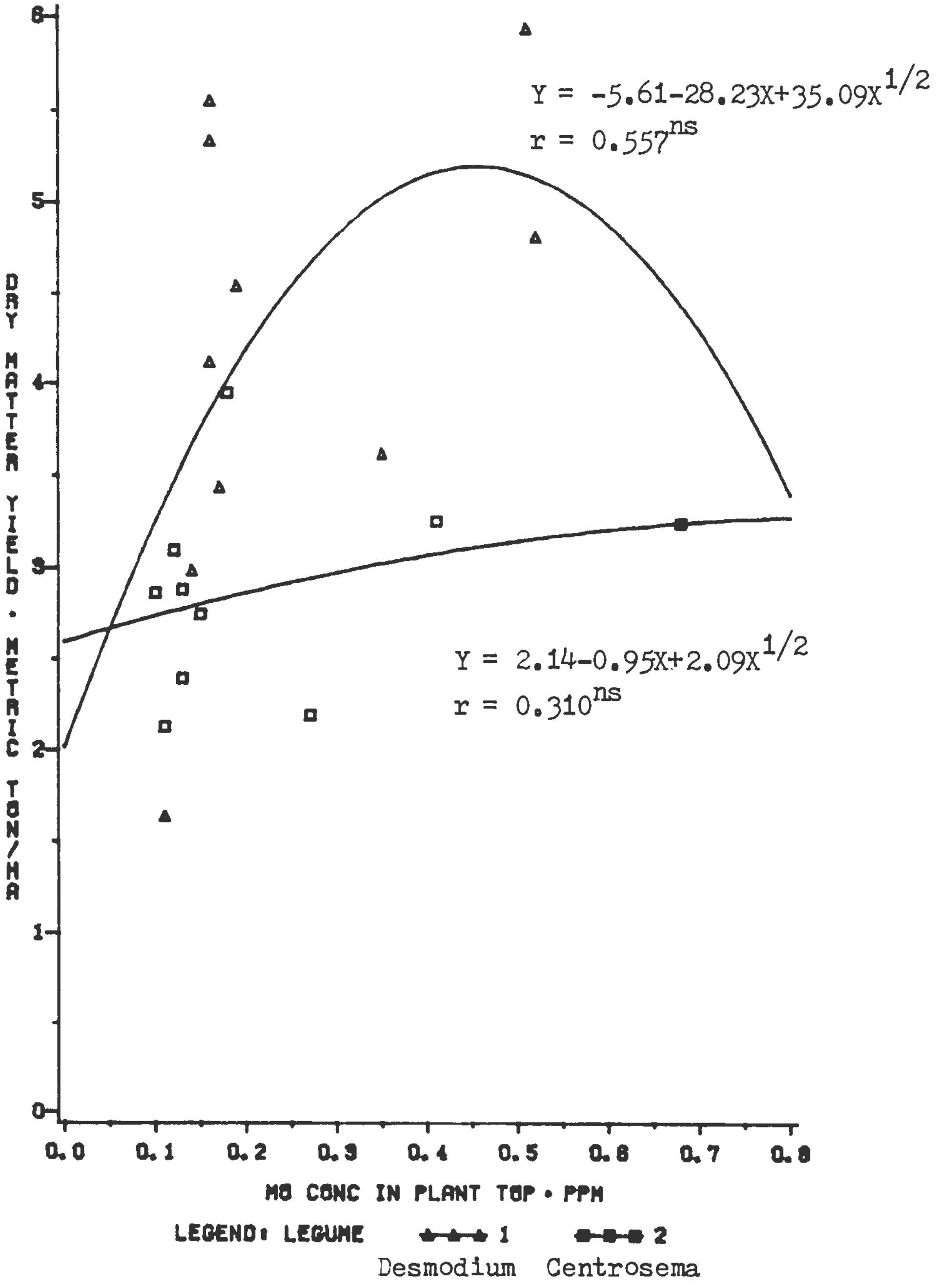


Fig. 1.46. Relationship between dry matter yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (First cutting).

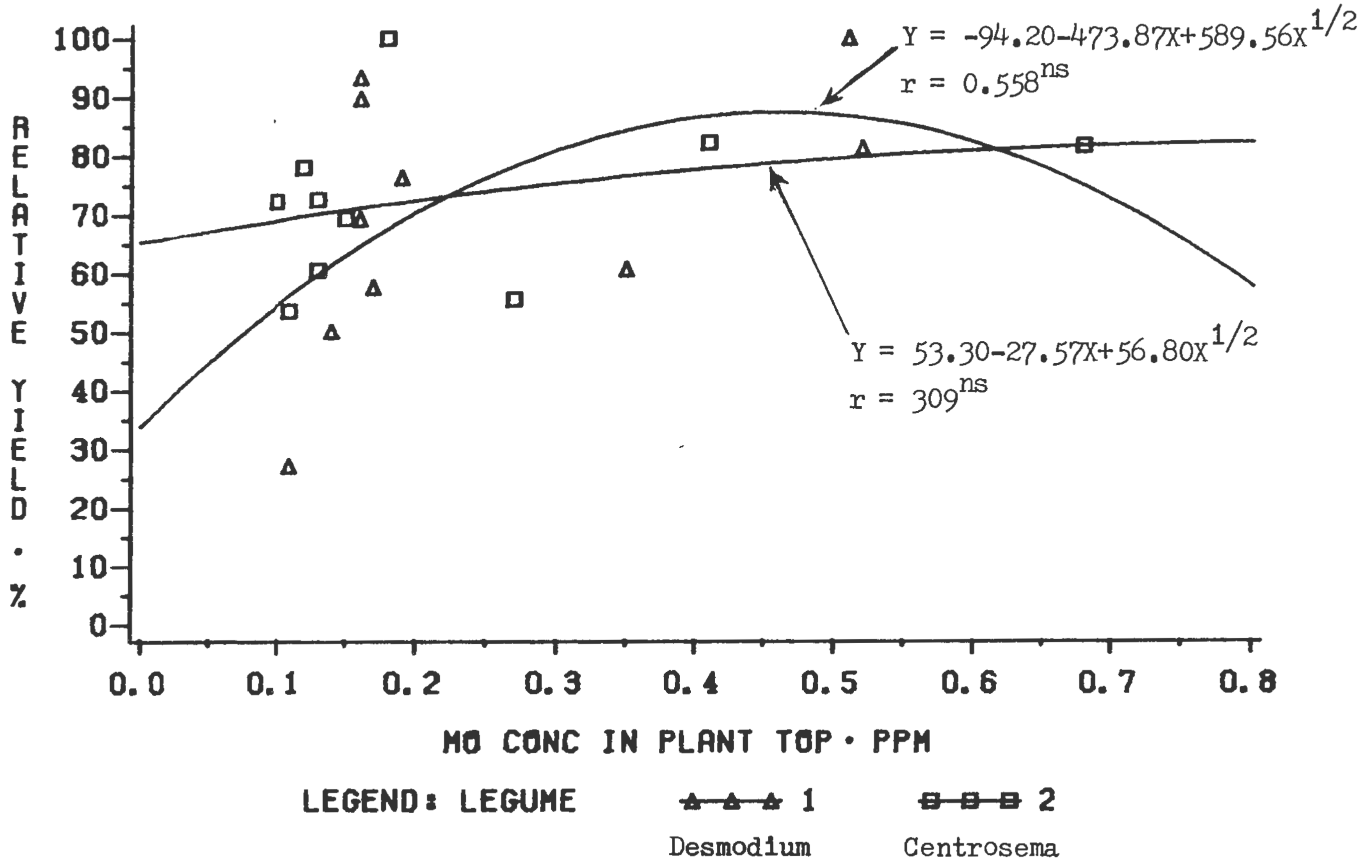


Fig. 1.47. Relationship between relative yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (First cutting).

Second Cutting:

There was no significant correlation between dry matter yield and Mo concentration in the top of desmodium at this cutting. The correlation coefficient of the relationship was 0.681 (p = 0.11). However the scatter diagram showed that yield of desmodium tended to increase as Mo concentration increased in the Mo concentration range of 0 to 0.3 ppm (Fig. 1.48). There was also no significant correlation between yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.322 (p = 0.68).

There was also no significant correlation between relative yield and Mo concentration of desmodium. The correlation coefficient of the relationship was 0.680 (p = 0.11). According to a scatter diagram (Fig. 1.49), relative yield increased as Mo concentration increased in the Mo range of 0 to 0.3 ppm. There was also no significant correlation between relative yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.292 (p = 0.73).

Third Cutting:

There was a highly significant correlation between dry matter yield and Mo concentration in the top of desmodium with the correlation coefficient of 0.840. According to a scatter diagram (Fig. 1.50), yield increased as the Mo concentration increased within the Mo concentration range of 0 to 0.3 ppm. As the Mo concentration increased above 0.45 ppm, yield tended to decline. There was no significant correlation between yield and Mo concentration of centrosema. The correlation coefficient of the relationship was 0.226 (p = 0.83).

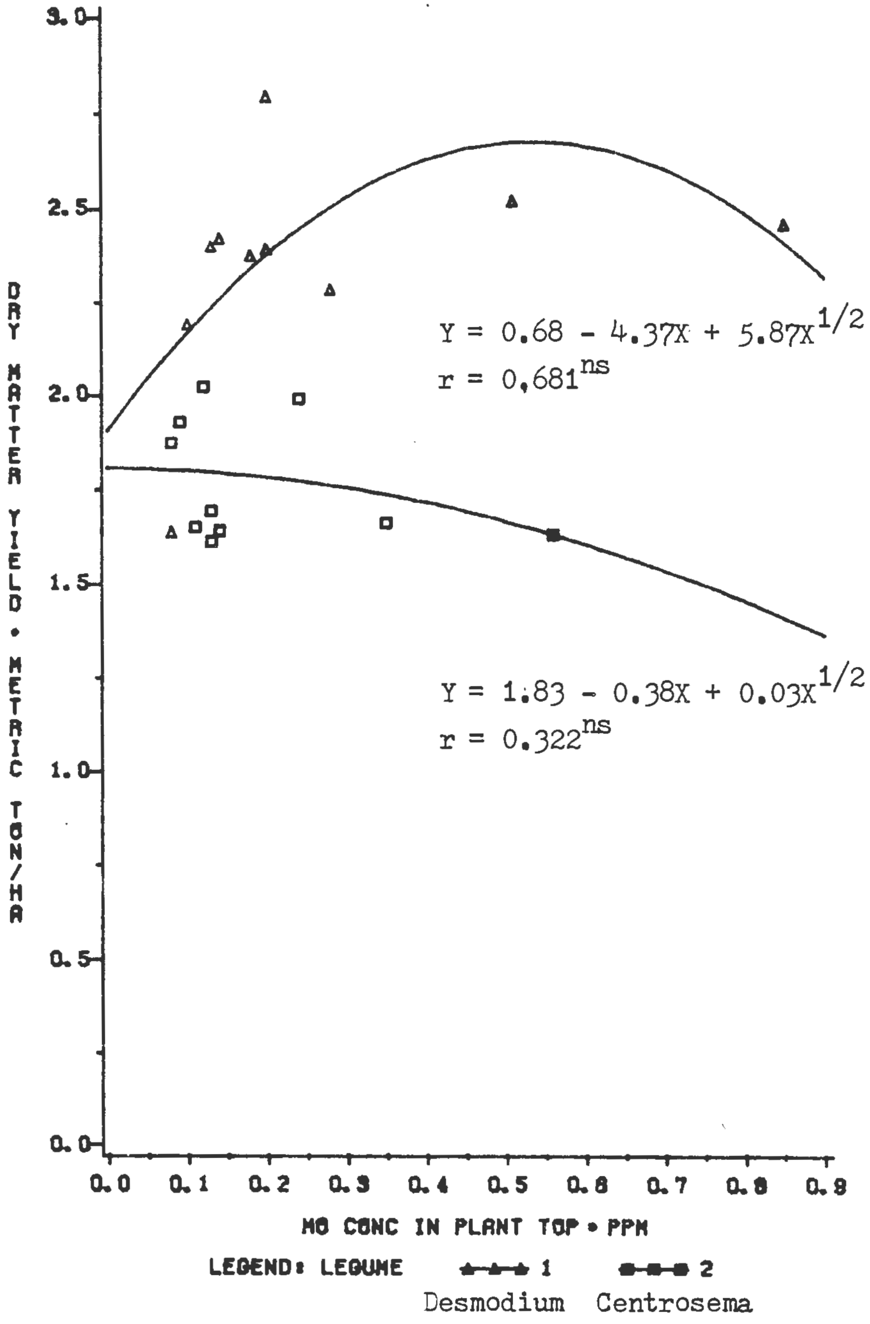


Fig. 1.48. Relationship between dry matter yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Second cutting).

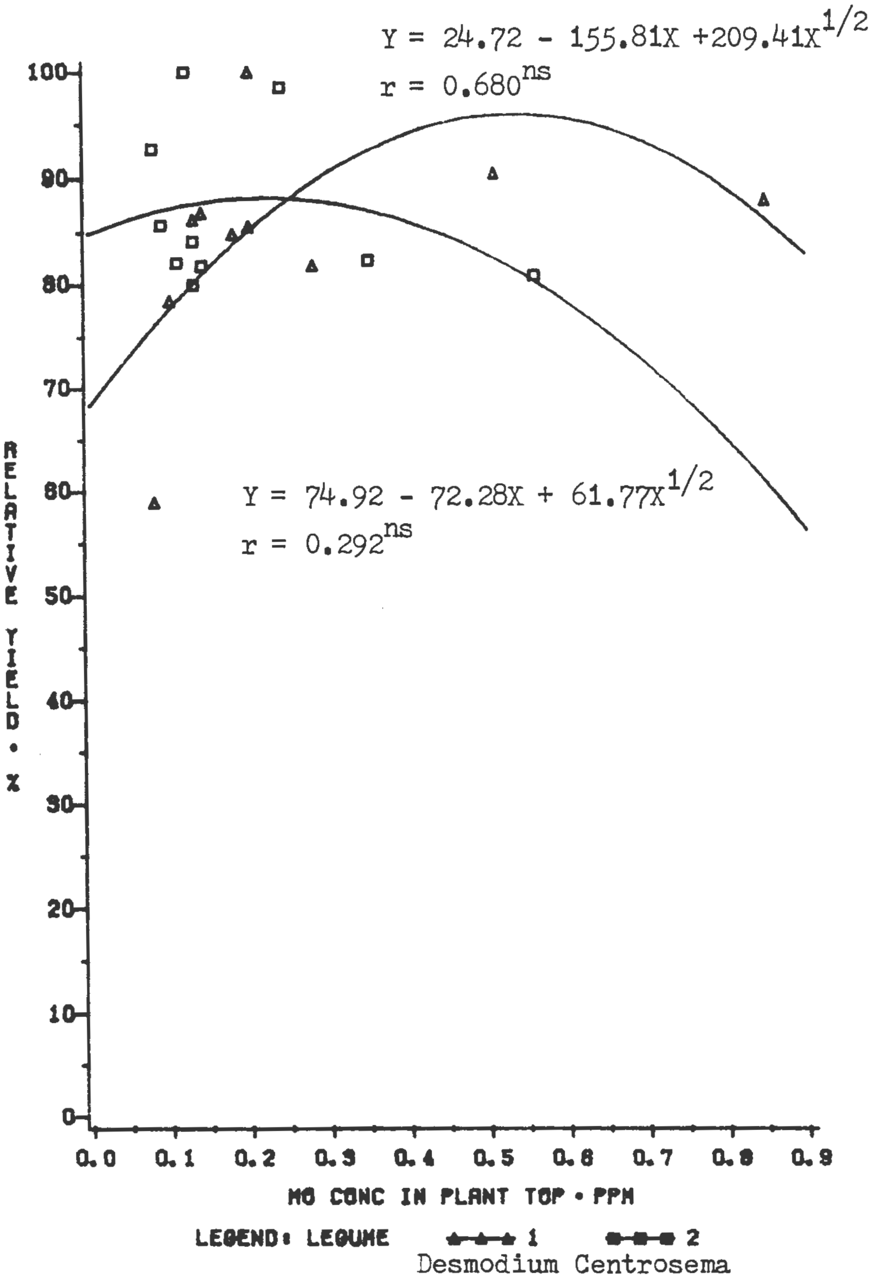


Fig. 1.49. Relationship between relative yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Second cutting).

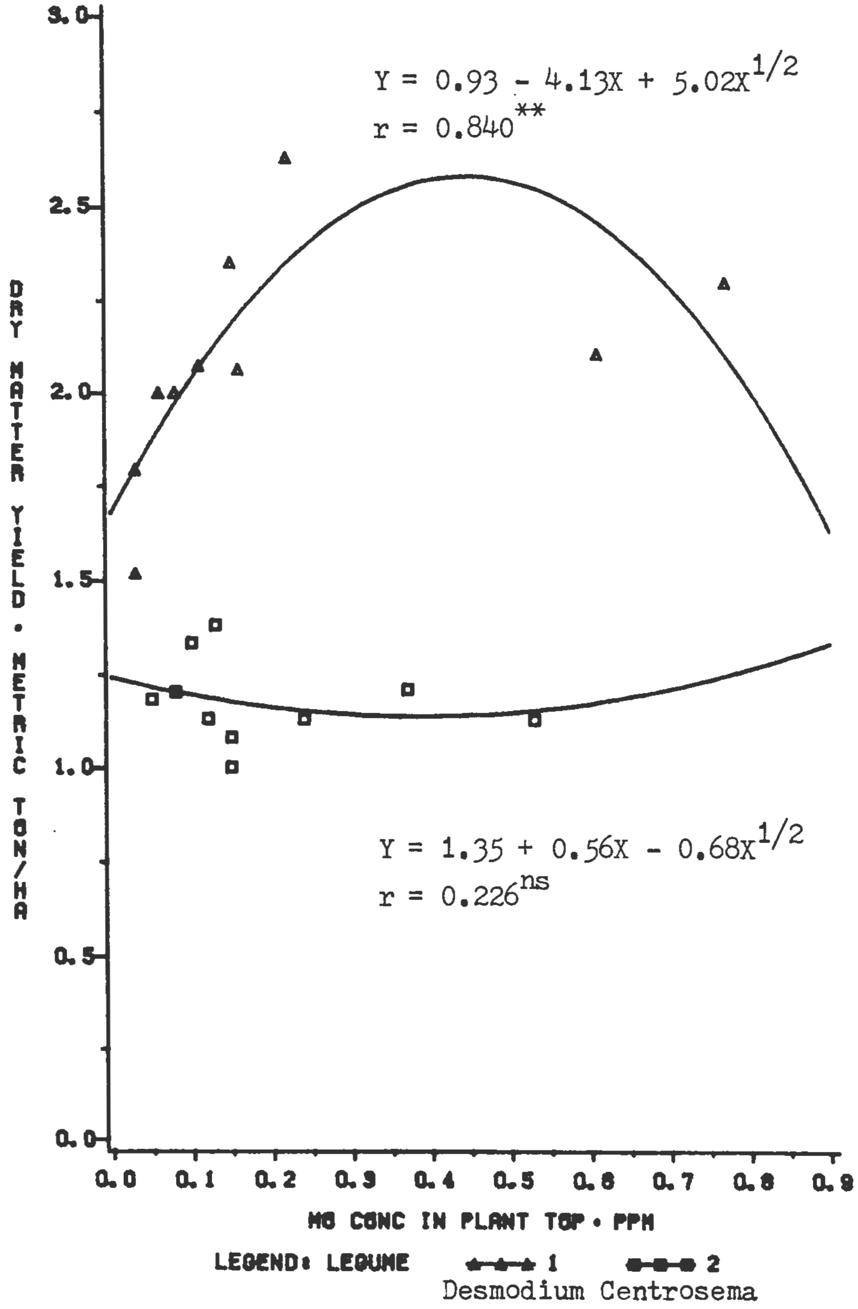


Fig. 1.50. Relationship between dry matter yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Third cutting).

There was a highly significant correlation between relative yield and Mo concentration in the top of desmodium with the correlation coefficient of 0.838. According to a scatter diagram (Fig. 1.51), Mo concentrations of 0.13 and 0.3 ppm were associated with 80% and 95% maximum yields, respectively. There was no significant correlation between relative yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.228 (p = 0.83). The approximate critical Mo level could not be defined for centrosema at this cutting.

Fourth Cutting:

There was no significant correlation between dry matter yield and Mo concentration in the top of desmodium. The correlation coefficient of the relationship was 0.710 (p = 0.09). According to a scatter diagram, yield of desmodium tended to increase as Mo concentration increased (Fig. 1.52). There was also no significant correlation between yield and Mo concentration of centrosema. The correlation coefficient of the relationship was 0.627 (p = 0.17).

There was also no significant correlation between relative yield and Mo concentration in the top of desmodium (Fig. 1.53). The correlation coefficient of the relationship was 0.710 (p = 0.09). There was no significant correlation between relative yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.627 (p = 0.17).

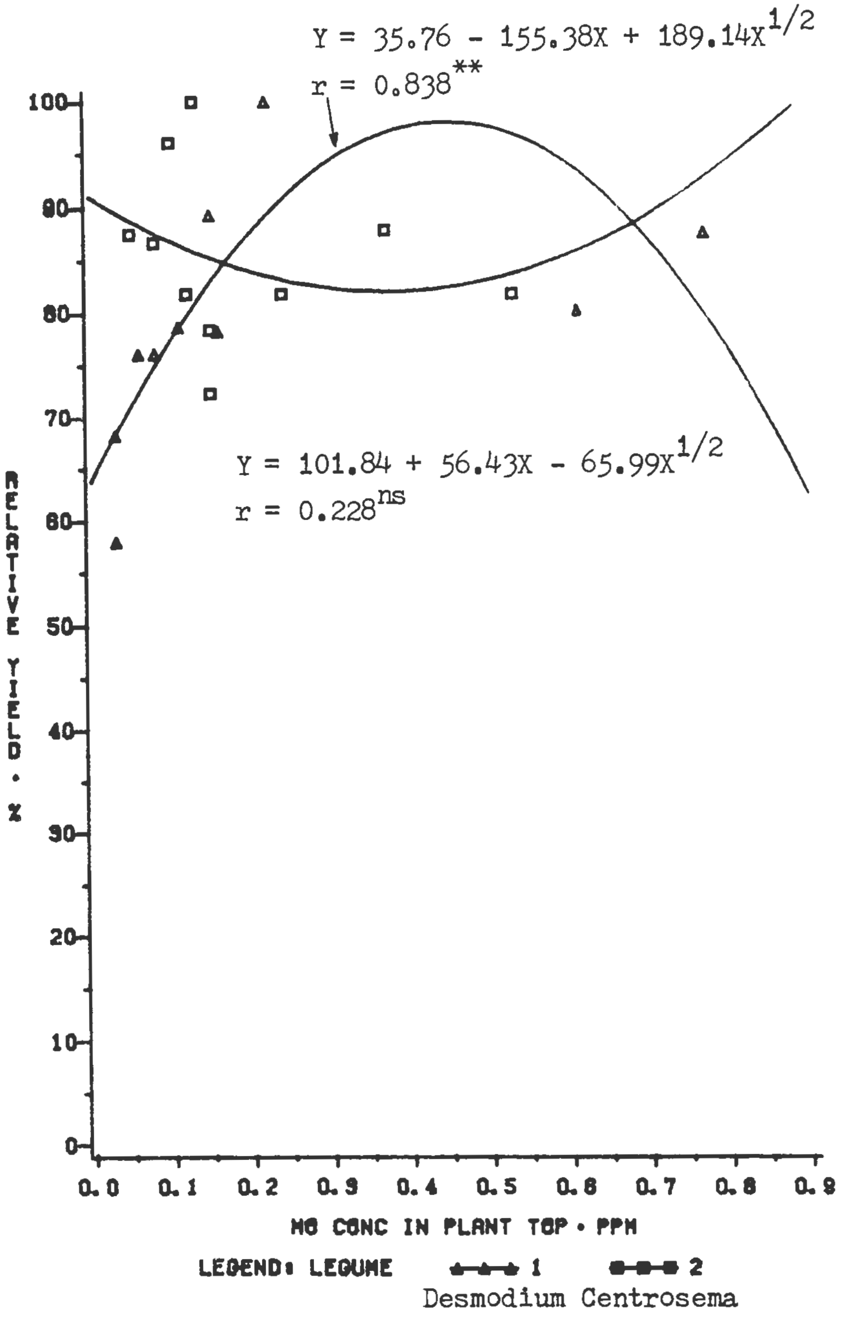


Fig. 1.51. Relationship between relative yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Third cutting).

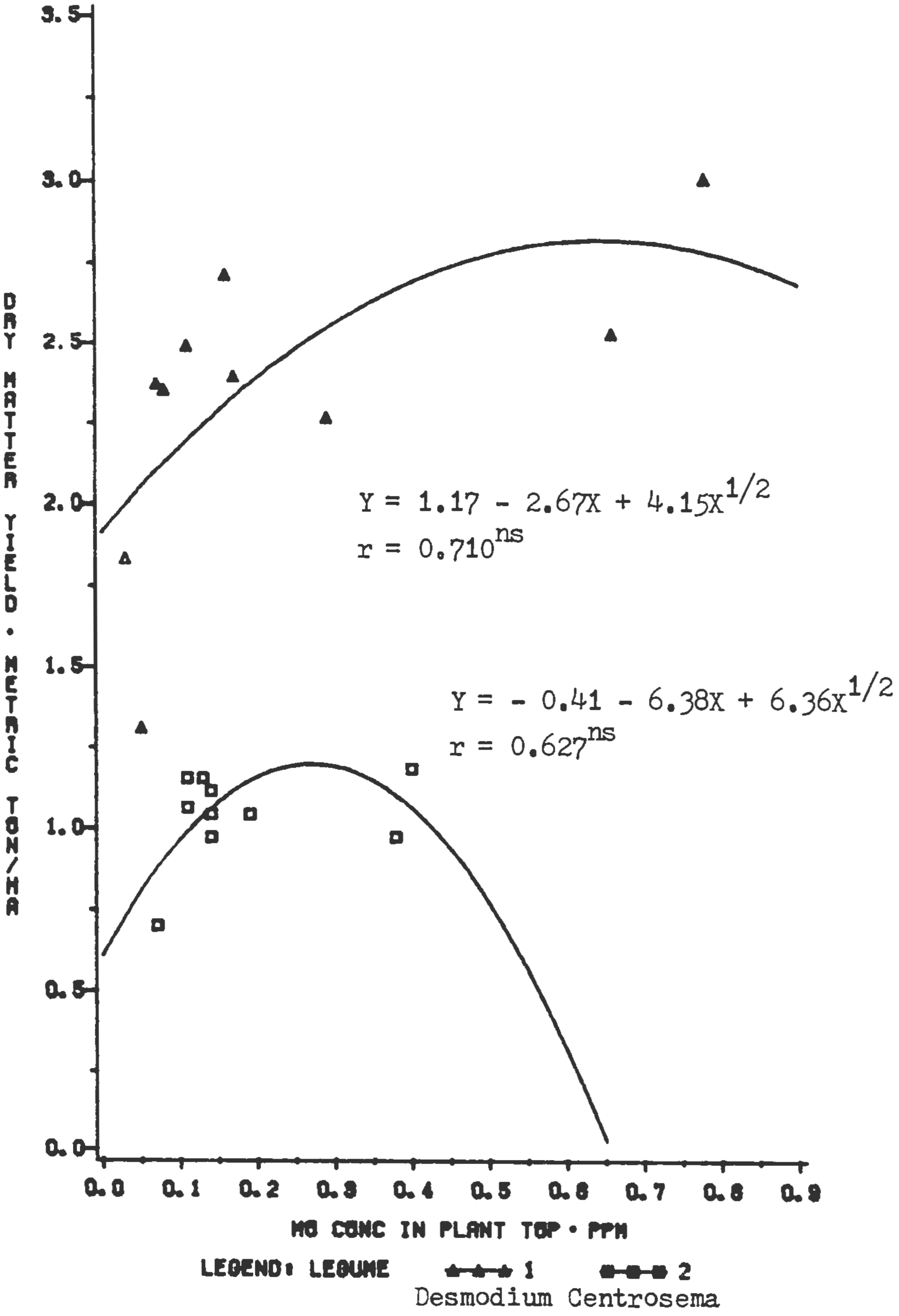


Fig. 1.52. Relationship between dry matter yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fourth cutting).

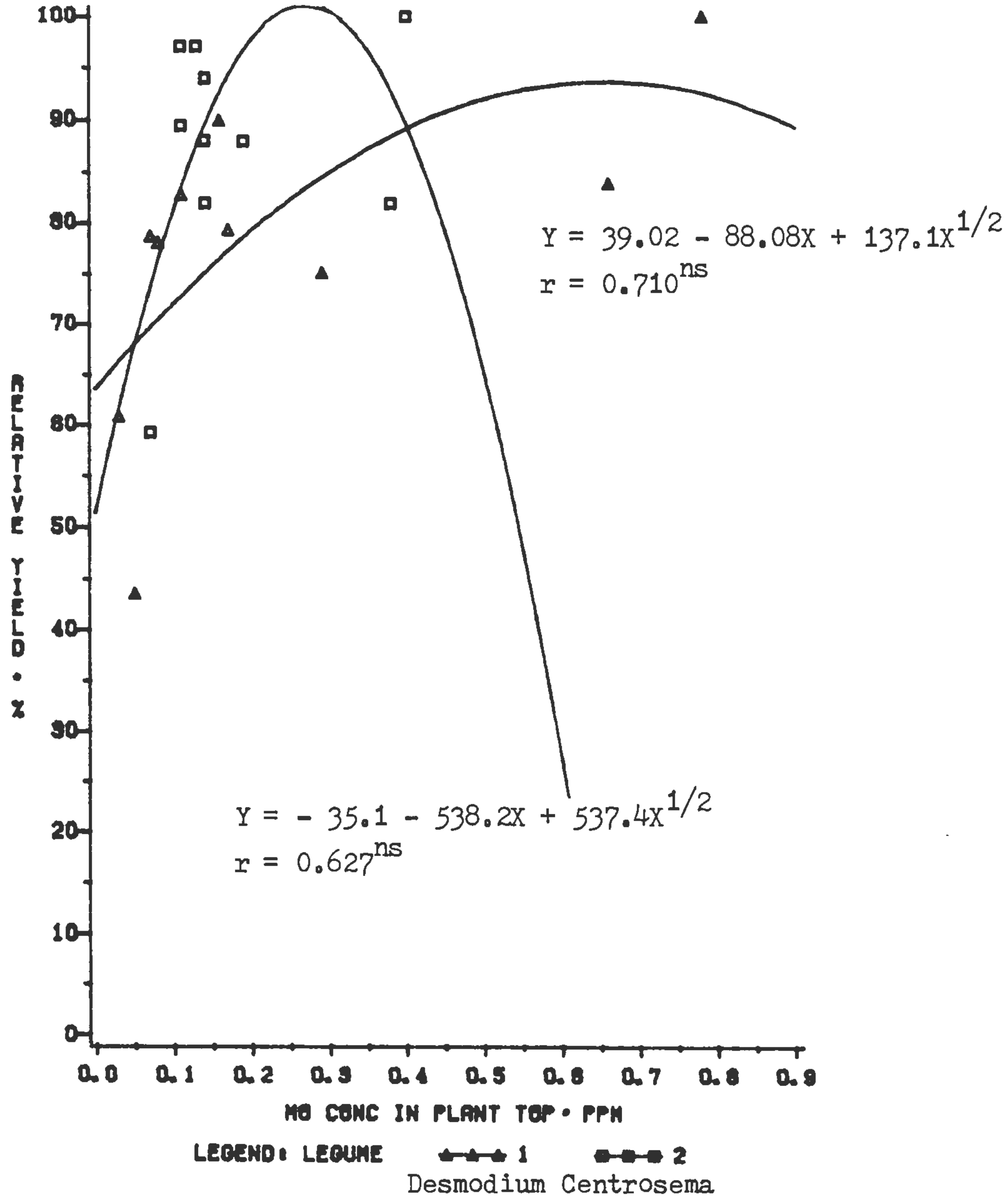


Fig. 1.53. Relationship between relative yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fourth cutting).

Fifth Cutting:

There was no significant correlation between dry matter yield and Mo concentration in the top of desmodium. The correlation coefficient of the relationship was 0.738 ($p = 0.06$). Although there was no significant correlation, yield of desmodium tended to increase as Mo concentration increased within the Mo concentration range of 0 to 0.15 ppm (Fig. 1.54). It should be noted that the critical Mo concentration range was lower than the range from the previous cuttings. The low level of Mo concentration in the top may have been caused by the low supply of Mo in soil and/or high requirement for Mo of nodule. Yield of desmodium receiving Mo fertilizer was still high despite low Mo concentration in the top, suggesting that Mo concentration in nodule was still adequate for the requirement of plant for nitrogen fixation. There was no significant correlation between yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.297 ($p = 0.72$).

There was no significant correlation between relative yield and Mo concentration in the top of desmodium. The correlation coefficient of the relationship was 0.740 ($p = 0.06$). Despite the lack of significance, it can be estimated that critical Mo concentration was in the range of 0.05 to 0.15 ppm (Fig. 1.55). There was also no significant correlation between relative yield and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.344 ($p = 0.64$)

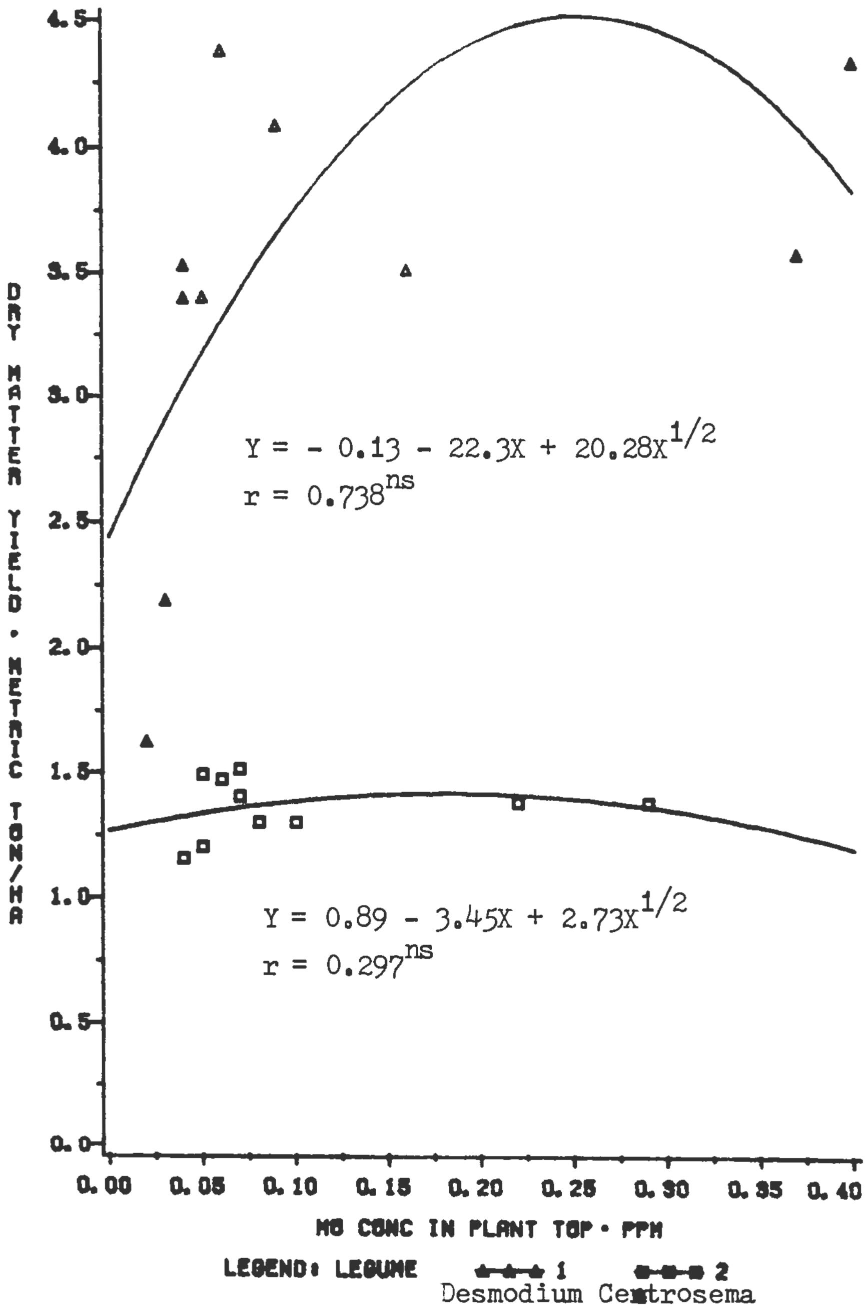


Fig. 1.54. Relationship between dry matter yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fifth cutting).

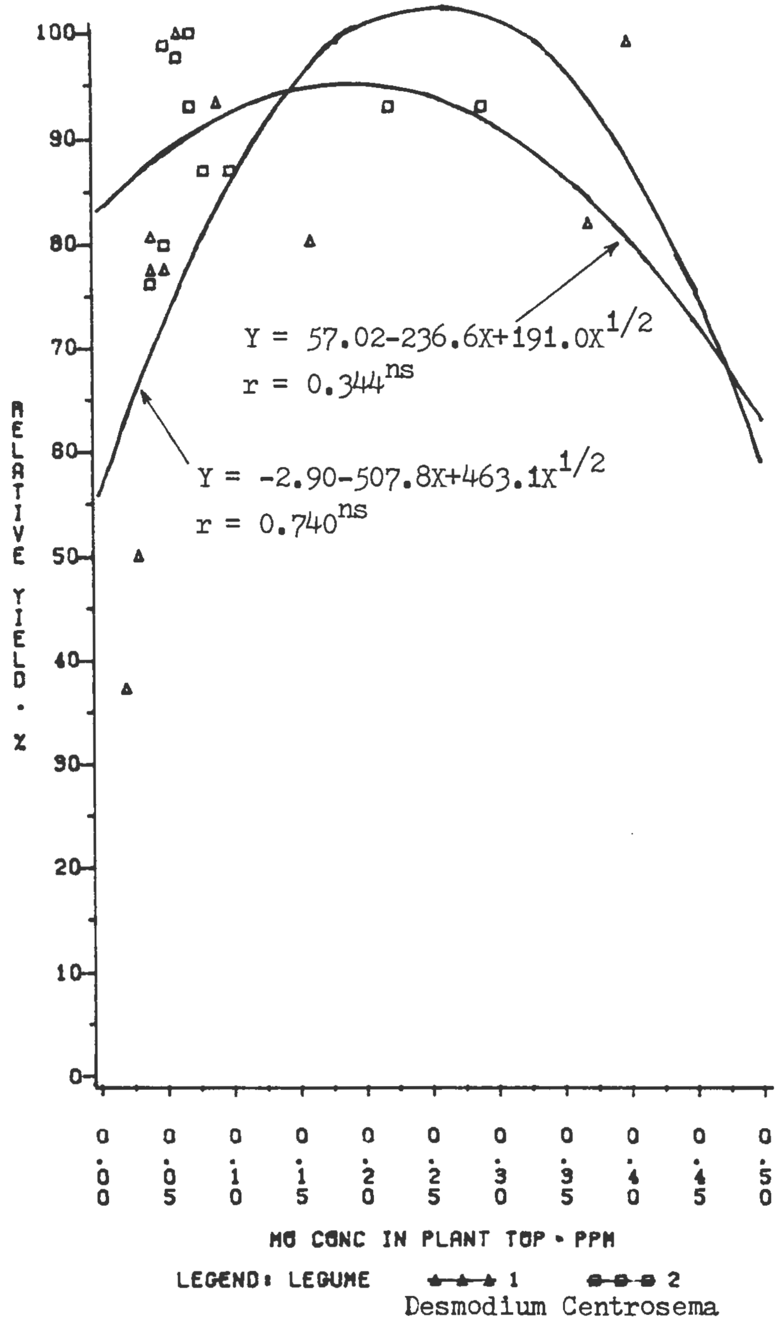


Fig. 1.55. Relationship between relative yields and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Fifth cutting).

Relationship between N and Mo Concentrations in Plant Top

First Cutting:

There was no significant correlation between N and Mo concentrations in the top of desmodium. The correlation coefficient of the relationship was 0.739 (p = 0.06). Despite the lack of significance, the scatter diagram showed a tendency that in the Mo concentration range of 0 to 0.3 ppm, N concentration increased as the Mo concentration increased (Fig. 1.56). There was also no significant correlation between N and Mo concentrations in the top of centrosema. The correlation coefficient of the relationship was 0.531 (p = 0.31).

Second Cutting:

There was a highly significant correlation between N and Mo concentrations in the top of desmodium with the correlation coefficient of 0.915. The scatter diagram indicated that in the Mo concentration range of 0 to 0.35, N concentration increases as Mo concentration increased (Fig. 1.57). Nitrogen concentration declined as Mo concentration increased above 0.35 ppm. There was no significant correlation between N and Mo concentrations in the top of centrosema. The correlation coefficient of the relationship was 0.684 (p = 0.10).

Third Cutting:

There was a highly significant correlation between N and Mo concentrations in the top of desmodium with the correlation coefficient of 0.875. Nitrogen concentration increased as Mo concentration increased within the Mo range of 0 to 0.4 ppm (Fig. 1.58). Nitrogen concentration

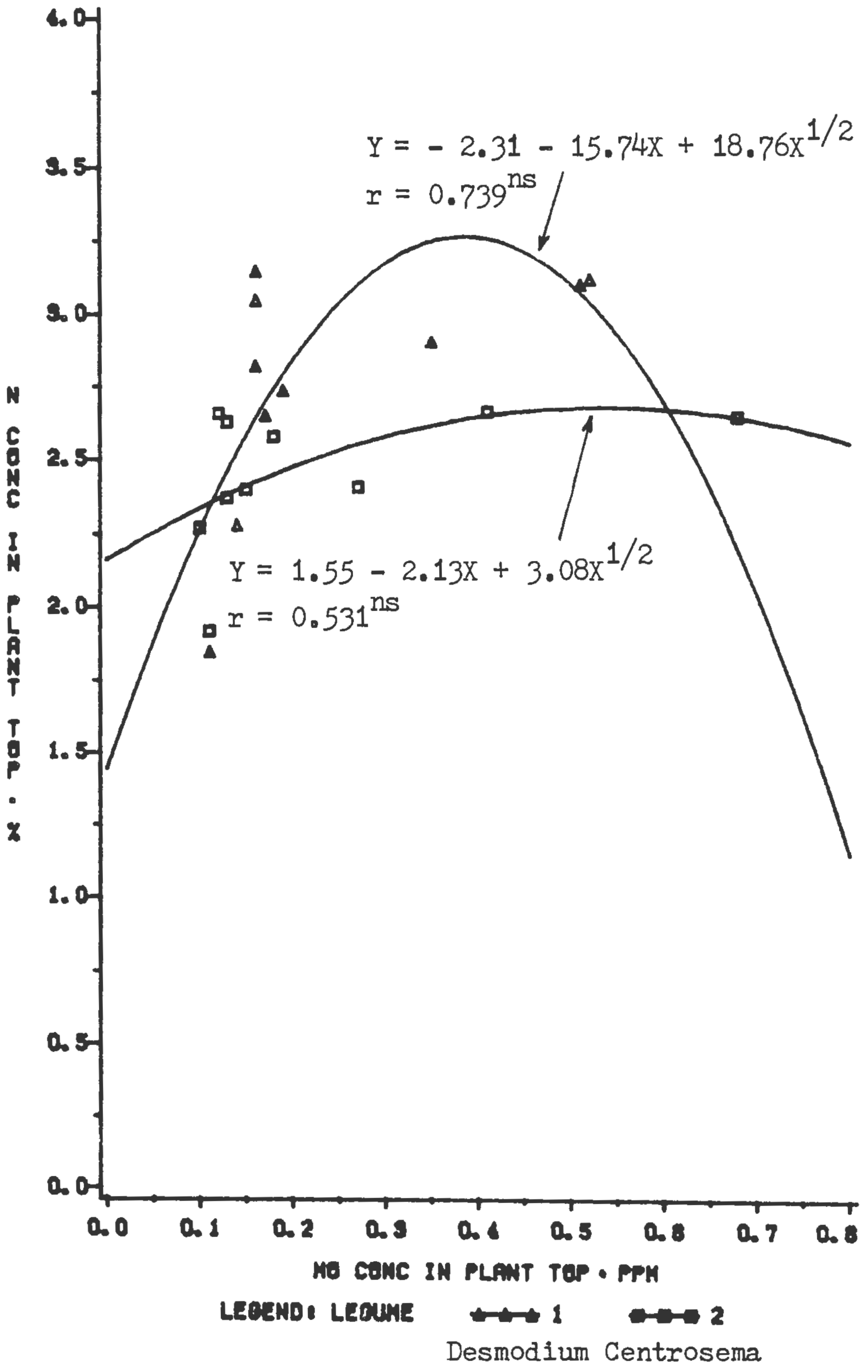


Fig. 1.56. Relationship between N and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (First cutting).

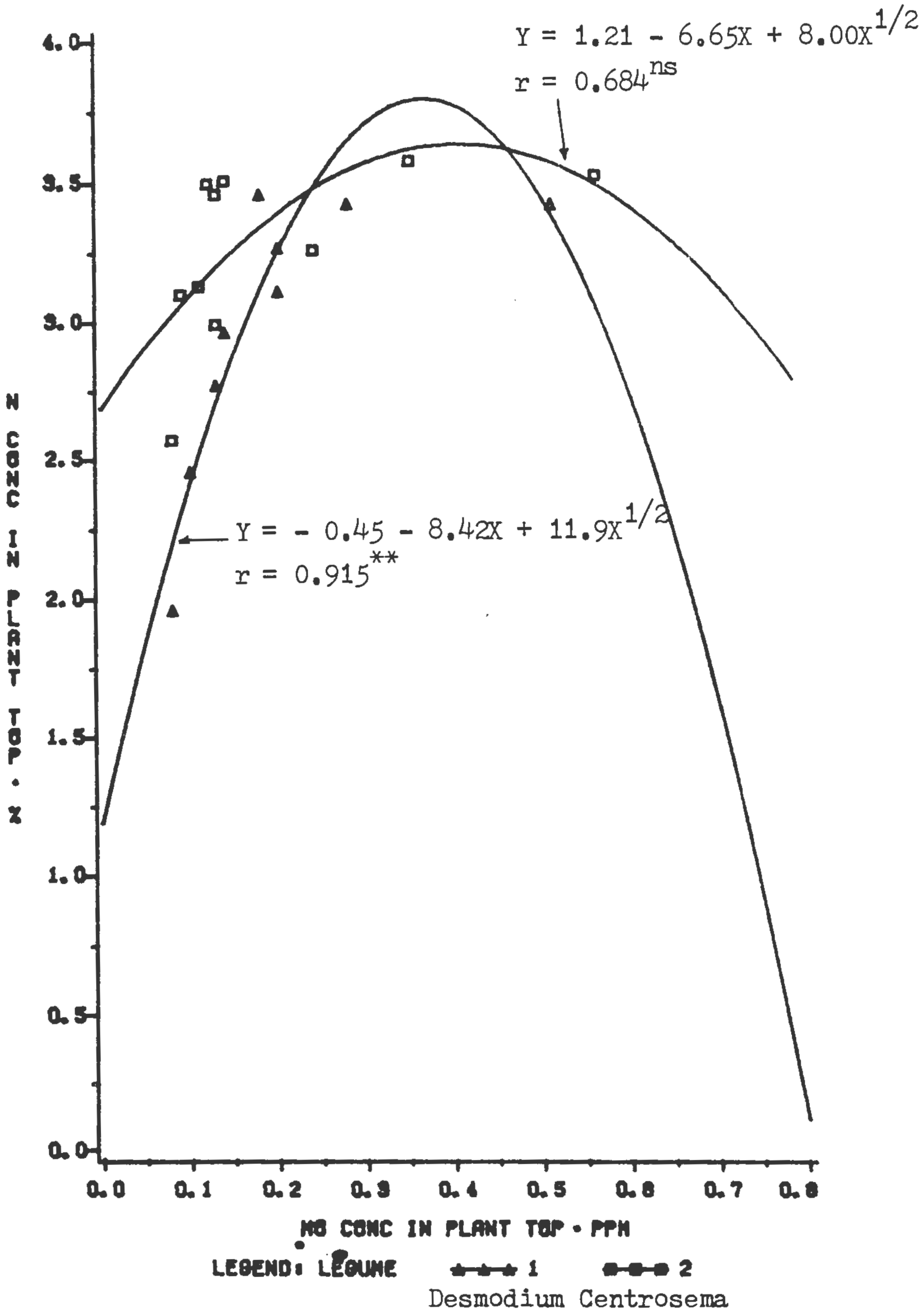


Fig. 1.57. Relationship between N and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in the Wahiawa soil (Second cutting).

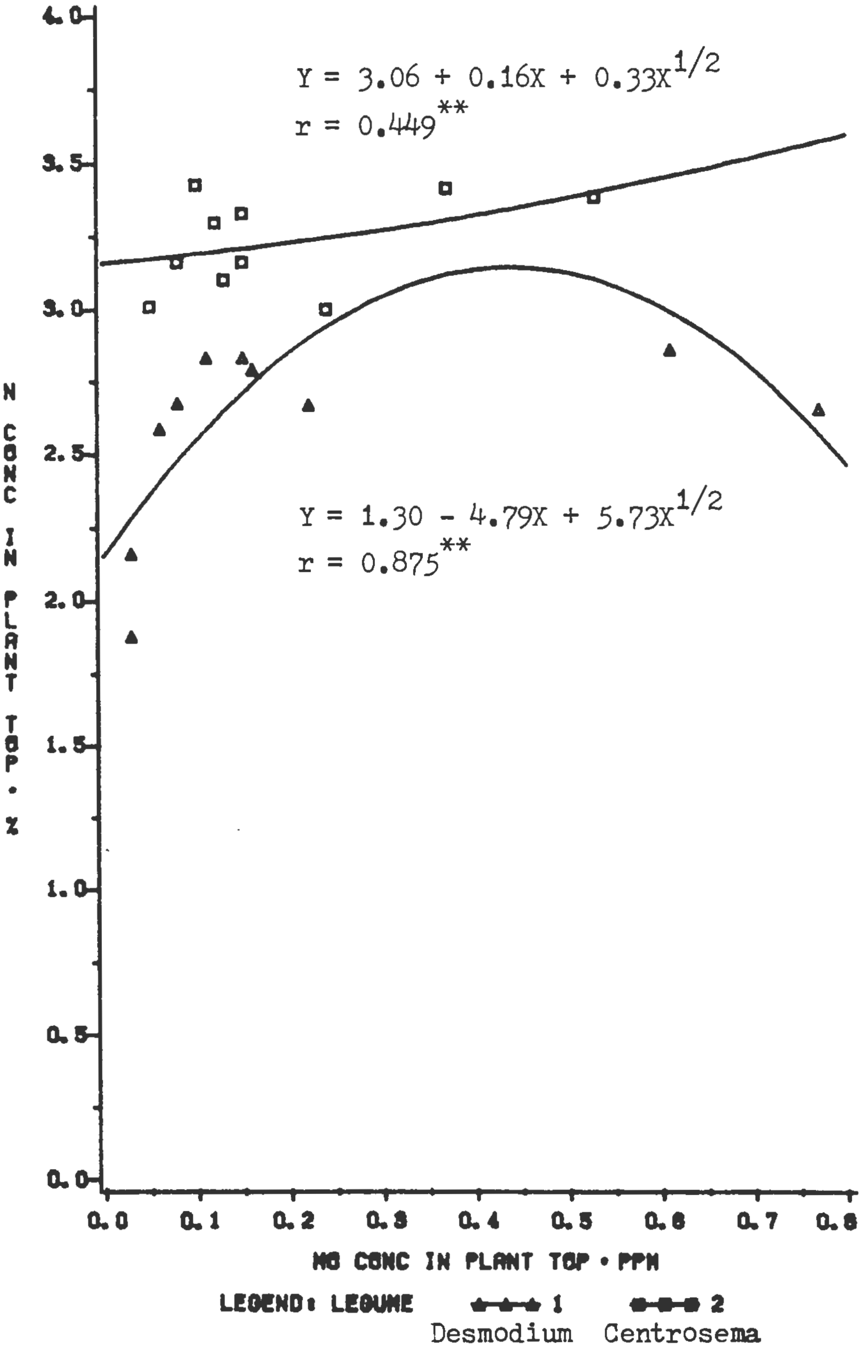


Fig. 1.58. Relationship between N and Mo concentrations in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Third cutting).

decreased as Mo concentration increased above 0.4 ppm. There was no significant correlation between N and Mo concentrations in the top of centrosema. The correlation coefficient of the relation was 0.449 (p = 0.45).

Fourth Cutting:

There was a highly significant correlation between N and Mo concentrations in the top of desmodium with the correlation coefficient of 0.887. According to the scatter diagram (Fig. 1.59), N concentration increased as Mo concentration increased. However, rate of increase declined when Mo concentration more than 0.4 ppm was observed and N concentration decreased when Mo concentration was more than 0.7 ppm. There was no **significant** correlation between N and Mo concentrations in the top of centrosema. The correlation coefficient of the relationship was 0.292 (0.73).

Fifth Cutting:

There was a highly significant correlation between N and Mo concentration in the top of desmodium with the correlation coefficient of 0.891. The scatter diagram (Fig. 1.60) indicated that N concentration increased as the Mo concentration increased within the Mo range of 0 to 0.3 ppm. Nitrogen concentration decrease when Mo concentration increased above 0.3 ppm. There was no significant correlation between N and Mo concentration in the top of centrosema. The correlation coefficient of the relationship was 0.245 (p = 0.80).

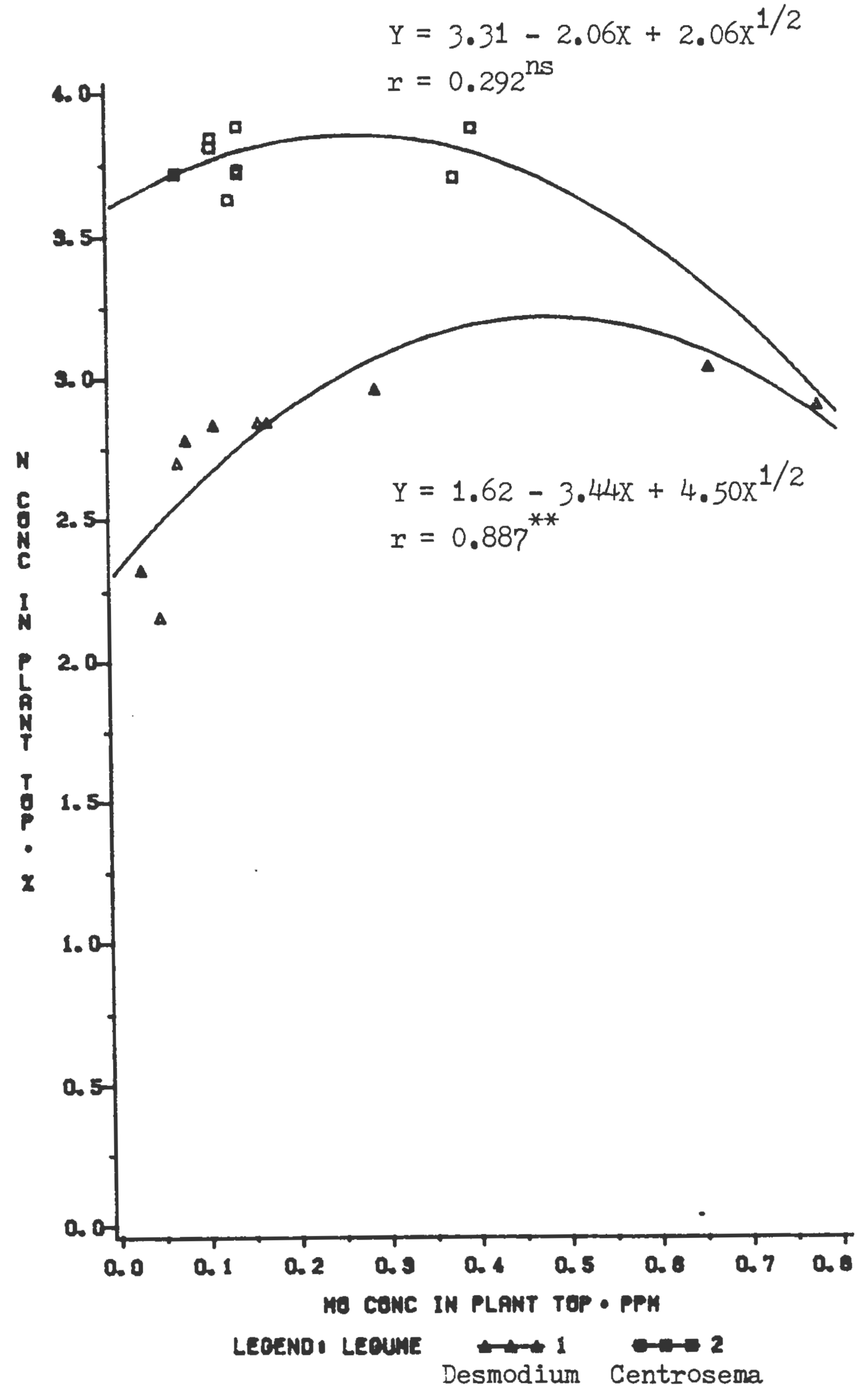


Fig. 1.59. Relationship between N and Mo concentrations in the tops of *Desmodium intortum* and *Centrosema pubescens* grown in the Wahiawa soil (Fourth cutting).

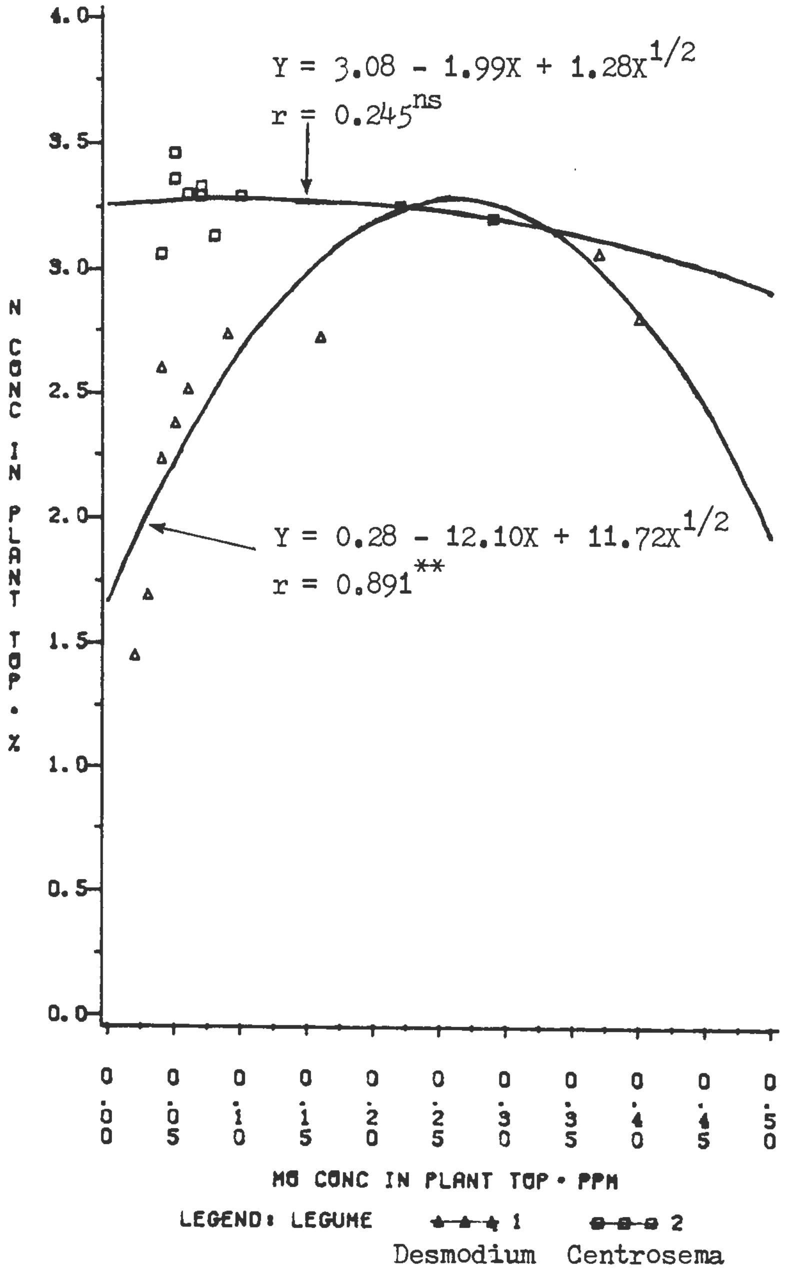


Fig. 1.60. Relationship between N and Mo concentrations in the tops of Desmodium intortum and Centrosema pubescens grown in Wahiawa soil (Fifth cutting).

SUMMARY

Molybdenum fertilizer increased dry matter yield of *Desmodium intortum* and *Centrosema pubescens* grown in Wahiawa soil. The results obtained from the greenhouse experiment indicated that soil-applied Mo at 2.0 and 4.0 kg Mo/ha were adequate for these legumes grown for one cutting in the greenhouse. Seed-applied Mo at 0.5 kg Mo/ha also increased yield of desmodium and centrosema. The efficiency of seed-applied Mo at 0.5 kg Mo/ha was similar to that of soil-applied Mo at 2.0 and 4.0 kg Mo/ha. Seed-applied Mo at 0.1 kg Mo/ha did not significantly increase legume yield, indicating that seed-applied Mo at this rate was not adequate for the requirement of legumes grown in the Wahiawa soil

Although Mo did not significantly increase dry matter yield of desmodium grown in the Paaloa soil in the greenhouse because of the Al toxicity, it still affected desmodium yield. When S and lime were applied, seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 4.0 kg Mo/ha increased yield by 98.3% and 106%, respectively, above the no-Mo treatment. Seed-applied Mo at 0.5 kg Mo/ha was as effective as soil-applied Mo at 4.0 kg Mo/ha. Seed-applied Mo at 0.1 kg Mo/ha was not adequate for the requirement of desmodium grown in the Paaloa soil.

The results obtained from the field experiment conducted with the Wahiawa soil confirmed that seed-applied Mo at 0.5 kg Mo/ha was as effective as soil-applied Mo at 2.0 and 4.0 kg Mo/ha. The field experiment also demonstrated that seed-applied Mo at 0.5 kg Mo/ha, as well as soil-applied Mo at 2.0 and 4.0 kg Mo/ha, were adequate for the requirements of desmodium and centrosema for at least five cuttings (from February 1983 until April 1984). At the first cutting, soil-applied at 1.0 and 2.0 kg Mo/ha did not significantly increase yield, suggesting that for a excellent stands of legumes, soil-applied Mo at 4.0 kg Mo/ha should be applied. The alternative method for excellent stands was to use seed-applied Mo at 0.3 or 0.5 kg Mo/ha. The field experiment also confirmed that seed-applied Mo at 0.1 kg Mo/ha was not adequate for legume need. The inadequacy of Mo obtained from seed-applied Mo at 0.1 kg Mo/ha rate became more evident at the later cuttings (cuttings 4 and 5); plants showed N deficiency symptoms.

Sulfur application at 50 kg S/ha did not increase dry matter yield of desmodium and centrosema grown in the Wahiawa soil. However, S significantly increased yield of desmodium grown in the Paaloa soil. Yield of desmodium receiving S increased by 40.3% above that of legume receiving no S.

Application of lime at a rate such that soil pH was raised to 6.0 increased dry matter yield of desmodium and centrosema grown in the Wahiawa soil. The influence of lime was obvious when Mo and S fertilizers were not applied. The influence of lime was even more evident for desmodium grown in Paaloa soil which has a low pH and high Al. When averaged over all Mo and S rates, yield of desmodium grown in limed soil was increased two-fold above that of legume grown in unlimed soil. When Mo and S were not added, yield of desmodium grown in limed soil was increased six-fold above that of legume grown in unlimed soil. These results indicated the beneficial effect of lime in alleviating Al toxicity in soil.

The influence of lime was also observed in the field experiment with the Wahiawa soil. When no Mo was applied, lime increased yields of desmodium and centrosema. However, when lime was applied along with 0.5 kg Mo/ha seed-applied or 4.0 kg Mo/ha soil-applied, yield response due to lime was not evident. Yield obtained from the 0.5 kg Mo/ha seed-applied treatment with lime was not significantly different from that obtained from the same seed-applied Mo treatment without lime. Yield obtained from the 4.0 kg Mo/ha soil-applied treatment, with or without lime, was also not significant different from each other. The influence of lime in increasing legume yield was similar from the first cutting to the fifth cutting, indicating the residual effect of lime.

Molybdenum fertilizer did not significantly affect nodule fresh weight and dry weight of desmodium grown in the Wahiawa soil. Molybdenum also did not significantly affect nodule number and nodule fresh weight of centrosema grown in the Wahiawa soil. However, Mo significantly increased nodule dry weight. Nodule dry weight of centrosema receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha was 83.3% and 106% higher than that of legume receiving no Mo.

Although Mo did not significantly affect nodule number of desmodium grown in the Paaloa soil, there was a trend indicating that seed-applied Mo had a detrimantal effect on Rhizobium bacteria. Nodule number of legume receiving seed-applied Mo was less than that of legume receiving no Mo or soil-applied Mo. Seed-applied Mo at 0.5 kg Mo/ha decreased nodule fresh weight and dry weight of desmodium.

Lime did not significantly increase nodule fresh and dry weights of desmodium grown in the Wahiawa soil. Lime significantly increased nodule number, nodule fresh and dry weights of centrosema grown in the Wahiawa soil. Lime markedly increased nodule number, nodule fresh and dry weights of desmodium grown in the Paaloa soil. Desmodium grown in unlimed Paaloa soil did not nodulate, whereas that grown in limed soil nodulated very well, suggesting that lime was one of the most critical factor for nodulation of desmodium grown in the Paaloa soil.

Sulfur fertilizer did not significantly affect nodule fresh and dry weights of desmodium grown in the Wahiawa soil. Sulfur did not significantly increase nodule number, nodule fresh and dry weight of desmodium grown in the Paaloa soil. It should be noted, however, that while S had no effect on nodule number of legume grown in limed soil which received soil-applied Mo, it tended to increase nodule number of legume grown in limed soil which received seed-applied Mo. Sulfur also significantly increased nodule fresh and dry weights of desmodium grown in limed soil which received seed-applied Mo. Sulfur did not significantly affect nodule number, nodule fresh and dry weight of centrosema grown in the Wahiawa soil.

Molybdenum, S and lime did not significantly increase nitrogenase activity of desmodium grown in the Wahiawa soil. However, nitrogenase activity tended to be higher when Mo was applied, either by seed coating or by soil application. Seed-applied Mo at 0.1 and 0.5 kg Mo/ha increased nitrogenase activity by 21.3% and 29.1%, respectively, above the no-Mo treatment. Soil-applied Mo at 2.0 and 4.0 kg Mo/ha also increased nitrogenase activity by 37.0% and 31.8%, respectively, above the no-Mo treatment. Molybdenum also tended to increase specific nitrogenase activity. Seed-applied Mo at 0.1 and 0.5 kg Mo/ha increased specific nitrogenase activity by 15.7% and 24.1%, respectively, above the no-Mo treatment. Soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased specific nitrogenase activity by 26.4% and 20.1%, respectively, above the no-Mo treatment.

Molybdenum fertilizer significantly increased nitrogenase activity of centrosema grown in the Wahiawa soil. Soil-applied Mo at 4.0 kg Mo/ha significantly increased nitrogenase activity by four-fold. Soil-applied Mo at 2.0 kg Mo/ha and seed-applied Mo at 0.5 kg Mo/ha increased nitrogenase activity by 199% and 166%, respectively, above the no-Mo treatment. Sulfur had no significant effect on the nitrogenase activity of centrosema. However, lime increased nitrogenase activity by 103% above the no-lime treatment.

Molybdenum fertilizer significantly increased N concentration in the top of desmodium grown in the Wahiawa soil. Soil-applied Mo at 2.0 and 4.0 kg Mo/ha increased N concentration by 27.9% and 42.9%, respectively, above the no-Mo treatment. Seed-applied Mo at 0.5 kg Mo/ha increased N concentration by 29.4% above the no-Mo treatment. Seed-applied Mo at 0.1 kg Mo/ha did not increase N concentration in plant top, indicating that seed-applied Mo at 0.1 kg Mo/ha did not supply an adequate amount of Mo for nitrogen fixation of desmodium. Molybdenum tended to increase N concentration in the top of centrosema grown in the Wahiawa soil.

Molybdenum did not significantly increase N concentration in the top of desmodium grown in the Paaloa soil because the growth of plant grown in unlimed soil was limited by Al toxicity which also affected N concentration in plant top. However, N concentration increased as Mo concentration in nodule increased.

The results from the field experiment confirmed that Mo fertilizer increased N concentration in plant top. Soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha increased N concentration in the top of desmodium and centrosema from the first to fifth cuttings. Seed-applied Mo at 0.3 and 0.5 kg Mo/ha increased N concentration in the top of desmodium and centrosema from the first to fourth cuttings. However, seed-applied Mo at 0.1 kg Mo/ha did not significantly increase N concentration of desmodium and centrosema. Nitrogen deficiency symptoms occurred in desmodium receiving no Mo from the first to the fifth cuttings. Nitrogen deficiency symptoms also occurred in plant receiving seed-applied Mo at 0.1 kg Mo/ha at the fourth and fifth cuttings. These results confirmed the results from the greenhouse experiments that seed-applied Mo at 0.1 kg Mo/ha did not adequately supply Mo for plant need.

At the fifth cutting, N concentration of desmodium receiving seed-applied Mo at 0.3 kg Mo/ha was significantly lower than those of legume receiving seed-applied Mo at 0.5 kg Mo/ha and soil-applied Mo at 1.0, 2.0 and 4.0 kg Mo/ha, suggesting that seed-applied Mo at 0.3 kg Mo/ha did not supply an adequate amount of Mo after the fourth cutting. Although N concentration of desmodium receiving seed-applied Mo at 0.5 kg Mo/ha was not significantly different from those of desmodium receiving soil-applied Mo at 1.0 and 2.0 kg Mo/ha, it was lower than that of legume receiving soil-applied Mo at 4.0 kg Mo/ha. The residual effect of seed-applied Mo at 0.5 kg Mo/ha appeared to be less than that of soil-applied Mo at 4.0 kg Mo/ha.

Sulfur did not significantly increase N concentration in plant top of desmodium grown in Wahiawa and Paaloa soils. Sulfur application decreased N concentration in the top of centrosema grown in the Wahiawa soil. However, it should be noted that N concentration of centrosema was quite low, indicating that factors other than Mo limited nitrogen fixation.

Lime had no significant effect on N concentration in the top of desmodium and centrosema grown in the Wahiawa soil. However, lime significantly increased N concentration in the top of desmodium grown in the Paaloa soil. Lime increased N concentration by 40.3% above the unlimed treatment. The effect of lime was contributed to its enhancement of nodulation. Lime also increased N concentration in the top of desmodium and centrosema grown in the Wahiawa soil in the field. However, the effect of lime was not evident when lime was applied along with Mo. Nitrogen concentration of legumes grown in limed soil which received seed-applied Mo at 0.5 kg Mo/ha or soil-applied Mo at 4.0 kg Mo/ha was not significantly higher than that of legumes grown in unlimed soil which received the same rates of Mo.

Molybdenum fertilizer increased Mo concentration in the top of desmodium and centrosema grown in the Wahiawa soil. Molybdenum concentrations in the top of legumes receiving soil-applied Mo were higher than those of legumes receiving seed-applied Mo. Molybdenum concentrations in the top of desmodium receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were 0.47 and 1.07 ppm, respectively. Molybdenum concentrations in the top of desmodium receiving seed-applied Mo at 0.1 and 0.5 kg Mo/ha were 0.10 and 0.25 ppm, respectively. Molybdenum concentrations of centrosema receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were 0.71 and 1.14 ppm, respectively. Molybdenum concentrations of centrosema receiving seed-applied Mo at 0.1 and 0.5 kg Mo/ha were 0.29 and 0.38 ppm, respectively.

Molybdenum fertilizer also significantly increased Mo concentration in the top of desmodium grown in the Paaloa soil. Molybdenum concentrations in the top of legume receiving soil-applied Mo at 2.0 and 4.0 kg Mo/ha were 0.73 and 1.44 ppm, respectively. Molybdenum concentrations in the top of legume receiving seed-applied Mo at 0.1 and 0.5 kg Mo/ha were 0.17 and 0.43 ppm, respectively.

Molybdenum fertilizer significantly increased Mo concentration in the top of desmodium and centrosema grown in the field with the Wahiawa soil. At the first cutting, Mo concentrations in the top of desmodium ranged from 0.11 to 0.52 ppm whereas those in the top of centrosema ranged from 0.11 to 0.60. Plant Mo concentrations in the later cuttings decreased when compared to those in the earlier cuttings. At the fifth cutting, Mo concentrations of desmodium ranged from 0.02 to 0.40 ppm whereas those of centrosema ranged from 0.04 to 0.29 ppm.

Sulfur fertilizer did not significantly affect Mo concentration in the tops of desmodium and centrosema grown in the the Wahiawa soil. Sulfur also did not significantly affect Mo concentration in the top of desmodium grown in the Paaloa soil.

Lime did not significantly increase Mo concentration in the top of desmodium grown in the Wahiawa soil in a greenhouse experiment. However, lime increased Mo concentration in the top of desmodium grown in the Paaloa soil. Lime also increased Mo concentration in the top of centrosema grown in the Wahiawa soil. Lime did not significantly increase Mo concentration in the top of desmodium and centrosema grown in the field.

Dry matter yield of desmodium and centrosema grown in the Wahiawa soil increased as the Mo concentration in nodule increased. Critical Mo concentrations in nodules of desmodium and centrosema associated with 80% maximum yield were 3.5 and 4.2 ppm, respectively.

Dry matter yield of desmodium grown in the greenhouse and in the field increased as Mo concentration in plant top increased. In the greenhouse experiment, critical Mo concentration in plant top associated with 80% maximum yield was 0.20 ppm. From the third cutting of the field experiment, critical Mo concentration in plant top associated with 80%

and 95% maximum yield were 0.13 and 0.30 ppm, respectively. Dry matter yield of centrosema grown in the Wahiawa soil did not correlate well with Mo concentration in plant top. However, it can be estimated that Mo concentration in plant top must be at least 0.20 ppm to be associated with 80% maximum yield.

Molybdenum fertilizer had no effect on S concentration in the top of desmodium and centrosema grown in the Wahiawa soil. Molybdenum fertilizer also had no significant effect on S concentration in the top of desmodium grown in the Paaloa soil. Sulfur application at 50 kg S/ha did not significantly increase S concentration in the top of desmodium and centrosema grown in Wahiawa soil. Sulfur fertilizer also did not significantly increase S concentration in the top of desmodium grown in Paaloa soil.

Lime had little effect on S concentration in the top of desmodium grown in the Wahiawa soil. However, lime significantly increased S concentration in the top of centrosema grown in the Wahiawa soil. Lime significantly decreased S concentration in the top of desmodium grown in the Paaloa soil. The reason that lime decreased S concentration was likely due to a "dilution effect" since lime markedly increased plant growth.

Molybdenum fertilizer tended to increase Cu concentration in the top of desmodium grown in the Wahiawa soil. However, Mo fertilizer did not significantly increase Cu concentration in the top of centrosema grown in the Wahiawa soil. Molybdenum also did not increase Cu concentration in the top of desmodium grown in the Paaloa soil. Sulfur had no significant effect on Cu concentration in the top of desmodium and centrosema grown in Wahiawa soil. Sulfur also had no significant effect on Cu concentration in the top of desmodium grown in the Paaloa soil. Lime increased Cu concentration in the top of desmodium and centrosema grown in the Wahiawa soil. However, lime decreased Cu concentration in the top of desmodium grown in the Paaloa soil.

The potential harmful effect of Mo regardind Mo-induced Cu deficiency in animals should not occur because Cu/Mo ratio was well above 3, which was the ratio under which Mo-induced Cu deficiency usually occurs. The Cu/Mo ratios in the top of desmodium grown in the Wahiawa soil in the greenhouse ranged from 11 to 153. In the field experiment conducted with the Wahiawa soil, Cu/Mo ratios in the top of desmodium ranged from 8 to 925 whereas those in the top of centrosema ranged from 10 to 369.

Molybdenum fertilizer decreased the concentrations of P, Ca, Mg, Si, Mn and Al in the top of desmodium and centrosema. The decrease in these nutrients was likely due to a "dilution effect" of growth. Molybdenum had no significant effect on K, Fe and Zn concentrations in plant top. Sulfur fertilizer had no significant effect on nutrient concentrations in plant top. Lime increased Ca concentration in plant top but lime had no effect on Fe concentration. Lime decreased P, Mg, Mn and Si concentrations and also tended to decrease K concentration in the tops of desmodium and centrosema.

CHAPTER II

EXTERNAL MOLYBDENUM REQUIREMENT OF *DESMODIUM INTORTUM* GROWN IN AN OXISOL AND ULTISOL

INTRODUCTION

The nutrient requirement of a crop can be expressed as the internal nutrient requirement and external nutrient requirement. The internal nutrient requirement can be defined as the concentration of a nutrient in the plant that is associated with near maximum yield, usually named the "critical concentration". The external nutrient requirement may be defined as the quantity of nutrient (or some proportional part of that quantity) that constitutes a minimum pool for adequate crop nutrition (Fox, 1981). The "external requirement" can also refer to the intensity of nutrition: the concentration of nutrient in the soil solution which is associated with adequate nutrition. The quantity of nutrient frequently takes on greater significance if it is considered in relation to the capacity of the soil to hold nutrient (Fox, 1981). This is the case of an Oxisol or an Ultisol which will highly adsorb Mo.

Various extraction procedures have been used to characterize soil Mo and attempts have been made to correlate these procedures with plant response. Among the popular procedures, acid ammonium oxalate extraction has been frequently used. Molybdenum extracted by acid ammonium oxalate has been found to correlate with field response where less than 0.15 ppm Mo was extracted (Grigg, 1960). Gupta and Mackay (1966) also used acid ammonium oxalate to extract Mo from podzolic soils. They obtained Mo values ranging from 0.02-0.23 ppm Mo. Barrow and Spencer (1971) found that acid ammonium oxalate-extractable Mo accounted for a large part of the regression between yield and estimates of Mo availability to plant. Sultana-

Ahmed and Rahman (1982) indicated that acid ammonium oxalate-extractable Mo was positively correlated with plant Mo concentration.

Barrow (1970) used adsorption isotherms to rank soils according to the amount of Mo adsorbed to maintain a solution concentration of 0.1 ppm Mo. Little and Kerridge (1978) related soil solution Mo, Mo adsorption isotherms and acid ammonium oxalate-extractable Mo to the response of tropical legumes to Mo fertilizer in the field with different soil types. They concluded that equilibration of soil samples with solutions of Mo and measuring the Mo adsorbed provided adsorption isotherms that indicated the best promise of predicting Mo response.

This part of the study was conducted in the greenhouse to: (1) relate Mo adsorption and desorption to the response of *Desmodium intortum* to Mo, and (2) determine the external Mo requirement of this legume grown in Wahiawa and Paaloa soils.

MATERIALS AND METHODS

1. Procedures

Pot experiments were conducted at the Agronomy and Soil Science Field Laboratory, Mauka Campus of the University of Hawaii at Manoa. *Desmodium intortum* was grown in the Wahiawa and Paaloa soils. Some chemical properties of both soils are shown in Table 2.1 for the Wahiawa soil and Table 1.1 for the Paaloa soil. Molybdenum adsorption curves of both soils were determined before applying treatments.

The experimental design used was a randomized complete block with seven treatments. Each treatment was replicated three times. The seven levels of Mo fertilizer were: 0, 0.5, 1.0, 2.0, 4.0, 8.0 and 16.0 kg Mo/ha. Legume was inoculated with the appropriate *Rhizobium* strain. Phosphorus, K, Mg, S, Zn, Cu and B were added to all treatments as part of a basal fertilizer application.

Thirty inoculated seeds of desmodium were planted in a 6-l pot. Three weeks after planting the seedling were thinned to three uniformly growing plants per pot. Through the experiment the pots were watered daily to 100% field capacity. The plants were harvested at 60 days after planting. They were cut approximately 1 cm above the soil surface and the roots were carefully removed from the soil with a minimum detachment of nodules. The roots with nodules were put into 500-ml glass bottles for acetylene reduction assessment. Plant samples were rinsed with deionized water and oven-dried at 70° C in an air-draft oven for three days for determining dry matter yield. The oven-dried samples were ground (20 mesh)

Table 2.1.

Some chemical properties of Poamoha soil

pH (1:1, soil:water)	4.7
Available P (modified Truog) (ug/g)	128
Water soluble P (ug/ml)	0.05
Exchangeable bases (NH_4OAc extraction) (meq/100 g)	
K	0.69
Ca	2.23
Mg	1.19
Na	0.21
Exchangeable Mn (KCl extraction) (ppm)	28.5
Extractable Mo (acid ammonium oxalate extraction) (ppm)	0.22
Available S ($Ca(H_2PO_4)_2$ extraction) (ppm)	53.0

in a stainless steel Wiley Mill and kept for elemental analysis.

2. Treatment Preparation

2.1. Molybdenum Adsorption Curves

Adsorption studies were conducted at room temperature (about 23° C). Twenty five grams of soil was weighed into each of 11 500-ml glass bottles to which were added 250 ml of 0.001 M $CaCl_2$ containing Mo at 0, 0.25, 0.5, 1.0, 2.0, 4.0, 8.0, 16.0, 32.0, 64.0 and 128.0 ppm Mo as Na_2MoO_4. The reciprocating bottles were shaken by a mechanical shaker for 24 hours. The mixture was filtered and the filtrate was analyzed for Mo. Molybdenum which was not detected in the filtrate was considered to have been absorbed. Molybdenum adsorbed was plotted against Mo concentration in the supernatant solution.

2.2. Molybdenum Desorption Curves

The Mo-equilibrated soils obtained from adsorption studies were used to establish desorption curves. The air-dried soil receiving each Mo level was extracted with acid ammonium oxalate solution to determine available Mo in soil. Acid ammonium oxalate-extractable Mo was considered to be the available Mo in soil since it consisted of Mo in soil solution and semi-available Mo (Mo adsorbed by Fe and Al oxides). The available Mo was plotted against Mo adsorbed by soils.

2.3. Fertilizers and Lime Used

Fertilizers and lime used in the experiments were as follow:

2.3.1. Basal Fertilizers

- Calcium phosphate, monobasic ($Ca(H_2PO_4)_2$) at 600 kg P/ha.

- Potassium chloride (KCl) at 200 kg K/ha.
- Magnesium sulfate ($MgSO_4.7H_2O$) at 200 kg Mg/ha.
- Zinc sulfate ($ZnSO_4.7H_2O$) at 20 kg Zn/ha.
- Copper sulfate ($CuSO_4.5H_2O$) at 3 kg Cu/ha.
- Boric acid (H_3BO_3) at 2 kg B/ha.

2.3.2. Molybdenum Fertilizer

- Sodium molybdate ($Na_2MoO_4.2\ H_2O$).

2.3.3. Lime

- Calcium hydroxide ($Ca(OH)_2$).

Basal fertilizers, except the phosphate fertilizer, were used in solution form. Each chemical was weighed at the required amount for the whole treatment, mixed together in solution form and made up to a volume of 630 ml. The solution fertilizer was used at 30 ml per pot. Phosphate fertilizer was weighed separately at the required amount for each pot and mixed with the soil along with the solution fertilizer.

2.4. Soil Preparation

The soil was sieved through a 1-cm sized sieve to preclude gravels, stones and large roots and spread on benches to dry. After one week of air-drying, the soil was ground and thoroughly mixed to attain homogeneity, a soil sample was taken to determine moisture content as well as to conduct soil analysis for determining some chemical properties of soil. Four kilograms of oven-dried soil was put into a 6-l plastic pot for subsequent liming and fertilizer application. For the Paaloa soil, $Ca(OH)_2$ at the amount needed to raise soil pH to 5.2 according to the lime requirement curve was mixed with soil and the limed soil was incubated for 2 weeks before fertilizer application and seeding.

3. Measurements of Responses

3.1. Yield Response

Relationship between dry matter yield, relative yield and available Mo obtained from desorption study were evaluated. The external Mo requirement of desmodium was also determined.

3.2. Nodulation Response

Relationships between number of nodule, nodule weight and available Mo were evaluated.

3.3. Nitrogenase Activity

Relationship between nitrogenase activity and available Mo was evaluated. The available Mo associated with optimum nitrogenase activity was also determined.

3.4. Nitrogen Concentration

Relationship between N concentration and available Mo was evaluated. Available Mo associated with optimum N concentration was determined.

3.5. Molybdenum Concentrations

Relationship between Mo concentration in plant top, Mo concentration in nodule,extractable Mo in soil after harvesting and available Mo were evaluated. Available Mo associated with critical Mo concentrations in the top and nodule was determined.

RESULTS AND DISCUSSION

Molybdenum Adsorption Characteristics

The Wahiawa and Paaloa soils used for the experiments varied in their Mo adsorption capacities as is evident from the adsorption curves presented in Fig. 2.1. In this study on external Mo requirements the concentration of 0.002 ug Mo/ml in solution was selected to be the standard concentration for comparing adsorption capacities of these soils. Barrow (1970) and Little and Kerridge (1978) who worked with Australian soils used a Mo concentration of 0.1 and 0.01 ug Mo/ml, respectively, as the standard Mo concentration in solution. However, Oxisols and Ultisols in Hawaii highly adsorb Mo so that the amount of Mo remaining in solution after equilibrating soil with 0.001 M $CaCl_2$ containing Na_2MoO_4 was lower than 0.01 ug Mo/ml at the Mo adsorption range of 0 to 128 ug Mo/g (Fig. 2.1). Moreover, the Mo concentration that the Australian workers used to equilibrate soil was higher than the Mo concentration used in this study.

The same pattern of the adsorption curves (Fig. 2.1) of Wahiawa and Paaloa soils reflected similar parent materials. Both soils have an olivine-basaltic origin. However, they were different in their adsorption capacities. The Wahiawa soil adsorbed 18.3 ug Mo/g at 0.002 ug Mo/ml in solution whereas the Paaloa soil adsorbed 28.8 ug Mo/g at the same concentration of Mo in solution. The difference in adsorption capacities of both soils was due to the differences in their clay mineralogy. The Paaloa soil contains less kaolinite but more gibbsite, goethite and haematite than Wahiawa soil. The fact that the Paaloa soil contains more Fe and Al oxides

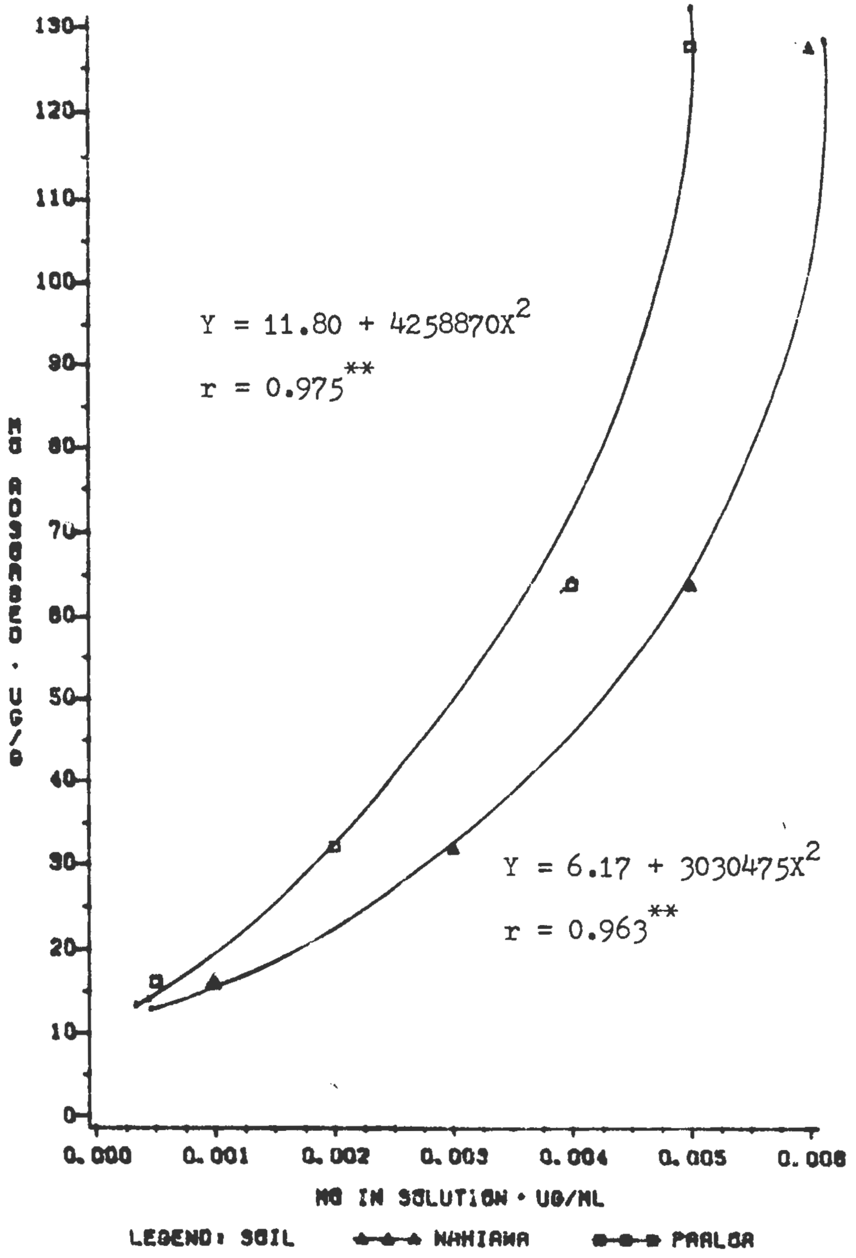


Fig. 2.1. Molybdenum adsorption curves for Wahiawa and Paaloa soils.

than the Wahiawa soil is the reason that the Paaloa soil adsorbs more Mo than the Wahiawa soil.

It should be mentioned that when Na_2MoO_4 at rates of 0 to 8 ppm was equilibrated with both Wahiawa and Paaloa soils, no Mo was detected in the solution. All of the Mo added appeared to be adsorbed by the soil. This adsorbed Mo cannot be desorbed by water extraction; therefore, acid ammonium oxalate solution was used to desorbed Mo that had been adsorbed by soil.

Molybdenum Desorption Characteristics

Molybdenum-equilibrated soils obtained from the previous Mo adsorption study were extracted by acid ammonium oxalate solution. Acid ammonium oxalate extractable Mo was considered to be available Mo. According to the desorption curve (Fig. 2.2), at the Mo adsorption range of 0 to 2.0 ug Mo/g, the available Mo released from equilibrated Wahiawa and Paaloa soils were similar. At the 2.0 ug/g Mo adsorbed level, Wahiawa soil released 0.08 ug Mo/ml whereas the Paaloa soil released 0.09 ug Mo/ml. As the concentration of Mo adsorbed increased above 2.0 ug Mo/g, the available Mo of the Paaloa soil was higher than that of Wahiawa soil. This effect was due to the finding from the adsorption study that the Paaloa soil adsorbed more Mo than the Wahiawa soil at the same equilibrated Mo level. When both soils was extracted by acid ammonium oxalate solution, the Paaloa soil would release more Mo than the Wahiawa soil.

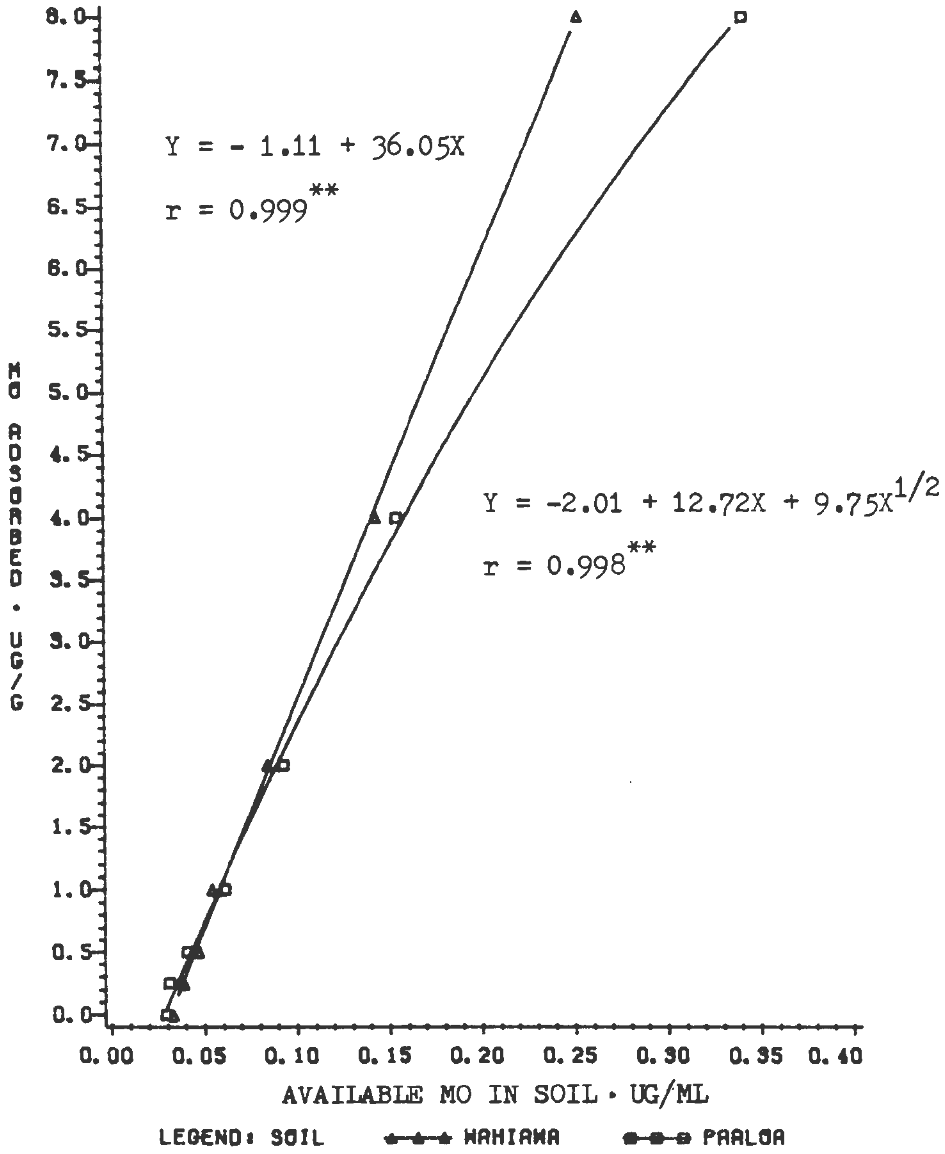


Fig. 2.2. Molybdenum desorption curves for Wahiawa and Paaloa soils.

Yield Response

Dry matter yields of desmodium grown in Wahiawa and Paaloa soils and receiving various rates of Mo (Table 2.2) were plotted against available Mo in soil as presented in Fig. 2.3. The yield response curves of desmodium grown in both soils were similar despite the overall yields of desmodium grown in the Wahiawa soil being lower than those of desmodium grown in the Paaloa soil. According to the response curves, yield of desmodium tended to increase as the available Mo increased up to approximately 0.2 ug/ml. When available Mo increased beyond this level, yield tended to decline, suggesting that high levels of Mo would have a toxic effect to plant growth.

Actual yields from each soil were converted to relative yield (percentage of the maximum yield) and plotted against corresponding available Mo as shown in Fig. 2.4. The available Mo associated with 95% maximum yield for Wahiawa and Paaloa soils were 0.09 and 0.10 ug Mo/ml, respectively. This results suggested that the external Mo requirement for soils high in kaolinite and oxides of Fe and Al like Wahiawa and Paaloa soils, could be estimated by available Mo since both soils that had different adsorption capacities had similar available Mo required to obtain 95% maximum yield. Predicted quantity of applied Mo required to establish 0.09 ug/ml available Mo for the Wahiawa soil was 4.4 kg Mo/ha (Fig. 2.5). For the Paaloa soil, predicted quantity of applied Mo required to established 0.10 ug/ml available Mo was 4.7 kg Mo/ha.

Table 2.2.

Dry matter yield and relative yield of _Desmodium_ _intortum_ in relation to Mo applied and the available Mo in Wahiawa and Paaloa soils.

Mo applied	Available Mo	Dry matter yield	Relative yield
kg/ha	ug/ml	kg/pot	%
	Wahiawa soil		
0.0	0.033	11.6 b	73.0
0.5	0.039	14.4 ab	90.6
1.0	0.047	14.0 ab	88.1
2.0	0.054	14.3 ab	89.9
4.0	0.084	15.9 a	100.0
8.0	0.143	14.9 ab	93.7
16.0	0.253	14.7 ab	92.5
	Paaloa soil		
0.0	0.029	10.9 b	52.9
0.5	0.031	15.9 ab	77.2
1.0	0.041	17.7 a	85.9
2.0	0.061	20.6 a	100.0
4.0	0.093	20.2 a	98.1
8.0	0.154	19.1 a	92.7
16.0	0.342	15.0 ab	72.8

* Values followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

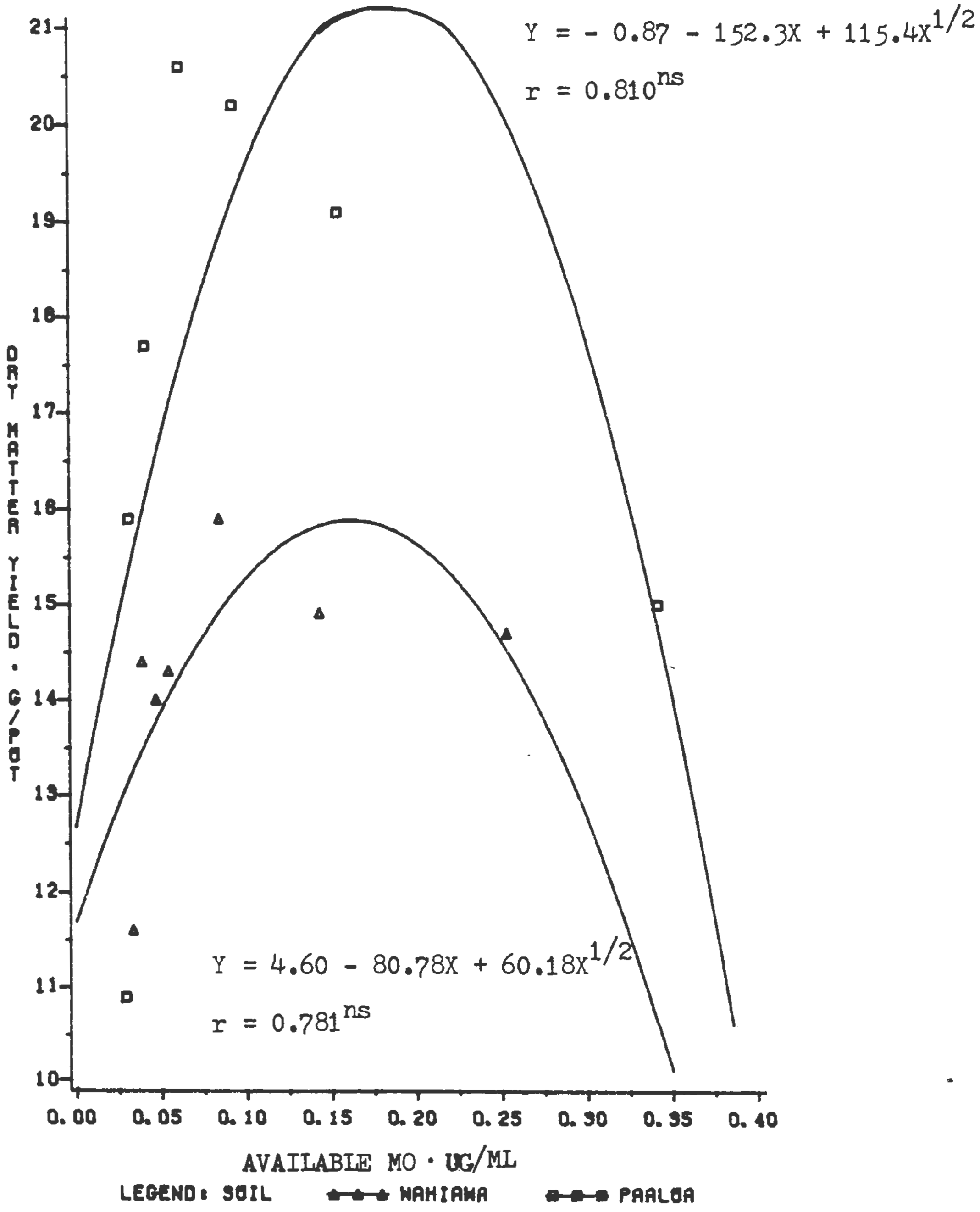


Fig. 2.3. Relationship between dry matter yield of Desmodium intortum and available Mo in Wahiawa and Paaloa soils.

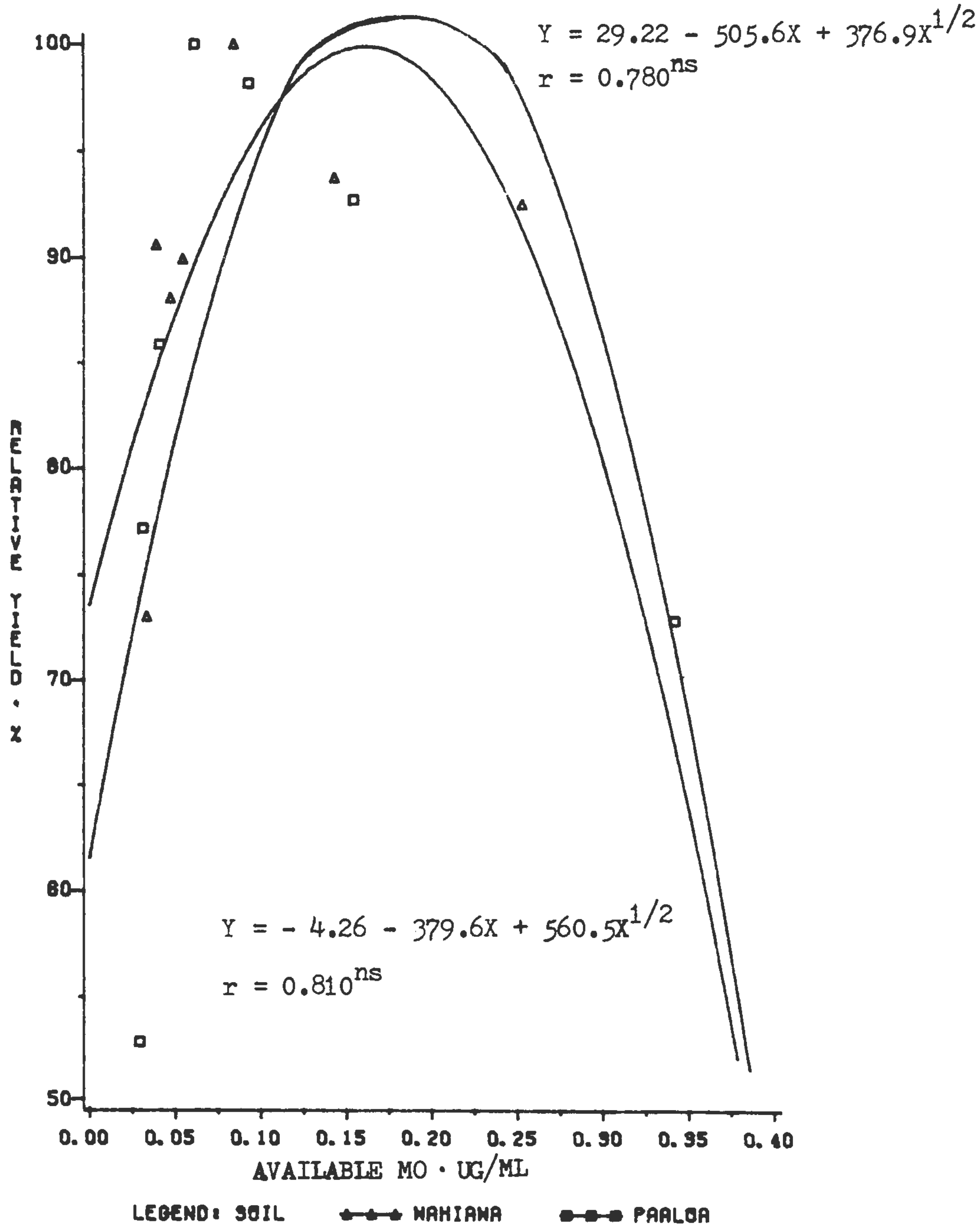


Fig. 2.4. Relationship between relative yield of Desmodium intortum and available Mo in Wahiawa and Paaloa soils.

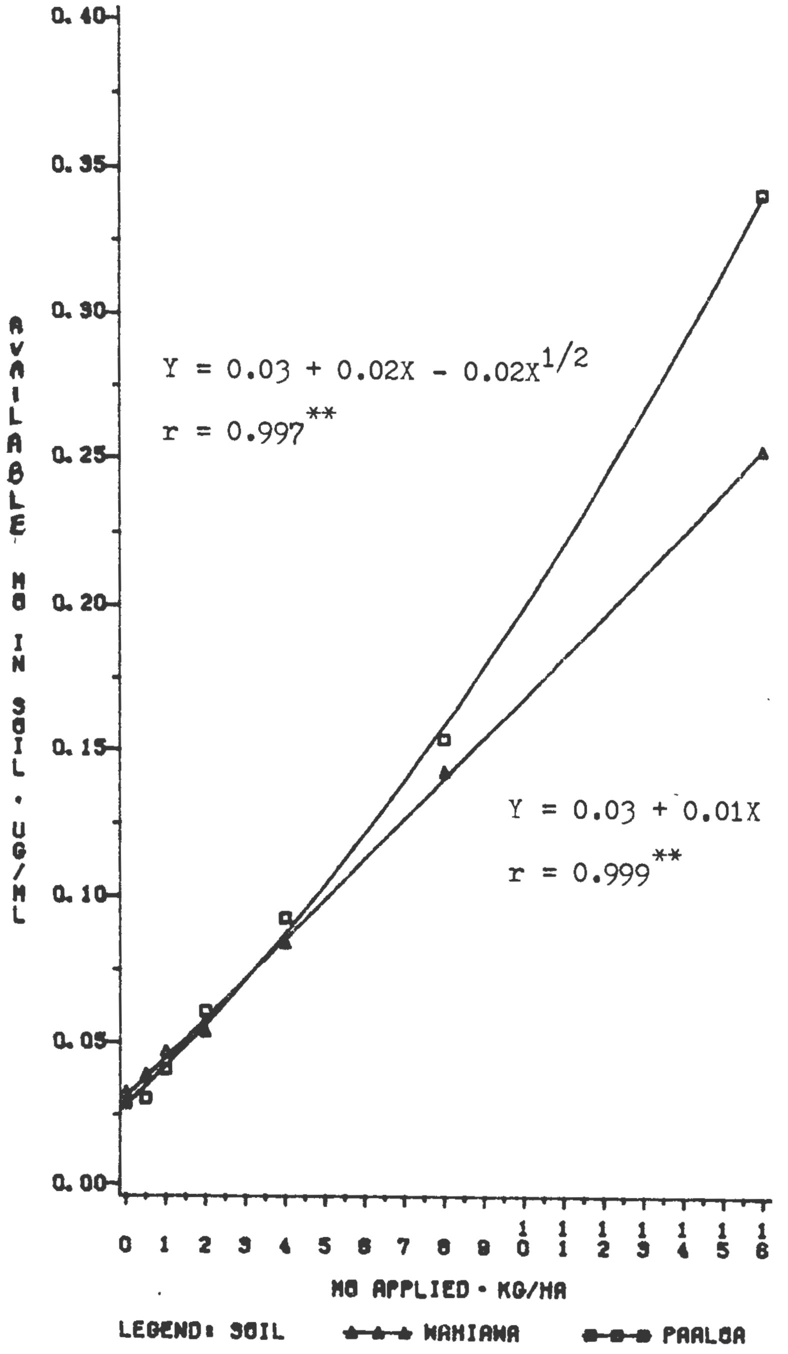


Fig. 2.5. Relationship between available Mo in Wahiawa and Paaloa soils, and the level of Mo applied.

Relationship between Mo Concentration in Plant Top and Available Mo in Soil

Molybdenum concentration in the top of desmodium grown in the Wahiawa soil increased from 0.03 ug/g at 0.033 ug/ml available Mo to 0.69 ug/g at 0.235 ug Mo/ml (Table 2.3). Molybdenum concentration in plant top associated with the available Mo at 0.09 ug/ml (external Mo requirement) was approximately 0.22 ug/g (Fig. 2.6).

Molybdenum concentration in the top of desmodium grown in the Paaloa soil increased from 0.10 ug/g at 0.029 ug/ml available Mo to 3.77 ug/g at 0.342 ug Mo/ml (Table 2.3). Molybdenum concentration associated with the available Mo at 0.10 ug/ml was approximately 1.29 ug/g (Fig. 2.6).

These results indicated that although the external Mo requirement of Wahiawa and Paaloa soils were quite similar (0.09 and 0.10 ug Mo/ml, respectively), the Mo concentration in plant tops of desmodium grown in both soils (0.2 and 1.3 ug/g, respectively) were different from each other. The reason that legume grown in Paaloa soil absorbed more Mo than Wahiawa soil was due to the fact that Paaloa soil was limed to pH 5.2 to eliminate Al toxicity. At this pH, the plant was likely to absorb more Mo than plant grown in Wahiawa soil which has a pH of 4.7. These findings suggest that it is necessary to consider the differences between some chemical properties of different soil types if plant Mo is to be used to evaluate the response of plant to Mo.

Table 2.3

Molybdenum concentration in the top and nodule of Desmodium intortum in relation to the level of available Mo in Wahiawa and Paaloa soils.

Available Mo	Mo concentration	
	Plant top	Nodule
ug/ml	ppm	
	Wahiawa soil	
0.033	0.03 c	1.65 c
0.039	0.04 c	2.80 c
0.047	0.07 c	3.09 c
0.054	0.09 c	4.34 c
0.083	0.15 c	8.58 b
0.143	0.40 b	10.82 b
0.253	0.69 a	16.51 a
	Paaloa soil	
0.029	0.10 d	3.27 e
0.031	0.14 d	4.37 e
0.041	0.27 d	6.41 de
0.061	0.44 d	9.24 cd
0.093	1.13 c	11.95 c
0.154	2.18 b	17.88 b
0.342	3.77 a	23.31 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

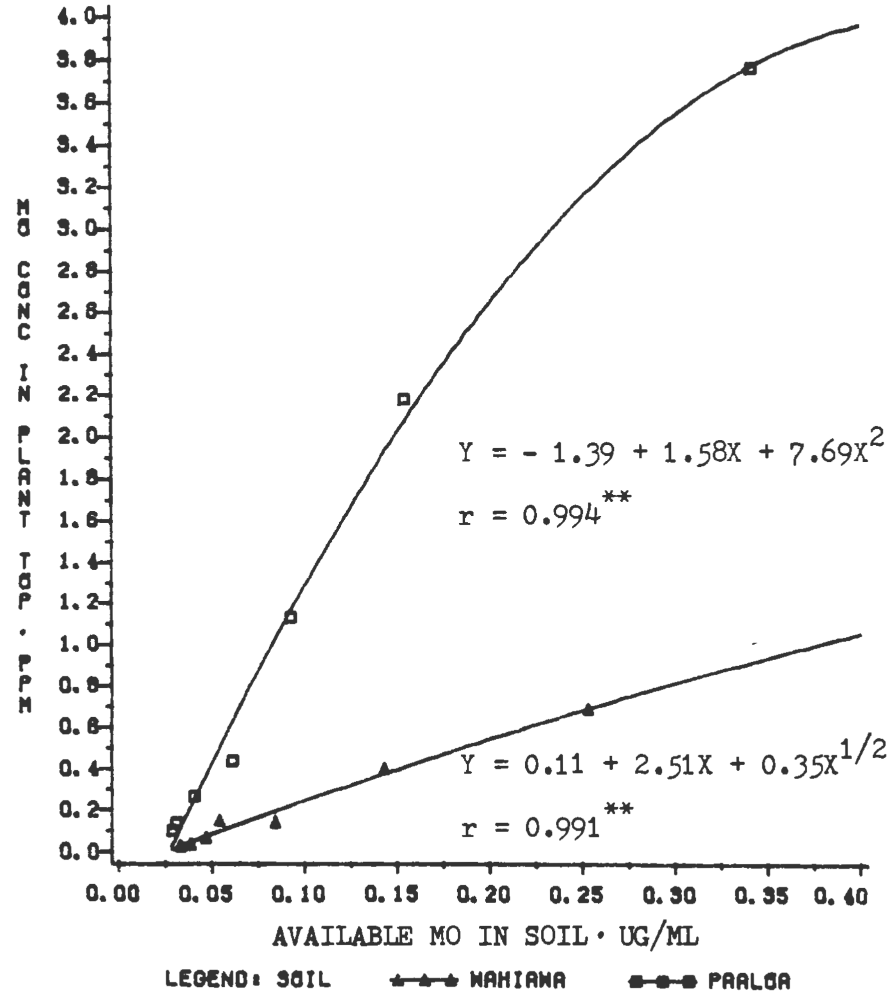


Fig. 2.6. Relationship between Mo concentration in the top of *Desmodium intortum* and available Mo in Wahiawa and Paaloa soils.

Relationship between Mo Concentration in Nodule and Available Mo in Soil

Molybdenum concentration in nodule of desmodium grown in Wahiawa soil increased from 1.65 ug/g at 0.033 ug/ml available Mo to 16.51 ug/g at 0.253 ug Mo/ml (Table 2.2). Molybdenum concentration in nodule associated with 0.09 ug Mo/ml was 7.99 ug/g (Fig. 2.7).

Molybdenum concentration in nodule of desmodium grown in Paaloa soil increased from 3.27 ug/g at 0.029 ug/ml available Mo to 23.31 ug/g at 0.342 ug Mo/ml (Table 2.2). Molybdenum concentration in nodlue associated with 0.10 ug Mo/ml was 13.42 ug/g (Fig. 2.7).

It was observed that Mo concentration in nodule of desmodium grown in the Paaloa soil was higher than that of legume grown in Wahiawa soil. Their pattern was the same as Mo concentration in plant top. However the different between Mo concentrations in nodules of desmodium grown in the two soils and receiving the same Mo treatment was less than that between Mo concentrations in plant top (Fig. 2.6 and 2.7). These results suggest that Mo concentration in nodule is a better criterion than Mo concentration in plant top to use in evaluating Mo response of plant grown in different soil types.

Relationship between Extractable Mo in soil after harvesting and Available Mo in Soil

Extractable Mo in Wahiawa soil after a crop of desmodium increased from 0.395 ug/g at 0.033 ug/ml available Mo to 1.40 ug/g at 0.253 ug Mo/ml (Table 2.4). Extractable Mo in Wahiawa soil associated with 0.09 ug Mo/ml was 0.60 ug/g (Fig. 2.8). Extractable Mo in Paaloa soil after a crop of

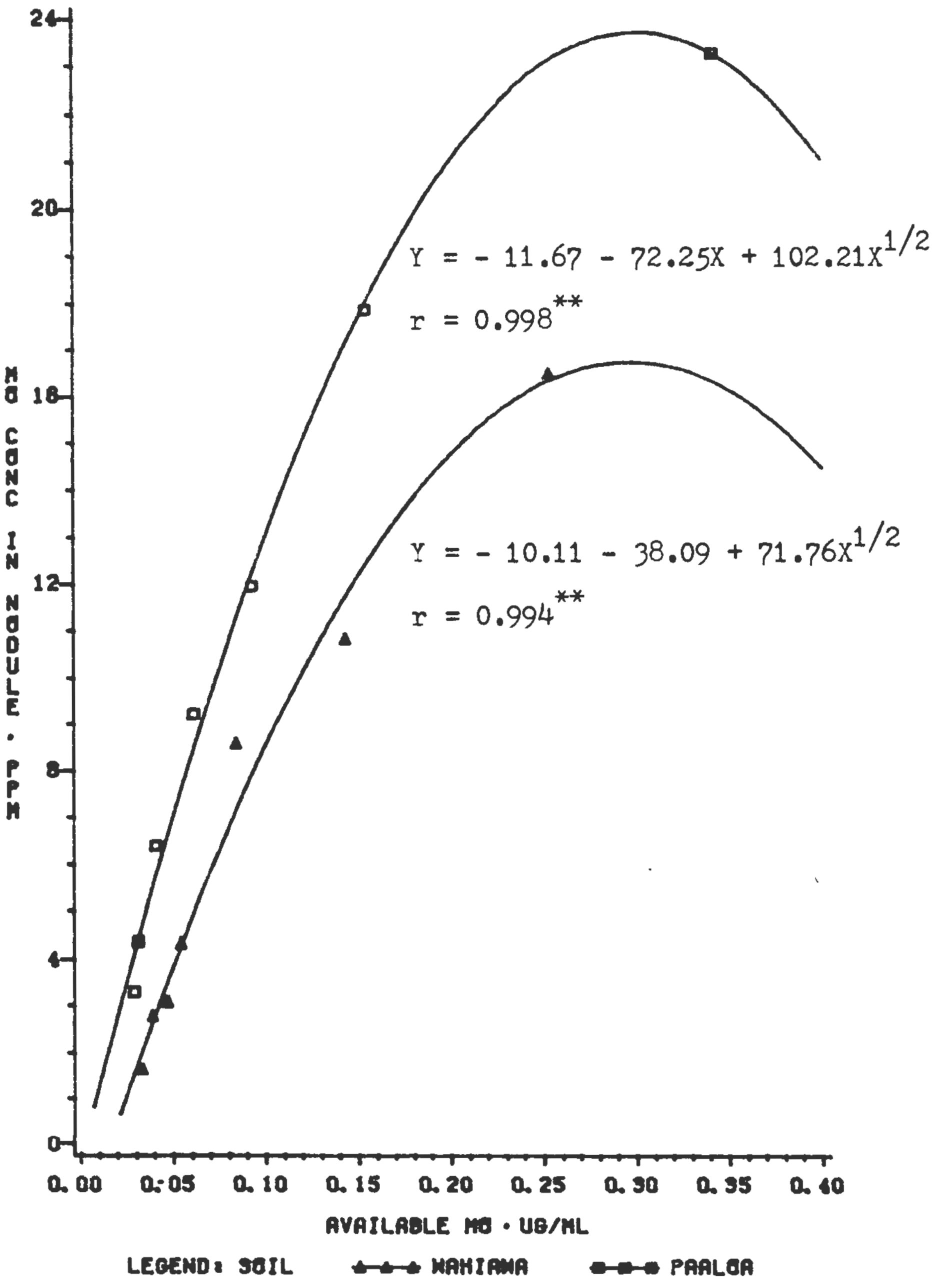


Fig. 2.7. Relationship between Mo concentration in nodule of *Desmodium intortum* and available Mo in Wahiawa and Paaloa soils.

Table 2.4.

Extractable Mo in soil after harvesting Desmodium intortum in relation to the level of available Mo in Wahiawa and Paaloa soils.

Available Mo	Extractable Mo
ug/ml	ug/g
Wahiawa soil	
0.033	0.395 c
0.039	0.438 c
0.047	0.443 c
0.054	0.548 bc
0.084	0.571 bc
0.143	0.800 b
0.253	1.400 a
Paaloa soil	
0.029	0.387 e
0.031	0.460 e
0.041	0.567 de
0.061	0.693 d
0.093	0.933 c
0.154	1.293 b
0.342	2.153 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

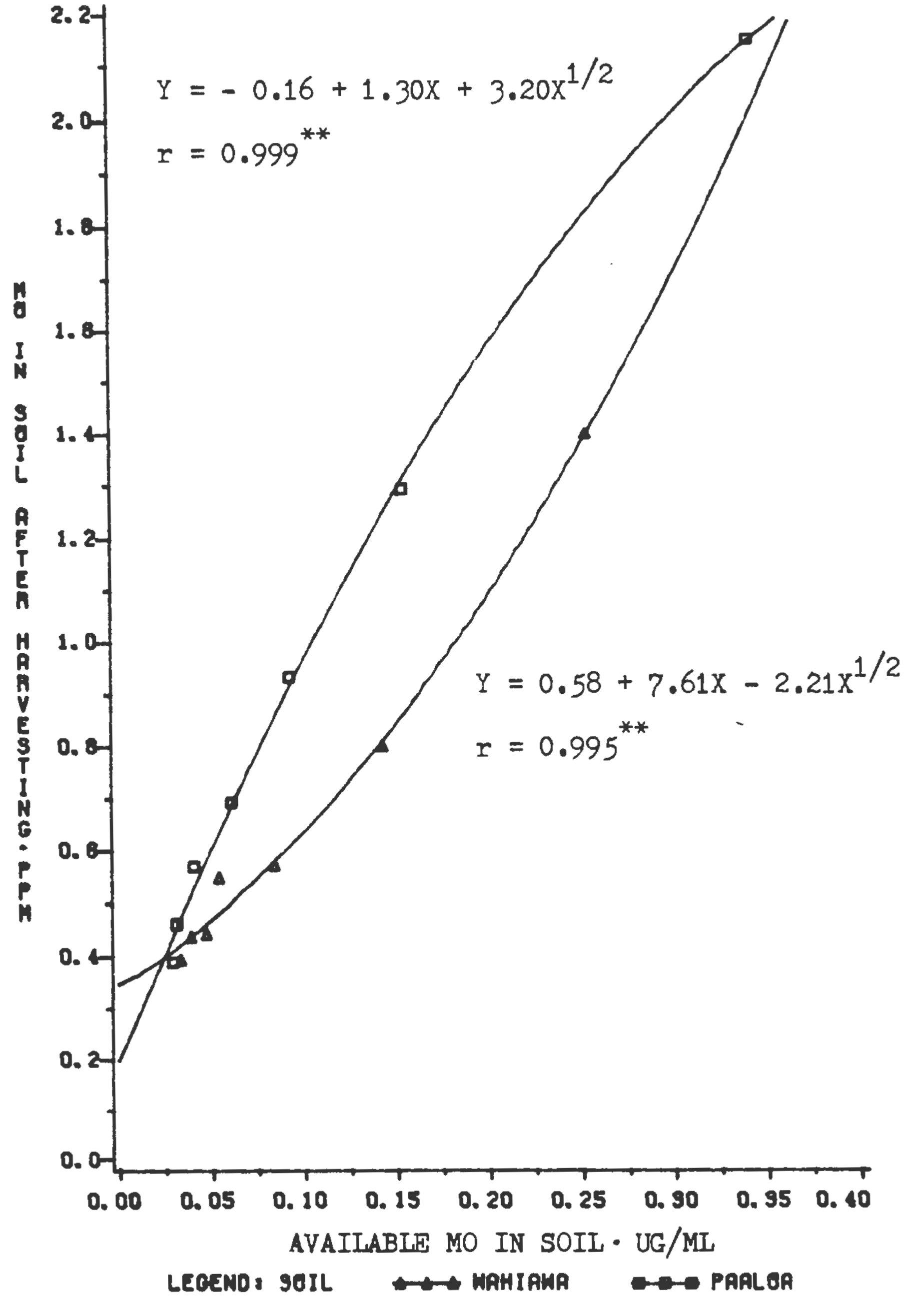


Fig. 2.8. Relationship between extractable Mo in soil after harvesting *Desmodium intortum* and available Mo in Wahiawa and Paaloa soils.

desmodium increased from 0.387 ug/g at 0.029 ug Mo/ml to 2.153 ug/g at 0.342 ug Mo/ml. Extractable Mo associated with 0.10 ug Mo/ml was 0.98 ug Mo/g (Fig.2.8).

Relationship between Nodulation and Available Mo in Soil

The nodule number of desmodium grown in Wahiawa soil increased from 103 at 0.033 ug/ml available Mo to 253 at 0.047 ug Mo/ml (Table 2.5). The number tended to decline when the available Mo was higher than 0.047 ug/ml. The maximum nodule number of desmodium grown in Paaloa soil was 559 at 0.061 ug/ml available Mo. This number was significantly higher than numbers at 0.031 and 0.342 ug Mo/ml but was not significantly higher than numbers at 0.029, 0.041, 0.093 and 0.154 ug Mo/ml (Table 2.5). It should be noted that nodule number of desmodium grown in Paaloa soil was high as compared to that of desmodium grown in Wahiawa soil. This difference was due to the fact that the Paaloa soil had higher pH (5.2) than the Wahiawa soil (4.7).

The nodule fresh weight of desmodium grown in Wahiawa soil increased from 1.07 g/pot at 0.033 ug/ml available Mo to 2.97 g/pot at 0.047 ug Mo/ml (Table 2.4). Nodule fresh weight of desmodium tended to decline when the available Mo was higher than 0.047 ug/ml. The nodule fresh weight of desmodium grown in Paaloa soil increased from 1.96 g/pot at 0.029 ug/ml available Mo to 4.76 g/pot at 0.061 ug Mo/ml. Nodule fresh weight tended to decline when the available Mo was higher than 0.061 ug/ml (Table 2.5).

The nodule dry weight of desmodium grown in Wahiawa soil increased from 0.21 g/pot at 0.033 ug/ml available Mo to 0.53 g/pot at 0.047 ug Mo/ml

Table 2.5.

Nodule number, nodule fresh weight and nodule dry weight of Desmodium intortum in relation to the level of available Mo in Wahiawa and Paaloa soils.

Available Mo	Nodule number	Nodule weight	
		Fresh weight	Dry weight
ug/ml		----------- g/pot	-----------
	Wahiawa soil		
0.033	103 b	1.07 b	0.21 c
0.039	107 b	1.51 b	0.27 bc
0.047	253 a	2.97 a	0.53 a
0.054	234 a	2.95 ab	0.45 ab
0.084	191 ab	1.92 ab	0.35 abc
0.143	175 ab	1.94 ab	0.36 abc
0.253	143 ab	1.75 ab	0.32 abc
	Paaloa soil		
0.029	456 ab	1.96 c	0.36 c
0.031	364 b	2.46 c	0.45 b
0.041	394 ab	3.27 abc	0.61 abc
0.061	559 a	4.76 a	0.91 a
0.093	431 ab	4.31 ab	0.81 ab
0.154	451 ab	3.49 abc	0.73 abc
0.342	364 b	2.77 bc	0.52 bc

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

(Table 2.5). Nodule dry weight tended to decline when the available Mo was higher than 0.047 ug Mo/ml. The nodule dry weight of desmodium grown in Paaloa soil increased from 0.36 g/pot at 0.029 ug/ml available Mo to 0.91 g/pot at 0.061 ug Mo/ml (Table 2.5). Nodule dry weight tended to decline when the available Mo was higher than 0.061 ug/ml. The trend of the relationship between nodule fresh weight of desmodium grown in the two soils and available Mo was similar to that of the relationship between nodule dry weight and available Mo.

Relationship between Nitrogenase Activity and Available Mo in Soil

The nitrogenase activity of desmodium grown in Wahiawa soil increased from 0.004 umole C_2H_2/plant/hr at 0.003 ug/ml available Mo to 0.056 umole C_2H_2/plant/hr at 0.047 ug Mo/ml (Table 2.6). The nitrogenase activity of legume tended to decline when the available Mo was higher than 0.047 ug Mo/ml.

The nitrogenase activity of desmodium grown in Paaloa soil increased from 0.033 umole C_2H_2/plant/hr at 0.029 ug/ml available Mo to 0.396 umole C_2H_2/plant/hr at 0.093 ug Mo/ml (Table 2.6). The nitrogenase activity of plant tended to decline when the available Mo was higher than 0.093 ug Mo/ml (Table 2.5).

Relationship between N Concentration in Plant Top and Available Mo in Soil

Nitrogen concentration in the top of desmodium grown in Wahiawa soil was increased from 1.50% at 0.033 ug/ml available Mo to 1.83% at 0.143 ug Mo/ml (Table 2.6). Nitrogen concentration tended to decrease when the

Table 2.6.

Nitrogenase activity and N concentration in the top of Desmodium intortum in relation to the level of available Mo in Wahiawa and Paaloa soils.

Available Mo	Nitrogenase activity	N concentration
ug/ml	umole C_2H_2/plant/hr	%
Wahiawa soil		
0.033	0.004 a	1.50 b
0.039	0.007 a	1.49 b
0.047	0.056 a	1.65 ab
0.054	0.052 a	1.59 ab
0.084	0.015 a	1.63 ab
0.143	0.010 a	1.83 a
0.253	0.011 a	1.69 ab
Paaloa soil		
0.029	0.033 b	2.78 a
0.031	0.060 ab	2.85 a
0.041	0.196 ab	2.92 a
0.061	0.101 ab	2.96 a
0.093	0.396 ab	3.04 a
0.154	0.071 ab	3.05 a
0.342	0.055 ab	3.10 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

available Mo was higher than 0.143 ug/ml (Fig. 2.9).

Nitrogen concentration in the top of desmodium grown in Paaloa soil was increased from 2.78% at 0.029 ug/ml available Mo to 3.10% at 0.342 ug Mo/ml (Table 2.6).

According to the curves in Fig. 2.9, N concentration in the top of desmodium grown in Wahiawa or Paaloa soil increased as the available Mo increased up to a certain level (0.15 ug Mo/ml for Wahiawa soil and 0.25 ug Mo/ml for Paaloa soil) and then declined. This trend suggested that a high level of Mo was likely to decrease N concentration in plant top.

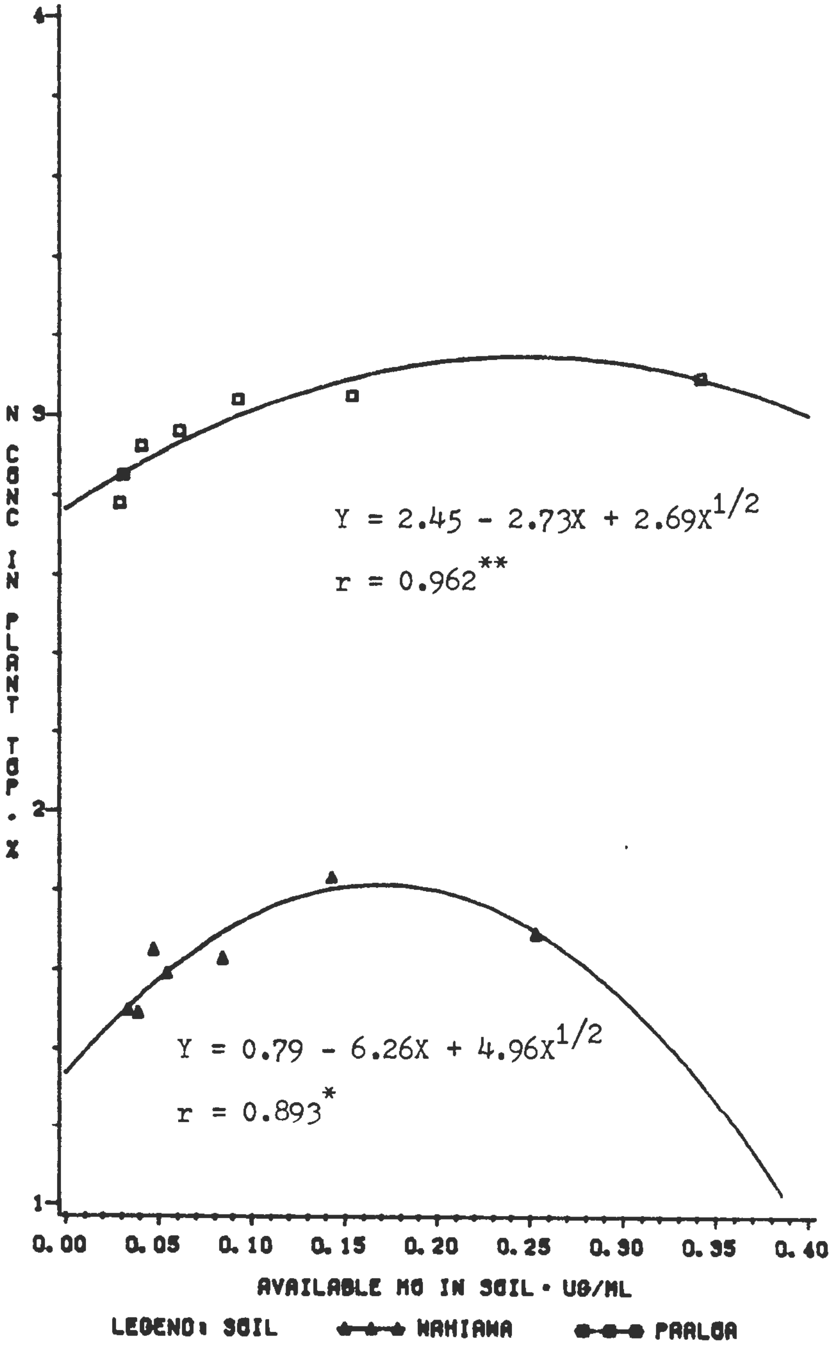


Fig. 2.9. Relationship between N concentration in the top of *Desmodium intortum* and available Mo in Wahiawa and Paaloa soils.

SUMMARY

The available Mo in soil associated with 95% maximum yield of desmodium grown in Wahiawa and Paaloa soils were 0.09 and 0.10 ug/ml, respectively. Predicted quantity of applied Mo required to establish 0.09 ug/ml available Mo for Wahiawa soil was 4.4 kg Mo/ha. For Paaloa soil, predicted quantity of applied Mo required to establish 0.10 ug/ml available Mo was 4.7 kg Mo/ha.

Dry matter yield of desmodium grown in Wahiawa soil increased as extractable Mo after harvesting increased. Extractable Mo associated with 80% and 95% maximum yield were 0.40 and 0.60 ug/g, respectively. Yield of desmodium grown in limed Paaloa soil increased as extractable Mo increased. Extractable Mo associated with 80% and 95% maximum yield were 0.50 and 0.98 ug/g, respectively.

Dry matter yield of desmodium grown in Wahiawa soil increased as the Mo concentration in nodule increased. Critical Mo concentration in nodule associated with 80% and 95% maximum yield were 2.20 and 7.85 ppm, respectively. Yield of desmodium grown in limed Paaloa soil increased as Mo concentration in nodule increased. Critical Mo concentration in nodule associated with 80% and 95% maximum yield were 4.0 and 13.2 ppm, respectively.

Dry matter yield of desmodium grown in Wahiawa soil increased as Mo concentration in plant top increased. Critical Mo concentration in plant top associated with 95% maximum yield was 0.22 ppm. Critical Mo concentration in the top of desmodium grown in limed Paaloa soil and associated with 95% maximum yield was 1.30 ppm.

CHAPTER III

INFLUENCE OF MYCORRHIZAE ON YIELD AND MOLYBDENUM RESPONSE OF PASTURE LEGUMES GROWN IN ACID TROPICAL SOIL

INTRODUCTION

Vesicular-arbuscular mycorrhiza (VAM) has been known to aid host legume by increasing P uptake in infertile tropical soil. In a P-deficient situation of Wahiawa series (Tropeptic Eutrustox), plant concentration of P was enhanced by mycorrhizal association (Yost and Fox, 1979). Mycorrhizal hyphae will help the plant overcome uptake limitations in the case of nutrient like phosphate that diffuses slowly and cause depleted zones around roots. Conversely, mycorrhiza confers little additional advantage to ions such as sulfate which moves through soil by mass flow with no major limiting step on the way to the root surface. Although root interception and mass flow are considered to be the important mechanisms controlling the movement of molybdate to plant roots (Barber et al., 1966), molybdate does not move rapidly through soil by mass flow like sulfate, especially in soil that highly adsorbs molybdate (e.g. Wahiawa soil). In this type of soil molybdate may behave like phosphate in regard to its availability to plant. Moreover, reactions of phosphate and molybdate in soil are similar in that they specifically adsorbed and they continue to react slowly over a long period in soil. With this similarity between phosphate and molybdate, it was deemed possible that mycorrhiza may enhance Mo absorption by plant roots and increase yield with regard to Mo status in soils.

This part of the study was conducted in the greenhouse to evaluate the influence of mycorrhizae on the yield and Mo response of _Desmodium intortum_, _Centrosema pubescens_ and _Stylosanthes humilis_ .

MATERIALS AND METHODS

1. Procedures

A pot experiment was conducted at the Agronomy and Soil Science Field Laboratory, Mauka Campus of the University of Hawaii at Manoa. The soil was a Tropeptic Eutrustox, Wahiawa series. Some chemical properties of this soil are shown in Table 2.1. The soil was limed to pH 6 and it was fumigated with methyl bromide before applying treatments.

Treatments were composed of: three pasture legume species (*Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis*); three levels of Mo (none added, seed-applied Mo at 0.3 kg Mo/ha and soil-applied Mo at 2.0 kg Mo/ha); and two mycorrhizal inoculation rates (non-inoculation and inoculation). These treatments made up the 3x3x2 factorial arrangement in a randomized complete block design with three replications. The legumes were inoculated with the appropriate *Rhizobium* strains. Potassium, Mg, S, Zn, Cu and B were added to all treatments as part of the basal fertilizer application. Thirty percent (w/v) gum arabic solution was used as adhesive material for *Rhizobium* inoculation and seed-applied Mo treatment.

Thirty treated seeds of a legume were planted in a 6-l plastic pot. After four weeks from planting, the seedling were thinned to six uniformly growing plants per pot. Throughout the experiment the pots were watered daily to 100% field capacity. The plants were harvested at 90 days after planting. They were cut approximately 1 cm above the soil surface and the roots were carefully removed from the soil with a minimum detachment of nodules. The roots with nodules were put into

500-ml glass bottles for acetylene reduction assessment. After finishing the assessment, the nodules were removed and the roots were kept in a refrigerator for evaluation of mycorrhizal infection. Plant samples were rinsed with deionized water and oven-dried at 70° C in an air-draft oven for three days for determining dry matter yield. The oven-dried samples were ground (20 mesh) in a stainless steel Wiley Mill and kept for elemental analysis.

2. Treatment Preparation

2.1. Fertilizers and Lime Used

Fertilizers and lime used in the experiment were as follow:

2.1.1. Basal Fertilizers

- Potassium chloride (KCl) at 200 kg K/ha.
- Magnesium sulfate ($MgSO_4.7H_2O$) at 200 kg Mg/ha.
- Zinc sulfate ($ZnSO_4.7H_2O$) at 20 kg Zn/ha.
- Copper sulfate ($CuSO_4.5H_2O$) at 3 kg Cu/ha.
- Boric acid (H_3BO_3) at 2 kg B/ha.

2.1.2. Molybdenum Fertilizer

- Sodium molybdate ($Na_2MoO_4.2H_2O$) at 0.3 kg Mo/ha seed-applied and 2.0 kg Mo/ha soil-applied.

2.1.3. Lime

- Calcium hydroxide ($Ca(OH)_2$) at rate such that the pH of soil was raised 6.0.

Basal fertilizers were applied in solution form. Each chemical was weighed at the required amount for the whole treatment, mixed together in solution form and made up to 1620-ml volume. The solution fertilizer was applied at the rate of 30 ml per pot at planting.

2.2. Seed Preparation

Seeds of Centrosema pubescens and Stylosanthes humilis were scarified by immersing in concentrated H_2SO_4 for 10 minutes. After draining the acid, the seeds were washed many times with deionized water. The scarified seeds were spread to dry before they were treated with sodium molybdate and inoculated with Rhizobium strains.

2.3. Soil Preparation

The soil was sieved through a 1-cm sized sieve to preclude gravels, stones and large roots and spread on benches to dry. After air-drying and thorough mixing to attain homogeneity, a soil sample was taken to determine moisture content as well as to conduct chemical analysis for determining some soil chemical properties. Four kilograms of oven-dried soil was put into 6-l plastic pots for subsequent liming, fumigation and fertilizer application. Calcium hydroxide at an amount needed to raise soil pH to 6.0 according to a lime requirement curve was mixed with soil and the limed soil was incubated for 2 weeks before fumigation.

Soil in the pots was fumigated with 0.8 kg/27 pots of methyl bromide applied under a plastic cover. The cover remained in place for 48 hr. The purpose of the fumigation was to destroy mycorrhizae-producing propagules. Legume seeds were planted 1 month after fumigation.

2.4. Mycorrhizal Inoculation

The fungal symbiont used was Glomus mosseae, stock-cultured on corn roots (Zea mays) in 8-l pots filled with crushed basalt. Vesicular-arbuscular mycorrhizal inoculum consisted of coarsely chopped root fragments and the basalt from the corn pot cultures.

At planting, 25 g of mycorrhizal inoculum was placed approximately 2-cm deep in the soil. The treated legume seeds were then planted above the inoculum.

To ensure the introduction of similar rhizosphere microflora into both mycorrhizal and nonmycorrhizal treatments, root washings from the mycorrhizal-stock cultures were applied to all pots. Root washings were passed through a 325-mesh sieve to separate mycorrhizal spores before applying to the pots.

2.5. Mycorrhizal infection Measurement

Procedures described by Kormanik and McGraw (1982) were used to measure mycorrhizal infection.

Root segments were heated at 90° C in 10% KOH for at least 1 hr. The potassium hydroxide solution was poured off and the root segment were washed three times with deionized water. Washed roots were immersed in alkaline solution of H_2O_2 at room temperature until they were bleached (10-20 minutes). The root segments were thoroughly rinsed with deionized water to removed all the H_2O_2 and then acidified with 1% HCl. The acidified root segments were stained by simmering at 90° C for 10 to 60 minutes in lactic acid-acid Fuchsin solution. Stained roots were examined in an open petri dish with a dissecting microscope. Percentage of roots infected by mycorrhizae was estimated using Biermann and Linderman's method (1981) by viewing 20 randomly selected root segments from each treatment. The length of each root segment which contained fungal structures (vesicles or arbuscles), in addition to hyphae, was estimated and the total percentage of root infected was then calculated on the basis of 20 root segments examined.

3. Measurement of Responses

3.1. Mycorrhizal Infection

Mycorrhizal infection was evaluated by examining the fine roots (less than 2 mm diameter) of plants at harvest. Relationship between mycorrhizal infection and Mo concentration was determined.

3.2. Yield Response

Plants were observed for deficiency and/or toxicity symptoms. Dry matter yield was determined at harvest. The efficiency of Mo applied as seed coating and soil treatment regarding the influence of mycorrhizae was compared. The relationship between dry matter yield and mycorrhizal infection was determined.

3.3. Nodulation

The number and weight of nodules of pasture legumes were determined at harvest in order to assess the influence of mycorrhizae and Mo on legume nodulation.

3.4. Molybdenum Concentration

Molybdenum concentrations in plant top and nodule were determined.

Extractable Mo in soil after harvesting was also determined. The relationship between extractable Mo and mycorrhizal infection was evaluated.

RESULTS AND DISCUSSION

Mycorrhizal Infection

Methyl bromide fumigation eliminated vesicular-arbuscular mycorrhizal (VAM) infection; there was no VAM root colonization occurring in the uninoculated plant (Table 3.1). Mycorrhizal infection occurred in plant inoculated with the fungi. When averaged over all mycorrhizal and Mo levels, percentages of mycorrhizal infection of desmodium, centrosema and stylosanthes were not significantly different from each other. When averaged over all legume species and mycorrhizal inoculationlevels, seed-applied Mo at 0.3 kg Mo/ha significantly increased mycorrhizal infection (Table 3.1). However, soil-applied Mo at 2.0 kg Mo/ha did not significantly increase the infection. When individual legume was considered, it was observed that mycorrhizal infection of desmodium or centrosema receiving seed-applied Mo was significantly higher than those of the same legume receiving soil-applied Mo and no Mo. Mycorrhizal infection in stylosanthes receiving seed-applied and soil-applied Mo was significantly higher than that in stylosanthes receiving no Mo.

The influence of Mo concentration in increasing fungal infection has been reported (Joham, 1953). He found that high Mo concentration in the substrate increased the rate of infection of cotton leaves by Ascophyta gossypii. He reasoned that the Mo concentration of cotton leaves or a high Mo requirement of the fungi may have been responsible for a decrease in the physiological resistance of cotton to the infection of A. gossypii. Golov and Kazakhkov (1973) also reported that Mo

Table 3.1.

Influences of mycorrhizae and Mo on mycorrhizal infection of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in the Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
				%			
No Mo added	0 d*	25.0 bc	0 d	16.6 c	0 d	21.7 c	10.5 b
2.0 kg Mo/ha (soil applied)	0 d	24.9 bc	0 d	19.9 c	0 d	34.2 ab	13.2 b
0.3 kg Mo/ha (seed applied)	0 d	36.9 a	0 d	33.9 ab	0 d	38.4 a	18.2 a
	0	28.9	0	23.4	0	31.5	
Mean	14.5 a		11.7 a		15.7 a		

	No Myc.	Myc.
Mean	0 b	27.9 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

accumulated in soybean leaves mainly in areas infected with fungi resembling Ascophyta in structure. These findings suggested that there was a relationship between Mo concentration and fungal infection. Mycorrhizal fungi, although not of the same species of fungi that cause a disease in cotton and soybean, may also require high Mo concentration for their multiplication and infection. The influence of Mo concentration in mycorrhizal infection was likely since seed-applied Mo which provided high Mo concentration in a spot significantly increased percentage of mycorrhizal infection in this experiment.

Yield Response

New seedling viewed just after the cotyledon leaf stage were uniformly healthy and green. As seed reserves of nutrient were depleted, emerging new leaves of nonmycorrhizal plant appeared progressively more stunted, misshappen, and bronzed, with ultimate necrosis and abscission of some leaves as compared to leaves of mycorrhizal plant. After 4 weeks of growth in individual pots, treatment differences were strinking. Nonmycorrhizal desmodium, centrosema and stylosanthes were smaller than mycorrhizal plants. While nonmycorrhizal plants were stunted and grew very slowly, mycorrhizal plants grew steadily and appeared healthy. It was clear that mycorrhizae enhanced plant growth. When averaged over all legume species and Mo levels, dry matter yield of mycorrhizal plant was significantly higher than that of nonmycorrhizal plant (Table 3.2).

When averaged over all mycorrhizal and Mo levels, dry matter yields of desmodium and stylosanthes were not significantly different from each other. Yield of centrosema was significantly lower than that of desmodium

Table 3.2.

Influences of mycorrhizae and Mo on dry matter yields of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in the Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	g/pot						
No Mo added	0.13 b*	16.4 a	0.20 c	1.43 bc	0.37 c	10.9 b	4.91 a
2.0 kg Mo/ha (soil applied)	0.53 b	17.5 a	0.40 c	4.67 ab	0.60 c	18.3 a	7.00 a
0.3 kg Mo/ha (seed applied)	0.53 b	15.2 a	0.43 c	6.00 a	0.63 a	20.7 a	7.25 a
	0.40	16.4	0.34	4.03	0.53	16.6	
Mean	8.40 a		2.19 b		8.57 a		

	No Myc.	Myc.
Mean	0.43 b	12.3 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

and stylosanthes. The reason that yield of centrosema was low may be due to the low mycorrhizal infection (16.6% to 33.9%) in this study. Centrosema was likely to be sensitive to mycorrhizal infection. Crush (1974) reported that in phosphate-deficient soil, VAM strongly stimulated growth of Centrosema pubescens. He also found that the growth of Stylosanthes guyanensis was also strongly stimulated by VAM.

Although dry matter yields of legumes receiving soil-applied and seed-applied Mo were 42.6% and 47.7%, respectively, higher than that of legumes receiving no Mo, when averaged over all mycorrhizal levels and legume species, the differences were not significant. However, these results showed that Mo tended to increase yield. When individual legume was considered, mycorrhizal stylosanthes receiving soil-applied and seed-applied Mo significantly increased yield (Table 3.2).

Nodulation Response

Number of Nodules

Desmodium, centrosema and stylosanthes inoculated with G. mosseae formed nodules. Nonmycorrhizal plants did not nodulate (Table 3.3). These results demonstrated that VAM strongly stimulated nodulation of these legumes. When averaged over all mycorrhizal and Mo levels, the nodule number of centrosema was significantly lower than that of desmodium and stylosanthes. The reason that the nodule number of centrosema was less than that of the others legumes was probably due to its low percentage of mycorrhizal infection. Molybdenum did not significantly increase nodule number when averaged over all legume species and mycorrhizal levels. However, nodule number of mycorrhizal centrosema and

Table 3.3.

Influences of mycorrhizae and Mo on nodule number of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	number						
No Mo added	0	486	0	18	0	440	157 a*
2.0 kg Mo/ha (soil applied)	0	436	0	92	0	915	241 a
0.3 kg Mo/ha (seed applied)	0	409	0	67	0	736	202 a
	0	444	0	59	0	697	
Mean	222 a		30 b		349 a		

	No Myc.	Myc.
Mean	0 b	400 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

stylosanthes receiving seed-applied or soil-applied Mo tended to be higher than that of legume receiving no Mo.

Nodule Fresh Weight

Mycorrhizal legumes significantly produced higher nodule fresh weight than nonmycorrhizal legumes since nonmycorrhizal plants did not nodulate. When average over all mycorrhizal and Mo levels, nodule fresh weight of desmodium was significantly higher than those of centrosema and stylosanthes (Table 3.4). The nodule weight of centrosema was low because of its small nodule number (Table 3.3). Although stylosanthes had a larger nodule number than desmodium, the nodule size was much smaller than that of desmodium, so that nodule weight of stylosanthes was less than that of desmodium. When averaged over all legume species and mycorrhizal levels, Mo did not significantly increase nodule fresh weight of legumes.

Nodule Dry Weight

Mycorrhizal legumes produced significantly higher nodule dry weight than nonmycorrhizal legumes which had no nodule (Table 3.5). When averaged over all mycorrhizal and Mo levels, nodule weight of desmodium was significantly higher than those of centrosema and stylosanthes. Molybdenum application did not significantly increase nodule weight of desmodium, centrosema and stylosanthes in this experiment.

Table 3.4.

Influences of mycorrhizae and Mo on nodule fresh weight of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	g/pot						
No Mo added	0	3.35	0	0.15	0	0.24	0.62 a*
2.0 kg Mo/ha (soil applied)	0	2.90	0	0.75	0	0.95	0.76 a
0.3 kg Mo/ha (seed applied)	0	2.35	0	0.81	0	0.62	0.63 a
	0	2.87	0	0.57	0	0.60	
Mean	1.43 a		0.29 b		0.30 b		

	No Myc.	Myc.
Mean	0 b	1.35 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 3.5.

Influences of mycorrhizae and Mo on nodule dry weight of _Desmodium intortum_, _Centrosema pubescens_ and _Stylosanthes humilis_ grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	**Myc.**	**No Myc.**	**Myc.**	**No Myc.**	**Myc.**	
	g/pot						
No Mo added	0	0.65	0	0.02	0	0.09	0.13 a*
2.0 kg Mo/ha (soil applied)	0	0.55	0	0.15	0	0.27	0.16 a
0.3 kg Mo/ha (seed applied)	0	0.46	0	0.16	0	0.20	0.14 a
	0	0.55	0	0.11	0	0.18	
Mean	0.28 a		0.05 b		0.09 b		

	No Myc.	**Myc.**
Mean	0	0.28 a

*** Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.**

Molybdenum Uptake

Molybdenum Concentration in Nodule

The small amount of nodules of some treatments required the pooling of treatment replications for nodule analysis, thereby precluding statistical evaluation of the data. However, there was a trend indicating that Mo application increased Mo concentration in nodule (Table 3.6). When averaged over all legume species and mycorrhizal levels, Mo concentration of legumes receiving seed-applied Mo was 215% higher than that of legumes receiving no Mo. When averaged over all mycorrhizal and Mo levels, Mo concentration in nodule of desmodium was higher than those of centrosema and stylosanthes (Table 3.6). Molybdenum concentration of stylosanthes was also higher than that of centrosema. Molybdenum concentration in nodule of mycorrhizal and nonmycorrhizal plants could not be compared because nonmycorrhizal plants had no nodule to be analyzed. Although mycorrhizal plants contained high Mo concentration in nodule, it could not be concluded that mycorrhizae directly increased Mo concentration in nodule. In this case mycorrhizae enhanced Mo uptake indirectly through their stimulation of nodulation.

Molybdenum Concentration in Plant Top

The stunted growth of nonmycorrhizal plants required the pooling of treatment replications for Mo analysis, thereby precluding statistical evaluation of plant Mo data. However, there was a trend indicating that Mo application increased Mo concentration in the top with a greater effect from seed-applied Mo than from soil-applied Mo (Table 3.7). When averaged over all Mo and mycorrhizal levels, Mo concentration in

Table 3.6.

Influences of mycorrhizae and Mo on Mo concentration in nodule of Desmodium intortum, Centrosema pubescens and Stylosanthes humilis grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	ppm						
No Mo added	-	7.51	-	-	-	4.78	2.05
2.0 kg Mo/ha (soil applied)	-	7.31	-	5.59	-	4.35	2.88
0.3 kg Mo/ha (seed applied)	-	17.46	-	6.14	-	15.13	6.46
	-	10.76	-	5.05	-	8.09	
Mean	5.38		2.53		4.05		

	No Myc.	Myc.
Mean	-	7.97

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

Table 3.7.

Influences of mycorrhizae and Mo on Mo concentration in the top of *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	ppm						
No Mo added	0.49	0.30	1.02	0.63	0.55	0.33	0.55
2.0 kg Mo/ha (soil applied)	1.56	0.33	1.20	1.03	0.84	0.46	0.90
0.3 kg Mo/ha (seed applied)	1.38	1.15	1.40	1.11	1.42	1.47	1.32
	1.14	0.59	1.21	0.92	0.94	0.75	
Mean	0.87		1.07		0.85		

	No Myc.	Myc.
Mean	1.40	1.24

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

centrosema tended to be higher than that in desmodium and stylosanthes. When averaged over all legume species and Mo levels, Mo concentration in the top of mycorrhizal legumes tended to be lower than that of nonmycorrhizal plants (Table 3.7). This difference was probably due to a "dilution effect" of growth since dry matter yield of mycorrhizal plants was much greater than that of nonmycorrhizal plants. If plant Mo is used to evaluate Mo uptake by plant, mycorrhizae are not likely to enhance Mo absorption since nonmycorrhizal plant contained Mo similar to mycorrhizal plant.

Extractable Mo in Soil

When averaged over all legume species and mycorrhizal levels, extractable Mo in soil treated with seed-applied Mo at 0.3 kg Mo/ha was significantly higher than that in soil treated with no Mo (Table 3.8). When averaged over all Mo and mycorrhizal levels, extractable Mo in soil grown with desmodium, centrosema and stylosanthes was not significantly different from each other. When averaged over all legume species and Mo levels, extractable Mo in soil grown with mycorrhizal plants was significantly higher than that in soil grown with nonmycorrhizal plants. The reason that more Mo was extracted from the soil with mycorrhizal plants could be due to the influence of root activities (e.g. root exudation, rhizosphere activity) on Mo avialability. Mycorrhizal plants developed a much more extensive root system than nonmycorrhizal plants.

Table 3.8.

Influences of mycorrhizae and Mo on extractable Mo in soil after harvesting *Desmodium intortum*, *Centrosema pubescens* and *Stylosanthes humilis* grown in Wahiawa soil.

Mo treatment	Desmodium		Centrosema		Stylosanthes		Mean
	No Myc.	Myc.	No Myc.	Myc.	No Myc.	Myc.	
	ppm						
No Mo added	0.26	0.47	0.29	0.37	0.25	0.33	0.33 b*
2.0 kg Mo/ha (soil applied)	0.29	0.44	0.34	0.42	0.29	0.52	0.39 ab
0.3 kg Mo/ha (seed applied)	0.41	0.58	0.39	0.51	0.29	0.56	0.46 a
	0.32	0.50	0.34	0.44	0.28	0.47	
Mean	0.41 a		0.39 a		0.37 a		

	No Myc.	Myc.
Mean	0.31 b	0.47 a

* Values of each factor followed by the same letter are not significantly different at the 5% probability level according to Duncan's Multiple Range Test.

SUMMARY

Mycorrhizal infection at the range of 13.2 to 38.4% of root infection occurred in legumes inoculated with *Glomus mosseae* fungi. There was no mycorrhizal infection occurring in legumes that were not inoculated with the fungi. Seed-applied Mo at 0.3 kg Mo/ha increased mycorrhizal infection, suggesting that there was a relationship between Mo concentration in soil and fungal infection.

Mycorrhizae enhanced plant growth and increased dry matter yield of desmodium, centrosema and stylosanthes. Nonmycorrhizal plants were stunted and grew very slowly, whereas mycorrhizal plants grew steadly and appeared healthy. Yields of mycorrhizal legumes was 29-fold higher than that of nonmycorrhizal legumes. Molybdenum fertilizer did not increase yields of nonmycorrhizal legumes. However, Mo fertilizer increased yields of mycorrhizal centrosema and stylosanthes.

Mycorrhizae strongly stimulated nodulation of legumes. Desmodium, centrosema and stylosanthes inoculated with mycorrhizal fungi had formed nodules whereas those legumes uninoculated with the fungi had not nodulated. Molybdenum fertilizer did not significantly increase nodule nomber of these legumes. Nodule number of centrosema was less than those of desmodium and stylosanthes. Mycorrhizal legumes significantly produced higher nodule fresh and dry weights. Molybdenum fertilizer did not significantly increase nodule fresh and dry weights of legumes. Nodule fresh and dry weights of desmodium were higher than those of centrosema and stylosanthes.

Molybdenum concentration in nodule of mycorrhizal and nonmycorrhizal legumes could not be compared because nonmycorrhizal legumes did not nodulate. Molybdenum concentration in nodule of centrosema tended to be lower than those of desmodium and stylosanthes.

Molybdenum concentration in plant top of mycorrhizal legumes tended to be lower than that in plant top of nonmycorrhizal legumes due to a "dilution effect" of growth since dry matter yield of mycorrhizal legumes were makedly higher than that of nonmycorrhizal legumes. Molybdenum concentration in the top of centrosema tended to be lower than in desmodium and stylosanthes.

Extractable Mo in soil growing mycorrhizal plant was significantly higher than that in soil growing nonmycorrhizal plant. This effect could be due to the influence of plant roots on Mo availability in soil since mycorrhizal plant developed a much more extensive root system than nonmycorrhizal plant.

CONCLUSIONS

Molybdenum fertilizer increased dry matter yield of Desmodium intortum grown in Wahiawa and Paaloa soils. It also increased yield of Centrosema pubescens grown in the Wahiawa soil. Both seed treatment and soil application are practical methods of Mo fertilization. However, seed treatment requires a smaller amount of Mo than soil application. For soil-applied Mo, at least 1.0 kg Mo/ha was required for desmodium and centrosema grown in the Wahiawa soil. However, in order to obtain excellent stands of legumes grown in the Wahiawa soil in the field, soil-applied Mo at 4.0 kg Mo/ha should be used. Moreover, soil-applied Mo at 4.0 kg Mo/ha plus lime produced the maximum yields of desmodium and centrosema in the fifth cutting of the field experiment, indicating the combined residual beneficial effects of Mo and lime. Soil-applied Mo at 4.0 kg Mo/ha which was successfully used in the experimental field is close to the predicted quantity of applied Mo (4.4 kg Mo/ha) to obtain 95% maximum yield of desmodium grown in the Wahiawa soil in the greenhouse.

Molybdenum application rates that were effectively used in these greenhouse and field experiments are higher than the recommended rates (140-200 g Mo/ha) for tropical legumes grown in acid soils of sub tropical Queensland in Australia. The difference in Mo rates was likely due to the ability of the Hawaiian soils to absorb Mo. The Wahiawa (Oxisol) and Paaloa (Ultisol) soils in Hawaii strongly adsorb Mo. The Mo adsorbed to give an equilibrium soil solution value of 0.002 ug/ml for the Wahiawa and Paaloa soils were 18.3 and 28.8 ug/g, respectively.

Seed-applied Mo at 0.5 kg Mo/ha was as effective as soil-applied Mo at 4.0 kg Mo/ha up to the fifth cutting. Seed-applied Mo at 0.3 kg Mo/ha also provided enough Mo upto the fourth cutting but at the fifth cutting the residual effect of seed-applied Mo at this rate tended to decline. Seed-applied Mo at 0.1 kg Mo/ha did not supply an adequate amount of Mo for plant needs. Therefore, it is recommended that Mo fertilizer at a rate higher than 0.1 kg Mo/ha should be used if seed treatment is the method selected to apply Mo fertilizer. However, seed treatment should be used with caution since concentrated Mo fertilizer can kill rhizobia in the inoculant and cause nodulation failure. The results obtained from the greenhouse experiment with desmodium grown in the Paaloa soil revealed that seed-applied Mo at 0.5 kg Mo/ha tended to decrease nodule number of the legume. Seed-applied Mo at this rate decreased nodule fresh and dry weights. For the establishment of pasture legumes in areas that have never grown any species of legume before and where rhizobia population is expected to be small, seed-applied Mo at high rates (e.g. 0.5 kg Mo/ha) should not be used.

Sulfur did not increase dry matter yield of desmodium and centrosema grown in the Wahiawa soil. However, S increased yield of desmodium grown in the Paaloa soil. Lime increased yield of desmodium grown in Wahiawa and Paaloa soils and it also increased yield of centrosema grown in the Wahiawa soil.

Molybdenum and lime did not affect nodule fresh and dry weights of desmodium grown in the Wahiawa soil which has a pH of 5.2. Molybdenum also did not affect nodule number and nodule fresh weight of centrosema grown in the Wahiawa soil. Lime increased nodule number, nodule fresh and dry weights of centrosema grown in the Wahiawa soil. It markedly enhanced nodulation of desmodium grown in the Paaloa soil which has a pH of 4.7. Desmodium grown in the unlimed Paaloa soil did not nodulate, indicating that lime is one of the most critical factor influencing nodulation of desmodium grown in this soil. Nevertheless, desmodium grown in the Wahiawa soil which has a pH of 4.7 had formed nodules although lime was not applied to the soil. The difference in nodulation of desmodium grown in the two soils which have the same pH reflected the influences of other chemical properties of the two soils. The Wahiawa soil contains high Mn whereas the Paaloa soil contains high Al. Aluminum toxicity in the Paaloa soil severely affected nodulation more than Mn toxicity in the Wahiawa soil. The fact that root growth of plant that showed Al toxicity symptom was restricted was likely to be one of the primary reasons that the nodulation of plant was limited. *Desmodium intortum* appeared to be more sensitive to Al toxicity than to Mn toxicity.

confusing reword

Sulfur did not affect nodulation of desmodium and centrosema grown in the Wahiawa soil. It tended to alleviate the detrimental effect of seed-applied Mo to rhizobia. The nodulation of no do desmodium grown in the limed Paaloa soil and receiving seed-applied Mo with S was greater than that of desmodium grown in the comparable treatment without S.

?

Molybdenum, S and lime did not significantly increase nitrogenase activity of desmodium grown in the Wahiawa soil. However, nitrogenase activity tended to be higher when Mo was applied, either by seed treatment or by soil application. Molybdenum fertilizer also tended to increase nodule efficiency of desmodium grown in the Wahiawa soil. Molybdenum and lime increased nitrogenase activity of centrosema grown in the Wahiawa soil but S had no effect on nitrogenase activity.

Molybdenum fertilizer increased N concentration in the tops of desmodium and centrosema grown in the Wahiawa soil but it did not increase N concentration in the top of desmodium grown in the Paaloa soil. Sulfur and lime did not increase N concentration in the top of desmodium grown in the Paaloa soil.

Molybdenum fertilizer increased Mo concentration in nodule and in plant top of legumes grown in the Wahiawa and Paaloa soils. Dry matter yield of legumes increased as Mo concentrations increased. Molybdenum concentration in nodule was the best correlated index ($r = 0.770^{**}$) to relative yield as compared to Mo concentration in plant top and extractable Mo in soil. Critical Mo concentrations in nodule associated with 80% and 95% maximum yields of desmodium grown in the Wahiawa soil were 2.20-3.50 and 7.85 ppm, respectively. Comparable critical Mo concentrations of desmodium grown in the limed Paaloa soil (pH 5.2) were 4.0 and 13.2 ppm, respectively.

Although Mo concentration in plant top did not correlate very well with yield as compared to Mo concentration in nodule, Mo concentration in plant top can still be used to evaluate plant response to Mo fertilizer. Critical Mo concentrations in plant top associated with 80% and 95% maximum yields of desmodium grown in the Wahiawa soil were 0.13-0.20 and 0.22-0.30 ppm, respectively. Comparable critical Mo concentrations of desmodium grown in the limed Paaloa soil (pH 5.2) were 0.4 and 1.3 ppm, respectively. Molybdenum concentration in the top of centrosema grown in the Wahiawa soil must be at least 0.2 ppm to be associated with 80% maximum yield.

Sulfur fertilizer did not significantly affect Mo concentration of desmodium and centrosema. Lime did not increase Mo concentration in the top of desmodium grown in the Wahiawa soil but increased Mo concentration in that of desmodium grown in the Paaloa soil. Lime also increased Mo concentration in the top of centrosema grown in the Wahiawa soil in a greenhouse experiment but it did not increase Mo concentration in that of centrosema grown in the same soil in a field experiment. Molybdenum concentrations in the tops of desmodium and centrosema grown in the Wahiawa soil in the field experiment tended to be lower than those in the tops of legumes grown in the greenhouse experiment. The lower concentrations of Mo in legumes grown in the field, compared to those in the greenhouse, are likely due to differences in soil moisture and root density. Plant absorption of soil Mo increases with increased water availability (Jensen and Lesperance, 1971). Under greenhouse conditions, where soils were watered daily, legumes tended to absorb more Mo than under field conditions. Greater root density of legumes grown in the greenhouse also contributes to higher Mo uptake.

The potential harmful effect of Mo regarding Mo-induced Cu deficiency in legume-fed animals should not occur because the Cu/Mo ratio was well above 3 in this study. This is the ratio under which Mo-induced Cu deficiency usually occurs. The high Cu/Mo ratio probably occurred because ~~was due to the finding that~~ Mo concentration in plant top was not exceptionally high and Cu concentration was in a sufficiency range.

Molybdenum fertilizer and lime decreased the concentrations of P, Mg, Mn, and Si in the tops of desmodium and centrosema. Molybdenum fertilizer also decreased the concentrations of Ca and Al but had no effect on K, Fe, and Zn concentrations. Lime increased Ca concentration but tended to decrease K concentration. Sulfur had no effect on nutrient concentrations in plant top.

Not only Mo concentration in plant but also Mo concentration in soil can be used to evaluate plant response to Mo. Dry matter yield of legumes increased as the available Mo in soil increased. The soil-available Mo associated with 95% maximum yield of desmodium grown in Wahiawa and Paaloa soils were 0.09 and 0.10 ug Mo/ml, respectively. These findings indicate that the external Mo requirement of desmodium grown in soils high in kaolinite and oxides of Fe and Al (Oxisols and Ultisols) is 0.09-0.10 ug/ml. The results of the Mo adsorption study suggest that Mo adsorption curves can be used as a basis for Mo fertilization. Available Mo (acid ammonium oxalate-extractable Mo) is likely to be used to evaluate Mo status of soil regarding the response of plant grown in soil high in kaolinite and oxides of Fe and Al elsewhere.

Mycorrhizae enhanced plant growth and increased yield of desmodium, centrosema and stylosanthes. They strongly stimulated nodulation of legumes. Nonmycorrhizal plants did not nodulate, whereas mycorrhizal plants nodulated very well, indicating the dependence of these legumes on mycorrhizal association. Mycorrhizal did not increase Mo concentration in plant top so it can be surmised that they did not enhance Mo absorption. Seed-applied Mo at 0.3 kg Mo/ha increased the percentage of mycorrhizal infection, suggesting that there is a relationship between Mo concentration in soil and mycorrhizal infection.

The overall evaluations of the results indicate that _Desmodium intortum_ is more responsive to Mo fertilizer than _Centrosema pubescens_.

The lower response to Mo fertilizer of centrosema is likely due to the ability of this legume to absorb high quantity of Mo from the soil and/or its requirement of Mo is low when compared to desmodium. Moreover, centrosema seed contains more Mo than desmodium seed. The Mo reserves in the seed would supply some Mo to plant needs.

LITERATURE CITED

Adam, F. 1981. Alleviating chemical toxicities: Liming acid soil. p. 270-301. In G. F. Arkin and H. M. Tayler (ed.) Modifying the root environment to reduce crop stress. ASAE Monograph no. 4. Am. Soc. Agric. Eng.

Alary, J., P. Bourbon, J. Esclassan, J. C. Lepert, and J. Vandaele. 1981. Environmental molybdenum levels in industrial molybdenosis of grazing cattle. Sci. Total Environ. 19:111-119.

Allaway, W. H. 1968. Agronomic controls over the environmental cycling of trace elements. Adv. Agron. 20:235-274.

Allaway, B. J. 1973. Copper and molybdenum in swayback pastures. J. Agric. Sci., Camb. 80:521-524.

Andrew, C. S. 1977. The effects of sulphur on the growth, sulphur and nitrogen concentrations and critical sulphur concentrations of some tropical and temperate pasture legumes. Aust. J. Agric. Res. 28:807-820.

Andrew, C. S., and M. F. Robins. 1969a. The effect of phosphorus on the growth and chemical composition of some tropical and temperate pasture legumes. 1. Growth and critical percentages of phosphorus. Aust. J. Agric. Res. 20:665-674.

Andrew, C. S., and M. F. Robins. 1969b. The effect of potassium on the growth and chemical composition of some tropical and temperate pasture legumes. 2. Growth and critical percentages of potassium. Aust. J. Agric. Res. 20:999-1007.

Anthony, J. L. 1967. Fertilizing soybean in the hill section of Mississippi. Miss. Agric. Exp. Stn. Bull. 743.

Asimi, S., V. Gianinazzi-Pearson, and S. Gianninazzi. 1980. Influence of increasing soil phosphorus levels on interactions between vesicular-arbuscular mycorrhizae and Rhizobium in soybeans. Can. J. Bot. 58:2200-2205.

Barber, S. A., E. H. Halstead, and R. F. Follet. 1966. Significant mechanisms controlling the movement of manganese and molybdenum to plant roots growing in soil. Int. Soc. Soil Sci., Trans. Comm. II&IV:299-304.

Barea, J. M., and C. Azcon-Aguilar. 1983. Mycorrhizas and their significance in nodulating nitrogen-fixing plants. Adv. Agron. 36:1-54.

Barrow, N. J. 1970. Comparison of the adsorption of molybdate, sulfate, and phosphate by soils. Soil Sci. 109:282-288.

Barrow, N. J. 1972. Influence of solution concentration of calcium on the adsorption of phosphate, sulfate, and molybdate by soils. Soli-Sci. 113:175-179.

Barrow, N. J. 1973. On the displacement of adsorbed anions from soil. 1. Displacement of molybdates by phosphate and by hydrate. Soil Sci. 116:423-431.

Barrow, N. J. 1977. Factors affecting the molybdenum status of soils. p. 583-596. In W. R. Chappel and K. K. Petersen (ed.) Molybdenum in the environment. Vol. 2. Marcel Dekker Inc., New york.

Barrow, N. J., and T. C. Shaw. 1975. The slow reactions between soil and anions: 4. Effect of time and temperature of contact between soil and molybdate on the uptake of molybdenum by plants and on the molybdate concentration in the soil solution. Soil Sci. 119:301-310.

Barrow, N. J., and K. Spencer. 1971. Factors in the molybdenum and phosphorus status of soils on the Dorrigo Plateau of New South Wales. Aust. J. Exp. Anim. Husb. 11:670-676.

Biermann, B., and R. G. Lindermann. 1981. Quantifying vesicular-arbuscular mycorrhizae: A proposed method towards standardization. New Phytol. 87:63-67.

Bogdan, 1977. Tropical pasture and fodder plants. Longmans, London and New York. 475 p.

Boswell, F. C. 1980. Factors affecting the response of soybeans to molybdenum application. p. 417-432. In F. T. Corbin (ed.) World soybean research conference II: Proceedings. Westview Press. Boulder, Colorado.

Bowden, J. W., A. M. Posner, and J. P. Quirk. 1977. Ionic adsorption on variable charge mineral surfaces. Theoretical-charge development and titration curves. Aust. J. Soil Res. 15:121-136.

Burris, R. H., and R. V. Hageman. 1980. Electron partitioning from dinitrogenase to substrate and the kinetics of ATP utilization. p. 23-37. In W. E. Newton and S. Otsuka (ed.) Molybdenum chemistry of biological significance. Plenum Press, New York and London.

Burton, J. C., and R. L. Curley. 1966. Compatibility of Rhizobium japonicum and sodium molybdate when combined in a peat carrier medium. Agron. J. 58:327-329.

Carling, D. E., W. G. Riehle, M. F. Brown, and D. R. Johnson. 1978. Effects of vesicular-arbuscular mycorrhizal fungus on nitrate reductase and nitrogenase activities in nodulating and nonnodulating soybeans. Phytopathology 68:1590-1596.

Cooper, K. M., and P. B. Tinker. 1978. Translocation and transfer of nutrients in vesicular-arbuscular mycorrhizas. II. Uptake and translocation of phosphorus, zinc, and sulphur. New Phytol. 81:43-52.

Crush, J. R. 1974. Plant growth responses to vesicular-arbuscular mycorrhiza. VII. Growth and nodulation of some herbage legumes. New Phytol. 73:743-749.

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

Dawson, M. D., and H. S. Bhella. 1972. Subterrean clover (*Trifolium subterraneum* L.) yield and nutrient content as influenced by soil molybdenum status. Agron. J. 64:308-311.

deMooy, C. J. 1970. Molybdenum response of soybeans (*Glycine max*(L.) Merr.) in Iowa. Agron J. 62:195-197.

Donald, C. M., and D. Spencer. 1952. The control of molybdenum deficiency in subterranean clover by pre-soaking the seed with sodium molybdate solution. Aust. J. Agric. Res. 2:295-301.

Eady, R. R., S. Imam, D. J. Lowe, R. W. Miller, B. E. Smith, and R. N. F. Thorneley. 1980. The molecular enzymatology of nitrogenase. p. 19-35. *In* D. P. Stewart and J. R. Gallon (ed.) Nitrogen fixation. Academic Press, London.

Edmeades, D. C., C. E. Smart, and D. M. Wheeler. 1983. Effects of lime on the chemical composition of ryeglass and white clover grown on a yellow-brown loam. N.Z. J. Agric. Res. 26:473-481.

Elkins, D. M., and L. E. Ensminger. 1971. Effect of soil pH on the availability of adsorbed sulfate. Soil Sci. Soc. Am. Proc. 35:931-934.

Fox, R. L. 1981. External phosphorus requirements of crops. *In* Chemistry in the soil environment. ASA Spec. Pub. no. 40:223-239. Am. Soc. of Agron., Madison, Wis.

Franco, A. A., and D. N. Munns. 1981. Response of *Phaseolus vulgaris* to molybdenum under acid condition. Soil Sci. Soc. Am. J. 45:1144-1148.

Fried, M. 1948. The absorption of sulfur dioxide by plants as shown by the use of radioactive sulfur. Soil Sci. Soc. Am. Proc. 13:135-138.

Gibson, A. H. 1976. Limitations to dinitrogen fixation in legumes. p. 400-428. *In* W. E. Newton and C. J. Nyman (ed.) Proceeding of the 1 st international symposium on nitrogen fixation. Vol. II. Washington Stata University Press, Pullman, Washington.

Giddens, J. 1964. Effect of adding molybdenum compounds to soybean inoculant. Agron. J. 56:362-363.

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

Giddens, J., and H. F. Perkins. 1972. Essentiality of molybdenum for alfalfa on highly oxidized Piedmont soils. Agron. J. 64:819-820.

Gile, C. H., T. H. MacEwan, S. N. Nakhwa, and D. Smith. 1960. Studies in adsorption. Part XI. A system of classification of solution adsorption isotherms and its use in diagnosis of adsorption mechanisms and in measurement of specific surface area of soils. J. Chem. Soc. 3973-3993.

Gladstone, J. S., J. F. Loneragan, and N. A. Goodchild. 1977. Fixed responses to cobalt and molybdenum by different legume species, with influences on the role of cobalt in legume growth. Aust. J. Agric. Res. 28:619-628.

Golov, V. I., and Y. N. Kazakhov. 1973. Uptake of molybdenum by soybean and its residual effect when applied to soils in the far east. Soviet Soil Sci. 5:551-558.

Gonzalez, R., B. H. Appelt, E. B. Schalscha, and F. T. Bingham. 1974. Molybdate adsorption characteristics of volcanic-ash-derived soils in Chile. Soil Sci. Soc. Am. Proc. 38:903-906.

Gorlach, E., K. Gorlach, and A. Compala. 1969. The effect of phosphorus on the sorption and desorption of molybdates in the soil. Agrochimica 13:506-512.

Gray, L. E., and J. W. Gerdemann. 1973. Uptake of sulphur-35 by vesicular-arbuscular mycorrhizae. Plant Soil 39:687-689.

Grigg. J. L. 1960. The distribution of molybdenum in the soils of New Zealand. I. Soils of the north island. N.Z. J. Agric. Res. 3:69-86.

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

Gupta, U. C. 1979. Effect of methods of application and residual effect of molybdenum on the molybdenum concentration and yield of forages on podzol soils. Can. J. Soil Sci. 59:183-189.

Gupta, U. C., and D. C. MacKay. 1966. The relation of soil properties to exchangeable and water-soluble copper and molybdenum status in podzol soils of eastern Canada. Soil Sci. Soc. Am. Proc. 30:373-375.

Gupta, U. C., and L. B. MacLeod. 1975. Effects of sulfur and molybdenum on the molybdenum, copper and sulfur concentrations of forage crops. Soil Sci. 119:441-447.

Gupta, U. C., F. W. Calder, and L. B. MacLeod. 1971. Influence of added limestone and fertilizers upon the micronutrient content of forage tissue and soil. Plant Soil 35:249-256.

Hageman, R. V., and R. H. Burris. 1978a. Nitrogenase and nitrogenase reductase associate and dissociate with each catalytic cycle. Proc. Natl. Acad. Sci. U. S. A. 75:2699-2702.

Hageman, R. V., and R. H. Burris. 1978b. Kinetic studies on electron transfer and interaction between nitrogenase components from Azotobacter vinelandii. Biochemistry 17:4117-4124.

Hageman, R. V., and R. H. Burris. 1980. Electron allocation to alternative substrate of Azotobacter nitrogenase is controlled by the electron flux through dinitrogenase. Biochim. Biophys. Acta 591:63-75.

Hagstrom, G. R., and K. C. Berger. 1963. Molybdenum status of three Wisconsin soils and its effect on four legume crops. Agron. J. 55:397-399.

Hardy, R. W. F., R. D. Holstein, E. K. Jackson, and R. C. Burns. 1968. The acetylene-ethylene assay for N_2 fixation: Laboratory and field evaluation. Plant Physiol. 43:1185-1207.

Harris, H. B., M. B. Parker, and D. V. Phillips. 1971. Effect of seed treatment, method of application, and molybdenum content on emergence and yield of soybeans. Ga. Exp. Stn. Res. Rpt. 113.

Hawes, R. L., J. L. Sims, and K. L. Wells. 1976. Molybdenum concentration of certain crop species as influenced by previous application of molybdenum fertilizer. Agron. J. 68:217-218.

Hayman, D. S., and J. M. Day. 1978. Influence of VA mycorrhiza on plant growth. Uptake of molybdenum. Rothamsted Annu. Rep. for 1977. Rothamsted Exp. Stn., Harpenden, England p. 240.

Hayward, D. O., and B. M. W. Trapnell. 1964. Chemisorption. 2 nd Ed. Butterworths, London.

Higston, F. J., R. J. Atkinson, A. M. Posner, and J. P. Quirk. 1967. Specific adsorption of anions. Nature 215:1459-1461.

Hingston, F. J., A. M. Posner, R. J. Atkinson, and J. P. Quirk. 1968. Specific adsorption of anions on goethite. Intl. Congr. Soil Sci., Trans 9th (Adelaide, Australia) 1:669-678.

Hingston, F. J., A. M. Posner, and J. P. Quirk. 1972. Anion adsorption by goethite and gibbsite. 1. The role of proton in determining adsorption envelopes. J. Soil Sci. 23:177-192.

Ishizuka, J. 1982. Characteristics of molybdenum absorption and translocation in soybean plants. Soil Sci. Plant Nutr. 28 (1):63-77.

James, D. W., T. L. Jackson, and M. E. Harward. 1968. Effect of molybdenum and lime on the growth and molybdenum content of alfalfa grown on acid soil. Soil Sci. 105:397-402.

Jarrel, W. M., and M. D. Dawson. 1978. Sorption and availability of molybdenum in soils of western Oregon. Soil Sci. Soc. Am. J. 42:412-415.

Jensen, H. L. 1946. The nitrogen-fixing activity of legume root nodules. Aust. J. Sci. 9:118.

Jensen, E. H., and A. L. Lesperance. 1971. Molybdenum accumulation by forage plants. Agron. J. 63:201-204.

Joham, H. E. 1953. Accumulation and distribution of molybdenum in the cotton plant. J. Plant Physiol. 28:275-280.

Johansen, C., and P. C. Kerridge. 1981. Nitrogen concentration ranges in tropical pasture legumes responding to molybdenum. Trop. Grassland 15:107-111.

Johansen, C., P. C. Kerridge, P. E. Luck, B. G. Cook, K. F. Lowe and H. Ostrowski. 1977. The residual effect of molybdenum fertilizer on growth of tropical pasture legumes in a subtropical environment. Aust. J. Exp. Agric. Anim. Husb. 17:961-968.

Jones, L. H. P. 1956. Interaction of molybdenum and iron in soils. Science 123:1116.

Jones, L. H. P. 1957. The solubility of molybdenum in simplified systems and aquqous soil suspension. J. Soil suspension. J. Soil Sci. 8:313-327.

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

Kamprath, F, J. 1971. Potential detrimental effects from liming highly weathered soils to neutrality. Soil Crop Sci. Soc. Fla. Proc. 31:200-203.

Kanehiro, Y., J. L. Walker, and M. Asghar. 1983. Edaphic factors. p. 197-219. In R. L. Burt, P. P. Rotar, J. L. Walker, and M. W. Silvey (ed.) The role of Centrosema, Desmodium, and Stylosanthes in improving tropical pastures. Westview Press, Boulder, Colorado.

Kannan, S., and S. Ramani. 1978. Studies on molybdenum absorption and transport in bean and rice. Plant Physiol. 62:179-181.

Karimian, N., and F. R. Cox. 1978. Adsorption and extractability of molybdenum in relation to some chemical properties of soil. Soil Sci. Soc. Am. J. 42:757-760.

Kerridge, P. C., B. G. Cook, and M. L. Everett. 1973. Application of molybdenum trioxide in the seed pellet for sub-tropical pasture legumes. Trop. Grassland 7:229-332.

Kormanik, P. P, and C. C. McGraw. 1982. Quantification of vesicular-arbuscular mycorrhizae in plant roots. p. 37-46. In N. C. Schenck (ed.) Methods and principles of mycorrhizal research. The American Phytopathological Society. St. Paul, Minnesota.

Kubota, J., and W. H. Allaway. 1972. Geographic distribution of trace element problems. p.525-554. In J. J. Mortvedt, P. M. Giordano and W. L. Lindsay (ed.) Micronutrients in Agriculture. Soil Sci. Soc. Am., Madison, Wisconsin.

Kucey, R. M. N., and E. A. Paul. 1982. Carbon flow, photosynthesis, and N_2 fixation in mycorrhizal and nodulated faba beans (Vicia faba L.). Soil Biol. Biochem. 14:407-412.

Lambert, D. H., D. E. Baker, and H. Cole, Jr. 1979. The role of mycorrhizae in the interactions of phosphorus with zinc, copper, and other elements. Soil Sci. Soc. Am. J. 43:976-980.

Lavy, T. L., and S. A. Barber. 1963. A relationship between the yield response of soybean to molybdenum applications and the molybdenum content of the seed produced. Agron. J. 55:154-155.

Lee, T. E., and M. W. Loutit. 1977. Effect of extracellular polysaccharides of rhizosphere bacteria on the concentration of molybdenum in plants. Soil Biol. Biochem. 9:411-415.

Ljones, T., and R. H. Burris. 1978. Evidence for one-electron transfer by the Fe protein of nitrogenase. Biochim. Biophys. Res. Commun. 80:22-55.

Levesque, M., and M. Schnitzer. 1967. Organo-metallic interactions in soils: 6. Preparation and properties of fulvic acid-metal phosphates. Soil Sci. 110:109.

Lewis, A. H. 1943. The teart pastures of Somerset: 3. Reducing the teartness of pasture herbage. J. Agric. Sci. 33:58-63.

Little, I. P., and P. C. Kerridge. 1978. A laboratory assessment of the molybdenum status of nine Queensland soils. Soil Sci. 125:102-106.

Maynard, D. G.,J. W. B. Stewart, and J. R. Bettany. 1983. Use of plant analysis to predict sulfur deficiency in rapeseed (Brassica napus and B. campestris). Can. J. Soil Sci. 63:387-396.

Miltmore, J. E., and J. L. Mason. 1971. Copper to molybdenum ratio and molybdenum and copper concentrations in ruminant feed. Can. J. Anim. Sci. 51:193-200.

Misra, S. G., K. C. Mishra, and P. C. Mishra. 1977. Retention and release of molybdenum by soils. p. 597-618. In W. R. Chappell and K. K. Petersen (ed.) Molybdenum in the environment. Vol. 2. Marcel Dekker, Inc, New York.

Mortenson, L. E., and R. N. F. Thorneley. 1979. Structure and function of nitrogenase. Ann. Rev. Biochem. 48:387-418.

Mortvedt, J. J., and O. E. Anderson (ed.). 1982. Forage legumes: Diagnosis and correction of molybdenum and manganese problems. Southern Coop. Ser. Bull. 278.

Mosse, B. 1973. Advances in the study of vesicular-arbuscular mycorrhiza. Ann. Rev. Phytopathol. 11:171-195.

Mosse, B., C. L. Powell, and D. S. Hayman. 1976. Plant growth responses to vesicular-arbuscular mycorrhiza. IX. Interactions between VA mycorrhiza, rock phosphate, and symbiotic N_2 fixation. New Phytol. 76:331-342.

Munns, D. N., and R. L. Fox. 1976. Depression of legume growth by liming. Plant Soil 45:701-705.

Munns, D. M., and B. Mosse. 1980. Mineral nutrition of legume crops. p. 115-125. In R. J. Summerfield and A. H. Bunting (ed.) Advances in legume science. Royal Botanic Garden, Kew, London.

Oades, J. M. 1963. The distribution of iron compounds in soils. Soil Fert. 26:69-80.

Ostrowski, H., A. Diatloff, and H. R. Williams. 1978. Molybdenum responses of Macroptilium atropurpureum cv. siratro on a red volcanic soil in coastal south-east Queensland. Trop. Grassland 12:75-79.

Parker, M. B., and H. B. Harris. 1962. Soybean response to molybdenum and lime and the relationship between yield and chemical composition. Agron. J. 54:480-483.

Peterson, N. K., and E. R. Purvis. 1961. Development of molybdenum deficiency symptoms in certain crop plants. Soil Sci. Soc. Am. Proc. 25:111-117.

Petrie, S. E., and T. L. Jackson. 1982. Effects of lime, P, and Mo application on Mo concentration in subclover. Agron. J. 74:1077-1081.

Powell, C. L., and J. Daniel. 1978. Mycorrhizal fungi stimulate uptake of soluble and insoluble phosphate fertilizer from a phosphate-deficient soil. New Phytol. 80:351-359.

Purvis, E. R., and N. K. Peterson. 1956. Methods of soil and plant analyses for molybdenum. Soil Sci. 95:223-228.

Reddy, G. D., A. M. Alston, and K. G. Tiller. 1981. Effects of fertilizer on concentrations of copper, molybdenum, and sulfur in subterranean clover (Trifolium subterraneum). Aust. J. Exp. Agric. Anim. Husb. 21:491-497.

Redente, E. F., and E. B. Reeves. 1981. Interactions between vesicular-arbuscular mycorrhiza and Rhizobium and their effect on sweetvetch growth. Soil Sci. 132:410-415.

Reisenauer, H. M. 1963. Relative efficiency of seed- and-soil-applied molybdenum fertilizer. Agron. J. 55:459-460.

Reisenauer, H. M. 1965. Molybdenum. p. 1050-1058. In C. A. Black (ed.) Method of soil analysis. Part 2. Chemical and microbiological properties. American Society of Agronomy, Madison, Wisconsin.

Reisenauer, H. M., A. A. Tabikh, and P. R. Stout. 1962. Molybdenum reactions with soils and the hydrous oxides of iron, aluminum, and titanium. Soil Sci Soc. Am. Proc. 26:23-27.

Reyes, E. D., and J. J. Jurinak. 1967. A mechanism of molybdate adsorption on Fe_2O_3. Soil Sci. Soc. Am. Proc. 31:637-640.

Ross, J. P. 1971. Effect of phosphate fertilization on yield of mycorrhizal and nonmycorrhizal soybeans. Phytopathology 61:1400-1403.

Rossiter, R. C. 1966. Ecology of the mediterranean annual type pasture. Adv. Agron. 18:1-16.

Safir, G. R., J. S. Boyer, and J. W. Gerdemann. 1972. Nutrient status and mycorrhizal enhancement of water transport in soybean. Plant physiol. 49:700-703.

Sandell, E. B. 1959. Colorimetric determination of traces of metals. Ed. 3. Interscience Publishers, New York.

Schrauzer, G. N. 1976. Molybdenum in biological nitrogen fixation. p. 243-265. In W. R. Chappell and K. K. Petersen (ed.) Molybdenum in the environment vol. 1. Dekker, Inc. New York.

Schrauzer, G. N. 1980. Studies of the mechanism of biological nitrogen fixayion with functional model systems. p. 103-119. In J. Chatt, L. M. da Camara Pina, and R. L. Richards (ed.) New trends in the chemistry of nitrogen fixation. Academic Press, London.

Schrauzer, G. N., G. W. Kiefer, K. Tano, and P. A. Doemeny. 1974. The chemical evolution of a nitrogenase model VII. The reduction of nitrogen. J. Amer. Chem. Soc. 96:641-652.

Shah, V. K., and W. J. Brill. 1977. Isolation of an iron molybdenum cofactor from nitrogenase. Proc. Natl. Acad. Sci. U. S. A. 74:3249-3253.

Sherman, G. D., and R. J. Kapteyn. 1966. Re-examination of the molybdenum content of Hawaiian soils. Agron. J. 58:358-359.

Sim, J. L., R. E. Sigafus, and N. Tiaranan. 1974. Effect of lime, inoculant, and molybdenum pelleting of seed on growth and nitrogen content of crownvetch. Agron. J. 66:446-449.

Smith, S. E., and G. D. Bowen. 1979. Soil temperature, mycorrhizal infection and nodulation of Medicago truncatula and Trifolium subterraneum. Soil Biol. Biochem. 11:469-473.

Smith, S. E., and M. J. Daft. 1977. Interactions between growth, phosphate content and nitrogen fixation in mycorrhizal and non-mycorrhizal Medicago sativa. Aust. J. Plant Physiol. 4:403-413.

Smith, B. H., and G. W. Leeper. 1969. The fate of applied molybdate in acidic soils. J. Soil Sci. 20:246-254.

Smith, S. E., D. J. D. Nicholas, and F. A. Smith. 1979. Effect of early mycorrhizal infection on nodulation and nitrogen fixation in Trifolium subterraneum L. Aust. J. Plant Physiol. 6:305-311.

Spencer, K., and J. R. Freney. 1980. Assessing the sulfur status of field-grown wheat by plant analysis. Agron. J. 72:469-472.

Stout, P. R., and W. R. Meagher. 1948. Studies on the molybdenum nutrition of plants with radioactive molybdenum. Science 108:471-473.

Stout, P. R., W. R. Meagher, G. A. Pearson, and C. M. Johnson. 1951. Molybdenum nutrition of crop plants. I. The influence of phosphate and sulfate on the absorption of molybdenum from soils and solution cultures. Plant Soil 3:51-87.

Sultana-Ahmed, M., and L. Rahman. 1982. Evalution of extractants for available Mo in several soils of Bangladesh. Plant Soil 69:287-291.

Suttle, N. F. 1980. The role of thiomolybdates in the nutritional interactions of copper, molybdenum, and sulfur: Fact or fantasy? Ann. New York Acad. Sci. 355:195-207.

Szalay, A., and M. Szilagyi. 1968. Laboratory experiments on the retention of micronutrients by peat humic acids. Plant Soil 29(2):219-224.

Taal, S. 1979. Factors affecting soil molybdenum availability and molybdenum fertilization of tropical pasture legumes. M. S. Thesis. Univ. Hawaii, Manoa.

Theng, B. K. G. 1971. Adsorption of molybdate by some crystalline and amorphous soil clays. N.Z. J. Sci. 14:1040-1056.

Underwood, E. J. 1971. Copper. Trace elements in human and animal nutrition. Ed. 3. Academic Press, New York and London.

Underwood, E.J. 1976. Molybdenum in animal nutrition. p. 9-31. In C. W. K. Chappell and K. K. Peterson (ed.) Molybdenum in the environment. Dekker, New York.

Underwood, E. J. 1977. Trace elements in human and animal nutrition. Ed. 4. Academic Press, New York and London.

Vlek, P. L. G., and W. L. Lindsay. 1977. Thermodynamic stability of molybdenum minerals in soils. Soil Sci. Soc. Am. J. 41:42-46.

Vlek, P. L. G., Th. J. M. Blom, J. Beck, and W. L. Lindsay. 1974. Determination of the solubility product of various iron hydroxides and jarosite by chelation method. Soil Sci. Soc. Am. Proc. 38:429-432.

Whitehead, D. C. 1966. Nutrient minerals in grassland herbage. Commonwealth Agricultural Bureaux. Hurley, Birkshire. Publication no. 1/1966.

Widdowson, J. P., and T. W. Walker. 1971. Effect of lime and molybdenum on the yield and composition of white clover in a sequence of New Zealand zonal soils. N.Z. J. Agric. Res. 14:801-820.

Winter, H. C., and R. H. Burris. 1968. Stoichiometry of the adenosine triphosphate requirement for N_2 fixation and H_2 evolution by a partially purified preparation of _Clostridium_ _pasteurianum_. J. Biol. Chem. 243:940-944.

Wynne, K. N., and G. L. McClymont. 1955. Copper-molybdenum-sulphate interaction induction of hypercuprosis. Nature 175:471-472.

Younge, O. R., and M. Takahashi. 1953. Response of alfalfa to molybdenum in Hawaii. Agron. J. 45:420-428.

Yost, R. S., and R. L. Fox. 1979. Contribution of mycorrhizae to P nutrition of crops growing on Oxisol. Agron. J. 71:903-908.

Yost, R. S., and R. L. Fox. 1982. Influence of mycorrhizae on the mineral contents of cowpea and soybean grown in an Oxisol. Agron. J. 74:475-481.