

INFLUENCE OF FERTILIZATION ON NUTRIENT STATUS AND SIZE  
OF  
BARE-ROOT PINUS TAEDA L. SEEDLINGS

A Thesis

by

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May 1994

Major Subject : Forestry

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OF BARE-ROOT PINUS TAEDA L. SEEDLINGS

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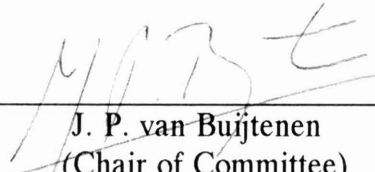
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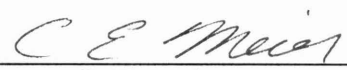
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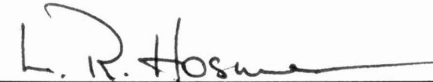
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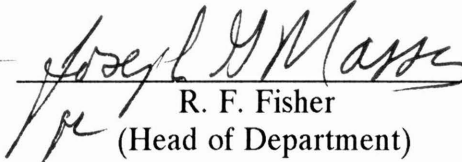
MASTER OF SCIENCE

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## ABSTRACT

Influence of Fertilization on Nutrient Status and Size

of Bare-root Pinus taeda L. Seedlings. (May 1994)

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The purpose of this study was to evaluate the influence of varying types and amounts of fertilizer on loblolly pine (Pinus taeda L.) seedlings. Morphological traits as well as nutritional status were measured. A comparison of the effects of constant versus increasing applications of fertilizer was included in this study. In addition, an effort was made to evaluate the influence of different treatment histories on two separate nursery compartments.

Seedlings grown in a southern forest nursery were fertilized with either pre-plant nitrogen, top-dress nitrogen or top-dress magnesium. Nitrogen was supplied as ammonium nitrate ( $\text{NH}_4\text{NO}_3$ ) and magnesium as magnesium sulfate ( $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ ). Seedlings were evaluated after ten weeks for shoot length, root length, and stem diameter, and again after one growing season for the aforementioned plus tissue concentrations of N, P, K, Ca, and Mg.

Ten week analysis showed no statistically significant results, perhaps due to lack of tissue differentiation over the short growing period. Sampling performed after one growing season showed that pre-plant applications of N had no effect on the seedlings except for mean needle:stem weight.

Top-dress applications of Mg had no significant effect on seedling morphology, but did significantly increase Mg and N foliage concentrations. Top-dress N application in compartment 4 resulted in higher needle to stem and needle to root weight, and higher N concentrations in foliage and stem tissue. Increasing applications of N resulted in significantly higher K stem and Mg root concentrations. Applications of N in compartment 7a increased mean needle weight, N concentrations, P foliar concentrations, and K stem and root concentrations. Application of N at an increasing rate significantly increased concentrations of N, Mg, and K.

Comparison between top-dress applications of N in compartment 4 and 7a showed that 7a seedlings had higher stem caliper, shoot length, stem weight, and needle weight.

Lower root measures in compartment 7a were probably due to heavier soil that caused loss of roots upon lifting. As a result, needle:root and shoot:root ratios were increased. Tissue concentrations of N and P were significantly higher for compartment 4 seedlings, whereas K and Mg concentrations were higher for compartment 7a.

The results of this study indicate that production of quality seedlings can be enhanced through minor changes in nursery practices that are currently already in place. Manipulation of seedlings by fine-tuning fertility levels can, in combination with existing moisture management, result in optimum seedling growth.

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## CHAPTER I

### INTRODUCTION

Forested lands are one of Texas' greatest natural resources. Approximately 23 million acres in Texas are forested. These forests provide a range of benefits to the people of Texas. In East Texas, forests provide not only wood products, but offer opportunities for employment, investment, and recreation (O'Laughlin and Williams 1989).

A key to maintaining this resource is the reforestation of harvested lands. Ninety-nine percent of forested land in Texas is planted with bare-root pine seedlings (Lord 1993). Because they may rely on the economics of plant production, both private landowners and public landholders can insure their continued success through carefully designed reforestation programs.

The goal of such reforestation programs is to realistically maximize the survival and growth of outplanted seedlings. Achieving this goal depends upon many factors including: seedling quality (i.e. seedling morphology and internal physiology), outplanting techniques, site conditions, and silvicultural practices following planting. Reforestation success is dependent to a great extent on quality of the seedlings. "The quality of planting stock is the degree to which that stock realizes the objectives of management (to the end of the rotation or achievement of specified sought benefits) at minimum cost. Quality is fitness for purpose" (Willen and Sutton 1980). That is fitness for planting in the forest. It has been pointed out that outplanting performance depends not only on seedling morphological traits that are evident through appearance, but also on preconditioning and the resultant physiological state of the seedling (Chavasse 1977). In order for seedlings to survive stress, they must be able to physiologically adapt to or tolerate their environment.

Morphology can give an indication of the condition of the seedlings and is normally the first consideration employed because of its proven value and ease of measurement. Assurance of the highest quality planting stock begins with the sowing of genetically superior seed. Subsequent growth of the seedlings must meet height standards established such that the seedling is not too small to survive, <13cm (5 in.) nor too large and difficult to

plant, >30cm (12 in.) (May 1984). In addition, physiological aspects such as cold hardiness, nutrient status and water uptake have been linked to seedling survival and growth (Alden and Hermann 1971). Because these traits are not readily recognized in the field, they require additional analysis.

Cultural practices in pine seedling nurseries prepare the seedlings for survival after planting. Nursery practices can be altered to manipulate seedling morphology and physiology, which subsequently influence survival. The reliance on seedling quality to enhance survival and growth is magnified in cases of extreme site conditions. It has been shown that such practices as fertilization, irrigation, and undercutting can alter the physiological state of nursery seedlings and consequently affect their outplanting success (van den Driessche 1980).

Of the important physiological attributes previously mentioned, this thesis addresses fertilization and its effect on morphology and nutrient status.

## CHAPTER II

### LITERATURE REVIEW

#### INTRODUCTION

Growing quality planting stock is the major focus of forest nurseries. But determining what is meant by quality has led numerous investigators to form their own definition. van den Driessche (1980) proposed to define quality based on three separate criteria. First, a seedling must survive transplantation. Second, because the seedling cannot go into shock, rapid growth should follow. Finally, the stock should be suitable for the planting method employed. More specific physical parameters such as shoot:root ratio, seedling size, or root collar diameter have been identified because they relate to these criteria. From the above goals, the concept of a "target" seedling has become the subject of serious research. The idea incorporates morphological and physiological traits that can be linked to reforestation success (Mexal and Landis 1990; Rose et al. 1990; Brissette and Carlson 1991; Omi 1991). As a result, now it is possible to more readily predict outplanted seedling performance based on those seedling traits. The target seedling concept recognizes that numerous traits are simultaneously involved in producing the desired response. In addition to morphological traits set up as culling standards, nursery personnel target certain physiological traits, such as plant moisture status or frost hardiness, as desirable traits to produce. Cultural practices are then governed by regimes needed to produce these target traits and seedlings specifically tailored to achieve the intended results for given sites.

#### MORPHOLOGICAL TRAITS

Desired morphology has been defined on the basis of a seedling's ability to meet certain goals. For example, the stem should be long enough (but not too long) to allow machine planting and should be strong (diameter) to withstand rain, ice and wind. The roots should reach or have the potential to grow below the root zone of competing grasses. In addition, the seedling should not be so large that the size or weight makes field handling difficult. It has been shown for example, that balanced, medium sized seedlings (17-33 cm stem height) with sturdy stems and well-developed, fibrous root systems will survive better and

have better initial growth than smaller or larger seedlings (Scarborough and Allen 1954, Wakeley 1954, Shoulders 1960, Silker 1960, Swearingen 1963, Hermann 1964, Meekins 1964, Hunt and Gilmore 1967, Carmean 1971, O'Gwynn 1972, Blair and Cech 1974, Williston 1974).

## SEEDLING GRADE

Morphology of planting material is generally recognized as important since specific parameters (e.g., shoot:root ratio) can affect field performance. Such a relationship can be seen in the studies of slash pine (*Pinus elliottii* Engelm. var. *elliottii*) and loblolly pine (*Pinus taeda* L.), which have shown that seedling grade greatly affects survival, height, and volume production (Blair and Cech 1974; South et al. 1985).

Seedling grade was traditionally a means of characterizing the capacity of the seedling to survive and grow well after outplanting (Blair and Cech 1974). In 1954, P.C. Wakeley established a grading system based on observable and measurable seedling traits (Table 1).

Table 1.  
Morphological grades<sup>1</sup> of uninjured<sup>2</sup> loblolly pine (Wakeley 1954).

Grade	Stem length	Stem diameter	Nature of stem	Bark on stem	Needles	Winter buds
1	5-12 in.	$\geq 3/16$ in.	stiff, woody	Usually on entire stem	Mostly 3's and 2's	Usually present
2	4-7 maybe 10	at least 1/8 in.	Moderately stiff	Lower part, often all over	Part at least 3's	Occasionally present
3	Usually <5		Weak, juicy	Often lacking	Mostly single, bluish	Usually absent

<sup>1</sup> Grades 1 and 2 usually considered plantable, and grade 3 culled.

<sup>2</sup> Any seedling with roots less than 5 inches long should be considered as grade 3, regardless of the quality of the tops.

Wakeley (1969) found that Grade 1 stock consistently outperformed Grade 3 stock in survival, mean total height, mean diameter breast height, percent of dominant plus codominant trees per acre, rough pulpwood per acre and sawtimber per acre. A 1985 study by South et al. found significant differences among these seedling grades for survival, height and volume production. Numerous additional studies have found a positive correlation between morphological grade and survival (Blair and Cech 1974, Burns and Brendemuehl 1971, Switzer and Nelson 1967). Correlations between shoot height, stem diameter, root volume, and shoot:root ratio and subsequent growth after 5 years in the field have been positive (Duryea 1984).

Although it is still considered the standard in the South today, it has been necessary to modify the Wakeley grading rules because of improved seedling quality brought about through reduced seedbed densities, improved insect and disease control, enhanced genetic quality, fertilization adjustments, root pruning and topcutting. Nursery cultural practices have led to a more desirable, uniform mix of seedlings. Today, many nurseries in the South do not invest time and money in grading, unless there is a specific need; such as seedlings for hand planting, for droughty sites or for wet sites. Southern pine seedling nurseries now deal more on a plantable versus non-plantable basis (May 1984).

Currently, there are differing opinions about what seedling attributes will insure success after outplanting (Table 2). Williston (1974) states that healthy, unbroken loblolly, slash or shortleaf pine seedlings should be culled if: 1) They lack secondary needles, 2) the root system is less than 5 inches long, or 3) the diameter at the root collar is less than 1/8 inch. Numerous studies have suggested an optimum shoot length be included (Silker 1960, Williston 1974, Wakeley 1969). A twenty year study by Clark and Phares (1961) concluded that shortleaf pine planting stock should be at least six inches tall with a stem caliper of 3/20 inch or more. Currently, the optimum seedling will also have some disease and insect resistance. A comparison of some target seedling attributes can be found in Table 2.

Pawsey (1972), in a study of Monterey pine *Pinus radiata* D. Don, found that survival and development were largely unaffected by grading or culling if size classes were planted separately. Because of this finding, he concluded that seedling size was not sufficiently reliable as a criterion of inherent vigor to warrant an overall rejection of small seedlings. Pawsey determined that plants below a minimum size of approximately five inches were usually inferior to larger ones, in growth rate more than survival.

Poorer grades have been known to outperform Grade 1 in some instances with respect to survival (Williston 1974, Venator 1983). This, it has been suggested, is due to some nurseries producing an internal chemical or physiological condition in the seedlings which

greatly influences their survival. Others attribute this to an interaction between seedling height and planting site. If planted on a droughty site, shorter seedlings may outperform

Table 2.  
Comparison of "Target" bare-root seedling attributes.

Investigator	Shoot Height	Stem Diameter	Root System	Special Attributes
Wakeley <sup>3</sup> (1954)	7.62-25.4cm	>3.18mm	>12.7cm	Stem stiff, woody, bark often allover, winter buds often present
Williston <sup>2</sup> (1974)		>3.18mm	>12.70cm	secondary needles present
Brissette & Carlson <sup>1</sup> (1991)		4.6mm	root vol. 3.1 cm <sup>3</sup>	Ht/Dia. <42.0 mm/mm Overwintering bud present
Clark & Phares <sup>1</sup> (1961)	>15.24cm	>3.81mm		
Barnett <sup>1</sup> (1991)	15-30 cm	1.6-5.0mm	10-20 cm >7 laterals	woody, secondary needles winter buds

1 Shortleaf pine (*Pinus echinata* Mill.)

2 Shortleaf or slash pine

3 Loblolly pine

taller seedlings because of a lower transpirational surface. Venator (1983) found that taller seedlings had higher outplanting mortality on extremely dry sites. On the other hand, taller seedlings may be better at competing with vegetation for water and nutrients. South et al. (1985) suggest a correlation between survival and height/diameter ratio that is lower for sites or years where soil moisture is adequate and higher where moisture is limited. Venator (1983) proposes that benefits of grading may only be recognized during times of stress, indicating that a generalized balance between top and root system may exist but manifestation will only appear during stress.



### Shoot Height and Seedling Mass

Shoot height can affect both survival and subsequent growth of seedlings at outplanting. In a study on Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco), Richter (1971) found a strong correlation between height at planting and height increment during growth. Height increment of the highest classes was more than twice that of the smallest classes. Growth rate of Monterey pine (*Pinus radiata* D. Don) in Australia, during the early years in the field was strongly influenced by initial stock size (Pawsey 1972). In an east Texas study by Hunt and Gilmore (1967), taller seedlings grew significantly faster during all four growing seasons. The larger seedlings survived and grew better on the best site. Silker (1960) noted that slash pine seedlings with larger than 15-23cm top, and diameters exceeding 0.38cm attained greater growth in good years, but did not consistently survive better in dry years.

In many instances, rate of growth during the early years in the field has been strongly influenced by the general overall size of the planting stock. In studies on white spruce, height and root collar diameter have been found to be well correlated with survival and growth ten years after planting (Mullin and Svaton, 1972). Positive correlations have been found between seedling dry weight and early height growth in coastal and interior Douglas-fir, Sitka spruce (*Picea sitchensis* (Bong.) Carr) and lodgepole pine (*Pinus contorta* Dougl.) (van den Driessche 1984). Additionally, survival and seedling dry weight were also correlated in coastal Douglas-fir and Sitka spruce.

In contrast, Chavasse (1977) found that growth of radiata pine and Douglas-fir was unrelated to the initial height of the seedlings over a period of a few years. Above a minimum size, there is also a question about initial seedling height as a predictor of survival (Mexal and Landis 1990). Puttonen (1989) believes this to be proof that seedling height may not be completely genetically controlled and that microenvironment plays a significant role at the planting site. Mullin and Svaton (1972) found that at heights above 20cm, seedling survival did not increase with increasing height. Pawsey (1972) found that survival in the field was practically independent of seedling size when competition from grasses is controlled. Comparisons between size classes showed that large seedlings grew less than other size classes the year of outplanting (Bickelhaupt 1988). It was suggested that these large seedlings were out of balance with large tops and average size root systems. One year later, the large seedlings did not grow any better than the culls and not as well as the medium size class.

### Stem Diameter

Stem diameter has been found to be a valuable measurement of seedling quality. It is generally accepted that seedlings with larger stem diameters have higher initial survival (Mexal and Landis 1990) and better outplanting success (Bunting 1975; Sutton 1979; Schmidt-Vogt 1981), but it must be noted that above a certain size, larger diameter seedlings may actually have a lower survival rate because of the loss of roots during lifting, which will change the shoot:root balance.

In a 1971 study by Anstey, height increment of one year old Monterey pine seedlings was only dependent on root collar diameter. Larger slash and loblolly pine seedlings produced 80% and 240% more volume, respectively, than average size seedlings when compared ten years after planting (South et al. 1985). South et al. (1988) found that average tree volume of 30-year-old loblolly pine was highly correlated with initial seedling diameter. They deduced that seedlings maintained their relative size difference as they grew. Chavasse (1977) suggested that root-collar diameter was a better indicator of seedling quality than shoot height in Monterey pine and Douglas-fir.

In general, stem diameter tends to be well correlated with other size characteristics, such as seedling dry weight, root weight and height (Switzer and Nelson 1963, Mexal and Landis 1990). Ritchie (1984) included the qualification that this only applies if other factors are equal.

### Root System

Root mass has been acknowledged as one of the most important factors critical to field performance (Hermann 1964, Duryea 1984, Ritchie 1984). But size of the root system is more meaningful when placed in relation to other plant parts, such as the stem or growing top. A large shoot requires a root system adequate for supplying water and nutrients. But as Ritchie (1984) points out, root weight or volume is not a very good indicator of the root system's ability to provide water and minerals. He suggested that some measure of root fibrosity or surface area is needed, but would be difficult and costly to determine. A tree seedling is more apt to survive and grow as well as a site permits if it produces new root growth within a few days after outplanting (Sutton 1979). Mortality is certain, if that root growth is not forthcoming. In addition, intimate root-soil contact must be maintained.

In a 1964 study by Hermann, Douglas-fir survival was significantly lower in seedlings with poor root systems compared to those with good root systems. Wakeley (1948)

hypothesized that initial survival and height growth of planted southern pines, on a well-drained soil, depends on an excess of water-intake over water loss. This excess of water-intake can be attributed to the formation of new root tissue promptly after planting (Sutton 1979, Carlson 1986). This new growth is critical because transplanted seedlings have reduced water and nutrient absorbing capacity due to root pruning during lifting, but the surface area available for transpiration remains at the pre-lifting levels (Brissette 1990). Brissette and Chambers (1992) propose that actually relatively little new root growth is needed to increase the capability of roots to absorb water. The new, unuberized roots are finer, with increased surface area that rapidly increase a seedling's ability to absorb water.

Because of the wide variation in a seedling's ability to regenerate new roots after planting, Stone (1955) regarded this ability to be an indication of their physiological status. It should be noted that a seedling's ability to regenerate new roots is commonly referred to as root-growth potential (RGP). Root-growth potential is generally developed while the seedling is growing in the nursery (Ritchie 1984), and may be affected by nursery cultural practices. Root-growth potential may also be influenced by method and timing of lifting and method and duration of storage. Hence, it is most important for nursery personnel to understand the consequences of their actions and how they will affect the program as a whole.

Root-growth potential can be directly related to field survival, and greater RGP will allow seedlings to better survive water stress during establishment (Brissette 1990). Brissette (1990) also considered RGP related to root system size and number of lateral roots present. He suggested that nursery practices that increase the root system size or the number of lateral roots should also increase RGP if all other factors remain constant. In contrast, Pawsey (1972) noted that growth rate was not appreciably influenced by the condition of the root system at time of planting. He considered regeneration of the roots to be independent of the number of existing lateral roots.

### Shoot:Root Ratio

Studies have shown average shoot:root ratios of 2:1 to be adequate (May 1984, Feret and Kreh 1986). There appears to be a difference of opinion when it comes to predicting seedling performance from shoot:root ratio. Some studies have shown a definite (positive or negative) correlation between shoot:root ratio and seedling outplanting performance (Larsen et al. 1988; Venator 1983; South et al. 1985), while others are skeptical about making that prediction (Rietveld 1989). In 1984, Lavender proposed several reasons for the differences of opinion. First, he noted that shoot:root ratio may vary as a result of age (size), genetics,

environment or cultural practices. In addition, the climate and vegetation of the planting site can vary widely. The method for determining shoot:root ratio is not standardized. Moreover, reports tend to omit the causes of different shoot:root ratios and the effects of the environment on the physiological parameters that determine the ratio. Hermann (1964) concluded that a high shoot:root ratio does not have to signal low survival if the seedlings have a well-developed root system.

### Mycorrhizae

Ectomycorrhizae may play a large role in reforestation programs. The association between plant roots and the fungus can result in increased growth and vigor of the plant. Much of this resultant growth is due to increased stress tolerance imposed by low soil fertility, low available moisture and root pathogens (Davey 1990). Mycorrhizae can moderate physiological responses to drought stress by providing resistance to soil water deficits and improved phosphorus absorption (Rietveld 1989). In addition to the fungal mantle that directly protects the roots from pathogens, many ectomycorrhizal fungi produce antibiotics that are used against some root pathogens (Marx 1969a, 1969b). A rapidly invading mycorrhizal fungus can give outplanted seedlings a competitive advantage, allowing them to dominate the site in a shortened period of time (Davey 1990). This will allow the trees to begin accumulation of photosynthate as wood and shorten rotation time.

Nursery inoculation can mean the difference between success or failure when seedlings are outplanted on extreme sites. The presence of mycorrhizae has been a suggested addition to the list of culling standards set up in forest nurseries (Mexal and South 1990). It has been proposed that mycorrhizae presence and grade together provide better criteria than grade alone (Williston 1974).

## NUTRITIONAL STATUS

Recently, it has been suggested that traits such as stem length, stem diameter, root length and dry weight are limited in their ability to predict seedling survival and growth. Wakeley (1948), noting the inconsistencies in the relation of morphological characteristics of seedlings to outplanting performance, concluded that there needed to be a physiological grade that truly represented a seedling's capability to survive and grow. In order to validly compare morphological traits, seedlings will need to be of the same physiological state. Those physiological qualities are much harder to determine than the morphological ones. Most of

this is due to the fact that they are not visible to the eye and may require destructive sampling to be determined. An additional drawback is that some physiological traits require destructive sampling over a period of time in order to obtain the desired trend information.

Williston (1974) stated that differences in physiological quality could be attributed to: 1) differences in mineral nutrition 2) differences in stored food reserves of the seedlings 3) differences in water tension under which the seedlings are grown and (4) fungicidal sprays, spreaders, adhesives, rodent repellent sprays, or other sprays applied at lifting time and presumably affecting the transpiration of the seedlings immediately after they are planted.

Seedling nutritional status has been the subject of increasing research because of the multiple roles which nutrients play in physiological processes (Clarkson and Hanson 1980; Mengel and Kirby 1982). It has been recognized that plant growth requires the presence of at least 17 macro- and micronutrients at proper levels and relative ratios. A variety of studies have evaluated the relationship between fertilization in the nursery and survival and growth after outplanting. Some have shown a positive correlation (Smith et al 1966; van den Driessche 1980b), while others have shown little correlation (Mullin and Bowdery 1977).

Verification of seedling nutritional status is generally determined through destructive sampling. Results are usually given as content or concentration, but sometimes as ratios (Ingstad 1979). Analysis is usually performed on separate plant components. Analysis of seedling shoots usually yields lower nutrient concentrations than foliar analysis. In addition, foliage from seedlings will show higher concentrations than foliage from mature trees (van den Driessche 1980a).

Poor outplanting performance has been linked to the influence of poor nutrient status on seedling functions such as inability to regenerate new roots and related impaired water uptake (Pharis and Kramer 1964). Pharis and Kramer concluded that under certain conditions, nursery fertilization of nutrient deficient plants influences water economy as well as tissue mineral content. Stress resistance may also be affected by nutritional status. Relationships between seedling mineral nutrition and height growth after outplanting have usually been positive (Switzer and Nelson 1963; Smith et al. 1966; Mullin and Bowdery 1977; van den Driessche 1980b). It should be noted that initial growth was not very well correlated to nutrient status, but subsequent growth was. In a study of white spruce, positive effects of nursery fertilization on tree height have been recorded (Mullin and Bowdery 1977). Height of loblolly pine, three years after planting, was correlated with foliar N content at lifting in a 1963 study by Switzer and Nelson. Larsen et al. (1988) found that foliar N content, foliar P concentration and foliar P content were all correlated with total height growth. Moreover, 25 percent of the height growth during those three years, was accounted for by foliar N content.

It has been suggested that higher nitrogen supply in the nursery can increase shoot dry weight which in turn may boost field performance. In a 1988 study by Troeng and Ackzell, size and mass development of Scots pine seedlings was largely determined by the amount of nitrogen supplied. In the same study, low nitrogen supplies favored root growth at the expense of needle and stem growth. Shoot height, root and shoot dry weights, and shoot:root ratios have been found to increase as N applications have increased (van den Driessche 1980b).

High soil fertility levels of N and P have been shown to reduce shoot:root ratios, but increase overall size of the stock. Studies have shown that survival and height growth of outplanted seedlings improved due to N fertilization in the nursery, that substantially increased seedling size at lifting (Bickelhaupt 1988; Smith et al. 1966). A similar study by van den Driessche (1980b), suggested that the major benefit was due to increased seedling size. Switzer and Nelson (1963) proposed that the overall result of increasing seedling size through N fertilization, was to increase the number of plantable seedlings.

Similar nutrient concentrations may be found in markedly different sized seedlings. This may be due to increased N fertilization, which increased seedling weight, but foliar N concentrations remain the same because of nutrient dilution. The time of year that sampling occurs can also affect the seedling tissue content. Seedlings are removed during a "lifting window" to optimize subsequent survival and growth. This window of time is based upon factors such as seedling development (size, bud formation, lateral root development, mycorrhizal development, stored food reserves), chilling requirements, and field planting conditions. Seedlings sampled during the latter part of the lifting window will have higher nutrient content due to increased dry weight accumulation in the latter part of the season. It should also be noted that tissue nutrient concentrations will be different for plants actively growing and those that are dormant.

van den Driessche (1980b) found a relationship between foliar N concentrations and seedling size in the field. Whether the increase in seedling size is due to better nitrogen status or planting a larger seedling with more roots and foliage, is not clear. Late season fertilization has been shown to increase seedling nutrient concentration with little or no effect on seedling size (van den Driessche 1985).

It has been shown that increasing N concentration of nursery seedlings may enhance growth after outplanting, but not necessarily survival (Switzer and Nelson 1963; Larsen et al 1988; Bickelhaupt 1988). In contrast, needle percent N was positively correlated with survival three years after outplanting in the forest (van den Driessche 1984). Larsen et al. (1988) found that both initial and subsequent field growth of loblolly pine was influenced by the foliar N

content at lifting. In a 1987 (van den Driessche) study with Douglas-fir and white spruce, survival appeared related to needle N concentration at the time of planting. It was proposed that maximum survival would be achieved with needle N concentrations of 2.1%. It should also be noted that spacing of the seedlings has been found to influence outplanting performance through its effect on N nutrition (van den Driessche 1984). Smith et al. (1966) found that nursery fertilization of Douglas-fir increased seedling size, then increased field survival from 70 to 95% and field height from 74 to 94 cm after four years. In addition optimal N levels for growth have been found to be different than those for survival.

The concept that increased fertilization will increase tissue mass is still a topic of discussion. van den Driessche (1987) found that increasing levels of fertilizer brought about a corresponding increase in dry weight. Fertilization also increased shoot growth compared with root growth. At the end of two growing seasons, seedlings had more dry weight in stems than in needles. In a 1988 study by Troeng and Ackzell, the amount of nitrogen supplied determined the size and dry matter development of Scots pine seedlings. Bickelhaupt (1988) found that application of nitrogen increased the percentage of large seedlings and decreased the percentage of cull seedlings.

Mineral nutrition has been shown to influence such traits as plant cold hardiness (Alden and Hermann 1971). The most well known consequence of excessive N applied during the summer is delayed dormancy and as a result, frost damage. This can be avoided if the plants have adequate carbohydrate reserves and if active growth has ceased (Weiser 1970). An additional consideration is that excessive use of N, early in the growth process, has often led to increased damping off.

Mineral nutrition can enhance moisture status of seedlings. Loblolly pine seedlings are most drought resistant when N is supplied in an optimal amount for growth (Pharis and Kramer 1964). Nitrogen levels that are too low or too high will further inhibit recovery and growth even after damage is already done. Ritchie (1984) found that N and K can reduce transpiration rate while P may increase it. He also suggested that N and K may improve tissue water relations by enhancement of turgor maintenance through osmotic adjustment.

It has been proposed by some that the form of nitrogen fertilizer (urea, nitrate, ammonium nitrate, ammonium sulfate) may account for the variability in response of nitrogen fertilized seedlings. In addition, a number of variables may affect the outcome of those studies: soil pH and organic matter, fertility of nursery soil, effect of fertilizer additions on soil microbes, frequency of application, and interactions between nutrients and seedlings.

The application of nitrogen fertilizers in bare-root nurseries in the South is a common practice. The rates, timing and analysis of the fertilizer are determined by the individual

nursery based on nursery conditions and management objectives. Traditionally, fertilizer has been applied as a granular top-dress, at constant rates through the early part of the growing season. These applications are usually complete blend, containing proportionally more N than other nutrients. A high concentration level is maintained to promote fast growth during the rapid development of the seedling. After the seedlings reach the juvenile phase and growth slows, a "finisher" blend, containing less N and more P or K, is often applied to encourage hardening off. Recently there have been studies to examine the concept of exponentially increasing fertilizer rates (Brissette et al. 1988; Timmer and Armstrong 1987). Justification for the concept argues that the exponential rate of application will better follow the relative growth rate and nutrient need of the plant. Fertilizer can then better match plant nutrient demand, and internal nutrient concentrations will remain constant even though dry mass increases.

Production of containerized red pine (*Pinus resinosa* Ait.) seedlings with exponentially increasing applications of N has been shown to generate larger seedlings with smaller shoot:root ratios (Timmer and Armstrong 1987). In the same study, height, dry matter and root development were significantly greater than in conventionally fertilized seedlings. The seedlings developed finer root systems with extensive branching and lateral root formation that allowed for greater absorptive capacity. These larger seedlings were associated with enhanced N and P uptake. Internal N concentrations were consistent with those found to be optimum for pine species grown at steady state nutrition. These treatments did not increase seedling height or shoot weight. It is theorized that the constant rate provides too much N early in the development of the seedling, and as a result may have inhibited root growth. A similar study by Brissette et al. (1988) suggested that N fertilization applied exponentially may produce morphologically better balanced seedlings. Fertilization applied exponentially seemingly had a greater effect on seedling roots than on their shoots.

Many southern nurseries suffer from a condition often called summer chlorosis. It has been suggested that this condition results from chlorophyll breakdown during hot weather (May 1984). There have been numerous suggested causes, such as N, S, Mg or Fe deficiency. Fertilizing to achieve recommended shoot tissue concentrations of conifer seedlings has been a means of alleviating this malady. Concentrations of 1.40-2.20%, 0.20-0.30%, 0.10-0.30% and 60-200 ppm are recommended for N, S, Mg and Fe respectively (Landis 1989). But because one of the most important functions of Mg in plants is the formation of chlorophyll, and because chlorophyll contains about 2.7% magnesium, as a constituent of the chloroplasts, it would appear that further study of Mg may be warranted. Although Fe is necessary for the formation of chlorophyll, it is not a constituent of the molecule.



Interpretation of nutritional results can be complicated by many factors. These include environmental effects, genetics, time of sampling, age of seedling, fertilization, soil fertility, nutrient interactions, soil pH, fumigation with methyl bromide, water status, and injury from insects and diseases. Evidence for the benefits of high fertility on outplanted seedlings is not clear. Factors such as lifting date and wrenching, to name a few, must be considered.

## NURSERY EFFECTS

Seedling performance is determined by both morphological and physiological characteristics, but expression of that potential is governed by conditions at the planting site (Puttonen 1989). It is generally accepted that nursery cultural practices influence seedling survival and subsequent growth. Switzer and Nelson (1963) demonstrated how the effects of nursery treatment were related to field height growth through such attributes as size and nitrogen content of the seedlings at lifting. Specific seedling morphological (e.g., shoot:root ratio) and physiological (e.g., nutrient status) characteristics may enhance growth and survival at time of planting. Silker (1960) suggested that the overall size of seedlings at a given age, or stem diameter alone, is a reliable index of early survival. Nambiar and Zed (1980) suggested keeping the same internal nutrient status of seedlings before and after planting to ensure survival and growth.

In addition to nutrition, other factors such as seedbed density, undercutting, top pruning, irrigation and cold storage are known to affect field performance (Ritchie 1984; Duryea 1984; Kennedy et al. 1987). These factors may alter the effect of a particular fertilization regime and make it difficult to evaluate the fertilizer effect. Morphology and nutrient status may need to be considered together to evaluate field performance when cultural practices modify the seedling's morphology. Burdett and Simpson (1984) made the observation that different types of stock will develop similar morphological traits after planting because they will all be adjusting to a common environment.

Once a crop is successfully established, irrigation, fertilization, and weed control should be carefully administered to assure a seedling crop reaches its target height and has 75 to 80 percent of its target diameter early enough to allow conditioning of the seedling (Barnett et al. 1984). At this point, treatments such as undercutting, wrenching, and nutrient and water management can be used to begin control of height growth and promote budset. If seedlings are raised under the best of conditions, then any reduction in stem diameter as a result of height growth control will be minimized.

In order to ensure that nutrition is not a limiting factor of the potential field performance, nursery managers must consider the effects of nutritional practices beyond the impact on seedling height or morphological grade. Duryea (1984) recommends that nursery personnel develop a better understanding of the effects of their current cultural practices so implementation of those practices will lead to improved seedling quality. These seedlings would be better matched to forest sites and improve future growth of that forest site. Seedling performance, at any particular site, could be readily determined by including the forester's hands-on knowledge in the evaluation process. If nursery personnel were to supply the forester with the physical and chemical measurements of the stock he received each year, that stock's performance could be evaluated under those particular conditions and give an indication of future performance.

## OBJECTIVES

The overall objective of this research was to evaluate the effect of macro-nutrient nutrition on the quality of loblolly Pine (*Pinus taeda* L.) seedlings.

Specific objectives were:

sub-objective 1 - evaluate influence of varying amounts of pre-plant nitrogen fertilizer on shoot length, root length, stem diameter and tissue concentrations of N, P, K, Ca, and Mg.

sub-objective 2 - evaluate influence of varying amounts of top dress magnesium fertilizer on shoot length, root length, stem diameter and tissue concentrations of N, P, K, Ca, and Mg.

sub-objective 3 - evaluate influence of varying amounts of top dress nitrogen fertilizer on shoot length, root length, stem diameter and tissue concentrations of N, P, K, Ca, and Mg.

sub-objective 4 - evaluate influence of different treatment history of two nursery compartments on shoot length, root length, stem diameter and tissue concentrations of N, P, K, Ca, and Mg.

## CHAPTER III

### EXPERIMENTAL DESIGN

In order to meet the objectives, the following hypotheses will be tested:

Hypothesis 1 - Applications of pre-plant ammonium nitrate fertilizer will alter shoot length, root length, root to shoot ratio, stem diameter and tissue concentrations of N, P, K, Ca, and Mg.

Hypothesis 2 - Applications of top-dress magnesium sulfate fertilizer will have an effect on shoot length, root length, root to shoot ratio, and tissue concentrations of N, P, Mg, K, and Ca.

Hypothesis 3 - Applications of top-dress ammonium nitrate fertilizer will influence shoot length, root length, root to shoot ratio, stem diameter and tissue concentrations of N, P, K, Ca, Mg.

Hypothesis 4 - Different treatment history of two nursery compartments will influence seedling quality as measured by shoot length, root length, stem diameter and tissue concentrations of N, P, K, Ca, Mg.

Each experiment will be set up using a randomized complete block design.

## CHAPTER IV

## MATERIALS AND METHODS

## GENERAL PROCEDURES

The experiments were carried out at the Texas Forest Service Indian Mound Nursery near Alto, Texas in Cherokee County. Experiments were carried out in four separate operational beds, each 1.2m wide within two different compartments (7a & 4). The soil at the nursery is an Amite fine sandy loam. Recent soil analyses can be found in Table 3.

Table 3.

Soil analysis of Texas Forest Service Indian Mound Nursery in 1987.  
Analysis performed by A & L Laboratories, Memphis, TN.

Compartment	pH	%O.M.	CEC <sup>1</sup>	%Clay	P <sup>2</sup>	Ca/Mg <sup>2</sup>	K/S/Fe <sup>2</sup>
4(bed 3)	5.2	1.8	3.9-L	14.8	134-VH	400-M 31-VL	125-VH 15-H 153-VH
7a(bed 4)	4.8	1.2	5.4-L	18.8	11-L	390-L 62-L	145-VH 28-VH 23-H

<sup>1</sup> Reported as meq/100g.

<sup>2</sup> Reported as parts per million - ppm.

The selected seed source for all experiments was superior loblolly, bred for improved growth rate and form by Texas Forest Service personnel. Seed was stratified, treated with Bayleton and Thiram (fungicides) and a repellent (bird), then sown by Texas Forest Service personnel using established nursery practices (May 1984).

Seeds were operationally sown to a target density of 290 seedlings per square meter the week of April 8 through April 15, 1988. A light mulch of composted post peelings was applied over the seeds to protect seeds from desiccation and displacement.

All experiments were irrigated according to operational nursery practices. The top dress schedule attempted to follow a normal treatment schedule except where indicated. Seedlings were undercut and side-pruned according to standard nursery techniques. Seedlings were not top pruned.

## FIELD PROCEDURES

Field measurements, destructive sampling and laboratory handling of collected materials were the same for all experiments.

### Experiment 1 - Pre-plant Nitrogen

Experiment 1 was designed to test the effects of pre-planting nitrogen on seedling morphology and physiology. Plots were 1.2 m (width of bed) x 6.15 m long and located within a single planting bed in compartment four. Plots were placed end to end in the bed running from northeast to southwest; however, measurements were not taken within 1 m of the plot ends. The experiments were located at least 14 m in from each end, to minimize any end effects due to tractor speed, tool depth etc..

Nitrogen was applied in the form of ammonium nitrate ( $\text{NH}_4\text{NO}_3$ ). Pre-plant treatments at five different rates were used. The rates were as follows : 0, 11, 22, 45 and 90 kg/ha (0, 10, 20, 40 and 80 lbs/acre) of elemental N. These were applied during bed preparation. The fertilizer was placed on the beds and then tilled in to a depth of approximately 10 cm.. The experiment was replicated four times in a randomized complete block design.

A total of 20 sampling plots, one per plot replicate, were established in the bed for the July 1988 and January 1989 target dates. Samples were taken from a randomly selected, 7.6 cm strip across the width of the bed. Needle, stem and root tissue were later analyzed for N, P, K, Ca, and Mg.

### Experiment 2 - Top-dress Magnesium

Experiment 2 was designed to test the effects of top-dress magnesium on seedling morphology and physiology. Procedures for experiment 2 were identical to those for experiment 1 except for plot dimension and placement, fertilizer applied and tissue analysis. The plots were 2.4 m long by 1.2 m wide, located within a single planting bed in compartment four. The plots were placed end to end in the bed running northeast to southwest; no samples were taken within 50 cm of the plot ends. They parallel the plots in experiment 1. The series of plots began at the same point as in experiment 1, 14 m in from the end of the bed.

Magnesium was applied as magnesium sulfate ( $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ ). The seedlings

received five different treatments; three with constant rates of application, one with an increasing rate of application and one untreated control.

This experiment contained four replications. Seedlings received magnesium fertilization as a top-dress every three weeks beginning the sixth week after sowing. The applications continued through August and included five total applications. The three constant treatments received 17, 28, 39 kg/ha (15, 25 and 35 lbs/acre) of elemental Mg, respectively. The non-constant rate increased exponentially in an effort to mimic plant growth. The exponentially increasing application rates were: 4.5, 9.0, 18.0, 36.1 and 72.3 kg/ha (4.03, 8.06, 16.12, 32.24 and 64.48 lbs/acre) of elemental Mg. The total applied fertilizer (5 applications) equaled that applied at the 28 kg /ha rate (140 kg/ha). Standard nursery top-dress applications were also applied.

On the July 1988 and January 1989 target dates, one square foot of seedlings per replication were lifted and analyzed for N, P, S, Mg, K, Ca. Sampling procedures were identical to those in experiment 1.

Foliar S concentration were evaluated to determine if fertilization with  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  had a significant impact on foliar Sulfur.

### Experiment 3 - Top-dress Nitrogen

Experiment 3 was designed to test the effects of top-dress nitrogen and fumigation with methyl bromide on seedling morphology and physiology. Procedures for experiment 3 were similar to those of experiment 2 except for the number of replications (three), fertilizer applied, and tissue analysis. This experiment was divided into two blocks with three replications within each block. One block was placed in compartment four (bed 3), parallel to experiment 2. This compartment was fumigated with methyl bromide in October 1986, two years prior to sowing. The other block was placed in compartment 7a (bed 4), which was fumigated October 1987.

Nitrogen was applied as ammonium nitrate ( $\text{NH}_4\text{NO}_3$ ). Seedlings received five different treatments. They received nitrogen fertilizer as a top-dress every three weeks beginning the sixth week after sowing. There was a total of five applications with the last occurring in late August. Nitrogen fertilizer was applied at the following constant rates : 0, 17, 34, and 67 kg/ha (0, 15, 30, and 60 lbs/acre), and at an exponential rate of elemental N. The exponential rate was as follows: 5.4, 10.8, 21.5, 43.0, and 86.1 kg/ha (4.8, 9.6, 19.2, 38.4, and 76.8 lbs/acre) per respective application week. The goal of the exponential rate was to

apply the same amount of total N as would be applied at the 34kg (30 lb) constant application rates (170 kg/ha).

July 1988 and January 1989 sampling for experiment 3 was carried out in the same manner as experiment 1. Tissue analysis was performed to determine concentrations of N, K, P, Ca and Mg.

### Field Measurements

Two observation plots, each 0.09 m<sup>2</sup> were located randomly within each rep/plot combination. The observation plots were 15.2 cm wide and 61 cm long. Measurements of seedling height and diameter within the observation plots were taken on a monthly basis. These measurements began four weeks after sowing and continued until lifting. Height was measured from the soil line to the tip of the needles or the terminal bud, when it was formed. Stem diameter was measured at or as close as possible to the soil line. Measurements continued through lifting in January 1989.

### Sampling

The initial destructive sampling, for mass measurements, was done on July 4, 1988. A second sampling was performed at lifting, on January 9th and 10th, 1989, for mass measurements as well as nutrient analysis. Time of lifting was selected to fall within the optimal lifting period for loblolly pine. Seedlings were undercut prior to lifting in order to more realistically assess treatment effects on root quality. Determination of which seedlings to lift was made on a random basis. One 0.09 m<sup>2</sup> plot of seedlings was lifted from each plot. This includes four replications in experiments one and two and three reps in each bed of experiments three. The seedlings were taken from a 7.6 cm section spanning the width of the bed.

The seedlings were placed in plastic bags grouped by measurement plot and stored on ice. Bundles were transported back to College Station, Texas and placed in 2°C cold storage.

All seedlings were rinsed with distilled water to remove soil and other debris from the roots and stems. When seedlings were cleaned, measurements were made of shoot length, root length and root collar diameter. The height was measured to the end of the needles or the bud, if formed. The root was measured to the end of the collected tap root.

Prior to chemical analysis, the seedling components were dried to equilibrium moisture content at 70°C in a forced air oven and separated into foliage, stem and root

components. July sample components were bulked together and weighed on a per plot basis. January sample components were weighed as individual seedling parts and ground to pass a 20 mesh screen using a Wiley Mill. Seedling component parts were then bulked for the January sample and placed in bags for nutrient analysis.

## ANALYTICAL PROCEDURES

### Laboratory Analysis

Approximately 0.40 g of ground oven-dried plant material was digested by a modified Kjeldahl digestion procedure (Parkinson and Allen 1975). Digestion was performed on an Orion-Scientific Instruments block digester. Nitrogen and P levels in the digests were determined colorimetrically by using indo-phenol blue and molybdenum blue methods, respectively (Allen et al. 1974). The colorimetric procedure analysis was carried out using an Orion-Scientific Instruments autoanalyzer (Scientific Instruments Co., Pleasantville, New York).

Determination of Ca, Mg and K content was done using a Perkin-Elmer 2380 Atomic Absorption Spectrophotometer (Perkin-Elmer, Norwalk, Connecticut). Digest samples and standards were diluted with a 5000/1000 ppm La-Cs mixture (Parkinson and Allen 1975).

Foliage was analyzed for sulfur concentration using a mixed nitric acid - perchloric acid digestion procedure (Jackson 1958), with the S concentration of digest solutions determined using an inductively coupled plasma emission spectrophotometer (ICP). Digest solutions and standards were analyzed by the Texas A & M University Department of Horticulture's ICP Analytical Center.

### Data Analysis

Statistical analysis was achieved using the General Linear Model (SAS Institute Inc.) on the means for a randomized complete block design. General Linear Model (GLM) analysis was performed on data from shoot length, root length and stem diameter, as well as component weights and tissue nutrient concentrations of N, P, K, Ca, Mg and S. Statistical significance due to fertilizer rate (all experiments) and soil (experiment 3) was determined ( $P = .05$ ) using the GLM procedure of SAS. Models and EMS tables are in Appendix A.

Component parts were individually analyzed or set up as ratios and compared. The following variables were determined for each experiment : mean needle weight, mean stem



weight, mean root weight, mean top weight = needle + stem weight, mean needle to stem weight, mean top to root weight, and mean needle to root weight.

## CHAPTER V

## RESULTS

## MORPHOLOGICAL TRAITS

Experiment 1 - Pre-plant Nitrogen

Analysis of pre-plant nitrogen data showed little significant influence on seedling dimensions(see Appendix B); however there was a significant block effect on root length. This may be an indication of the variability of the soil within the nursery. No significance was shown for any mass analysis. Rate of application significantly affected mean needle to stem weight, but not needle to root or shoot to root weight. Summaries of component data are found in Table 4.

Table 4.

Average shoot length(cm), stem caliper(mm) and root length(cm) of loblolly pine seedlings receiving applications of pre-plant nitrogen(January 1989). Means in each column followed by same letters are not significantly different at the 95% level.

Effect	Component		
	Caliper	Shoot length	Root length
<u>Block</u>			
A	3.79a	23.88a	19.96a
B	3.74a	23.18a	17.70b
C	3.71a	24.56a	17.37b
D	3.78a	25.25a	18.12a
<u>Rate</u>			
0 kg N/ha	3.80a	24.90a	17.92a
11 kg N/ha	3.75a	23.90a	18.44a
22 kg N/ha	3.62a	24.59a	18.74a
45 kg N/ha	3.84a	24.25a	18.56a
90 kg N/ha	3.77a	23.45a	17.78a

### Experiment 2 - Top-dress Magnesium

Block effects were observed on shoot length in top-dress applications of magnesium and also on mean shoot to root and mean needle to root ratios. No significant effects of fertilizer application were observed on seedling morphology. Table 5 summarizes the data means.

**Table 5.**

Average shoot length(cm), stem caliper(mm) and root length(cm) of loblolly pine seedlings receiving applications of top-dress magnesium(January 1989). Means in each column followed by same letters are not significantly different at the 95% level.

<b>Effect</b>	<b>Component</b>		
	Caliper	Shoot length	Root length
<u>Block</u>			
A	3.89a	26.53a	17.67a
B	3.74a	22.94b	17.36a
C	3.92a	23.18b	16.59a
D	3.95a	23.71b	16.29a
<u>Rate</u>			
0 kg Mg/ha	3.99a	25.67a	17.17a
17 kg Mg/ha	4.0 a	25.37a	16.86a
28 kg Mg/ha	3.94a	22.94a	16.99a
39 kg Mg/ha	3.68a	22.93a	16.97a
exp kg Mg/ha	3.77a	23.52a	16.89a

### Experiment 3 - Top-dress Nitrogen - Bed 3(compartment 4)

No significance due to rate of fertilizer application was observed on individual component (size or mass) analyses. Block effects were seen on stem caliper, shoot length and root length in top-dress applications of nitrogen in bed 3(compartment 4). A significant block effect was also indicated for mean stem weight, mean shoot (needle+stem) weight, and mean needle weight. Significant rate effects were seen on mean needle:stem and mean needle:root

weights, but not on mean shoot:root weights. Table 6 summarizes bed 3(compartment 4) data.

**Table 6.**

Average shoot length(cm), stem caliper(mm) and root length(cm) of loblolly pine seedlings receiving applications of top-dress nitrogen(compartment 4)(January 1989). Means in each column followed by same letters are not significantly different at the 95% level.

Effect	Component		
	Caliper	Shoot length	Root length
<u>Block</u>			
A	3.94a	23.49a	18.37a
B	3.56b	20.50b	16.45b
C	3.41b	20.75b	15.98b
<u>Rate</u>			
0 kg N/ha	3.38a	20.95a	16.37a
17 kg N/ha	3.65a	21.92a	16.02a
34 kg N/ha	3.70a	21.98a	17.98a
67 kg N/ha	3.81a	21.63a	16.54a
exp kg N/ha	3.65a	21.40a	17.76a

### Experiment 3 - Top-dress Nitrogen - Bed 4(compartment 7a)

Analysis of component parts, weights and weight ratios showed no significance due to rate of N application in bed 4(compartment 7a) except for mean needle weight. Table 7 catalogs component data as a summary.

### Experiment 3 - Combined Analysis

Comparisons between top-dress applications of nitrogen in beds 3(compartment 4) and 4(compartment 7a) showed a significant effect due to bed location for stem caliper, shoot length but not root length. A significant block within bed effect was detected for stem caliper and root length. Comparisons between top-dress nitrogen applications in beds 3(compartment

4) and 4(compartment 7a) showed significant bed effects for mean stem and needle weights, but not root weight (Figs. 1&2).

Table 7.

Average shoot length(cm), stem caliper(mm) and root length(cm) of loblolly pine seedlings receiving applications of top-dress nitrogen(compartment 7a)(January 1989). Means in each column followed by same letters are not significantly different at the 95% level.

Effect	Component		
	Caliper	Shoot length	Root length
<u>Block</u>			
A	4.16b	29.80a	15.38a
B	4.73a	29.59a	16.41a
C	4.20b	30.07a	15.41a
<u>Rate</u>			
0 kg N/ha	4.15a	30.23a	16.35a
17 kg N/ha	4.64a	29.61a	15.65a
34 kg N/ha	4.35a	30.74a	16.22a
67 kg N/ha	4.57a	29.45a	15.70a
exp kg N/ha	4.11a	29.06a	14.74a

Comparing weight ratios between beds 3(compartment 4) and 4(compartment 7a), showed that bed location had a significant effect on almost every independent variable in the model. These included mean shoot weight, mean stem weight, mean shoot:root weight, mean needle:root weight, mean needle:stem weight. In addition, a block within bed effect was seen for mean needle weight, mean stem weight, mean root weight, and mean shoot weight.

No statistically significant differences were found between treatments of the July sampling period - see Appendix D.

Height growth curves representing monthly measurements taken throughout the growing season can be found in Appendix E.

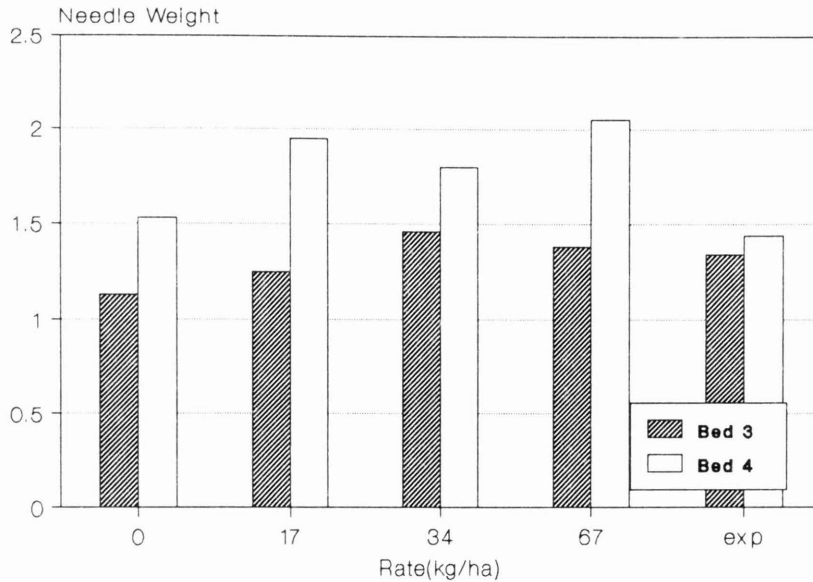


Figure 1. Comparison of mean needle weight(g) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N(January 1989). N=15.

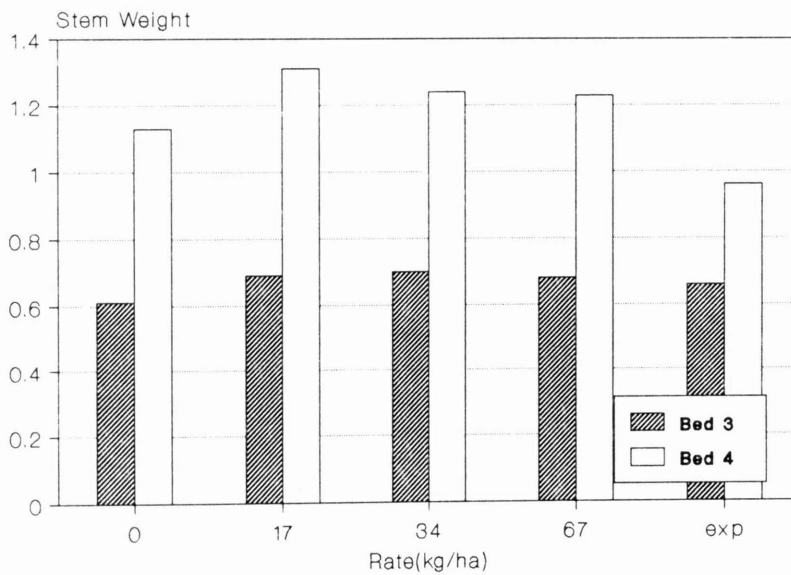


Figure 2. Comparison of mean stem weight(g) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N(January 1989). N=15.

## NUTRIENT CONCENTRATIONS

Statistical analysis was performed by comparing constant rates of application against other constant rates (0, 17, 34, & 67). The exponential application rate was not included as part of this first statistical analysis. The exponential rate was statistically compared only to that constant rate of application that matched the total amount of fertilizer applied.

### Experiment 1 - Pre-plant Nitrogen

Statistical analysis showed no significant effects due to rate of application on the nutritional status of the seedlings. Mean concentrations for components at the different rates are summarized in Table 8.

Table 8.

Mean nutrient concentrations of foliage, stem and root components of bed 1 loblolly pine seedlings receiving pre-plant applications of nitrogen(January 1989).

Nutrient	Rate(kg N/ha)	Mean wt %		
		Foliage	Stem	Root
Nitrogen	0	1.53	.66	.63
	11	1.61	.70	.66
	22	1.53	.67	.64
	45	1.54	.66	.65
	90	1.60	.70	.70
Phosphorus	0	.17	.17	.20
	11	.17	.18	.20
	22	.17	.18	.20
	45	.17	.17	.19
	90	.18	.17	.20
Calcium	0	.37	.24	.17
	11	.38	.26	.19
	22	.36	.25	.18
	45	.37	.24	.18
	90	.41	.24	.19
Magnesium	0	.07	.09	.06
	11	.07	.09	.06
	22	.07	.09	.06
	45	.07	.09	.06
	90	.07	.08	.06
Potassium	0	.56	.68	.66
	11	.57	.71	.66
	22	.56	.75	.63
	45	.58	.67	.63
	90	.63	.63	.65

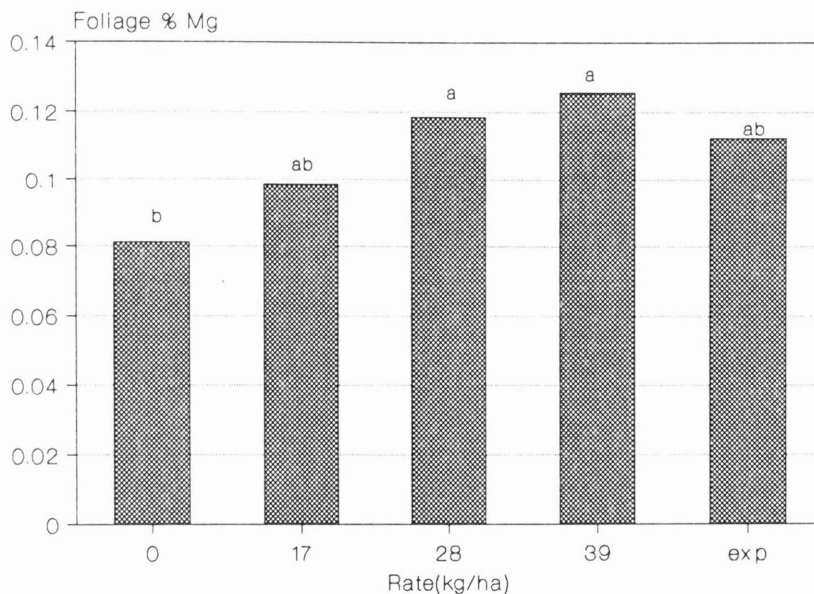


Figure 3. Mean foliar Mg concentration (weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (January 1989). Columns with same letter are not significantly different at the 95% level. N=8.

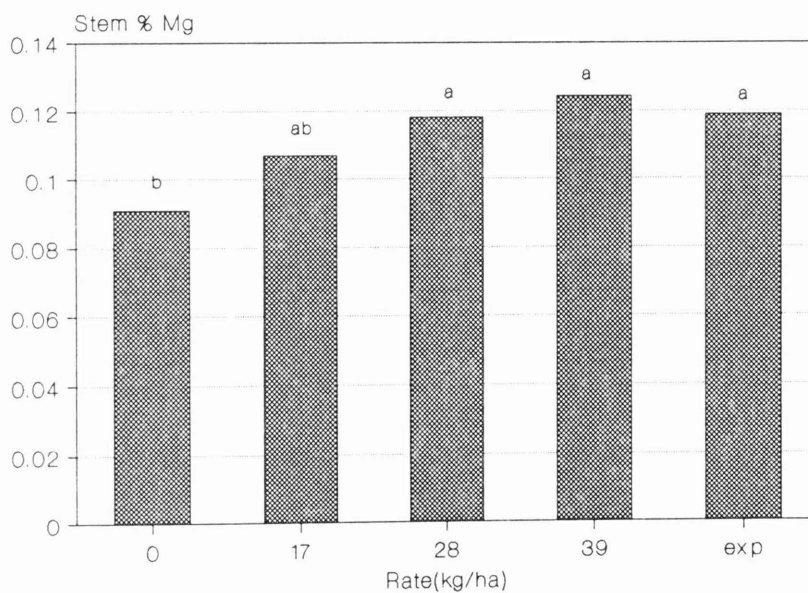


Figure 4. Mean stem Mg concentration (weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (January 1989). Columns with same letter not significantly different at the 95% level. N=8.



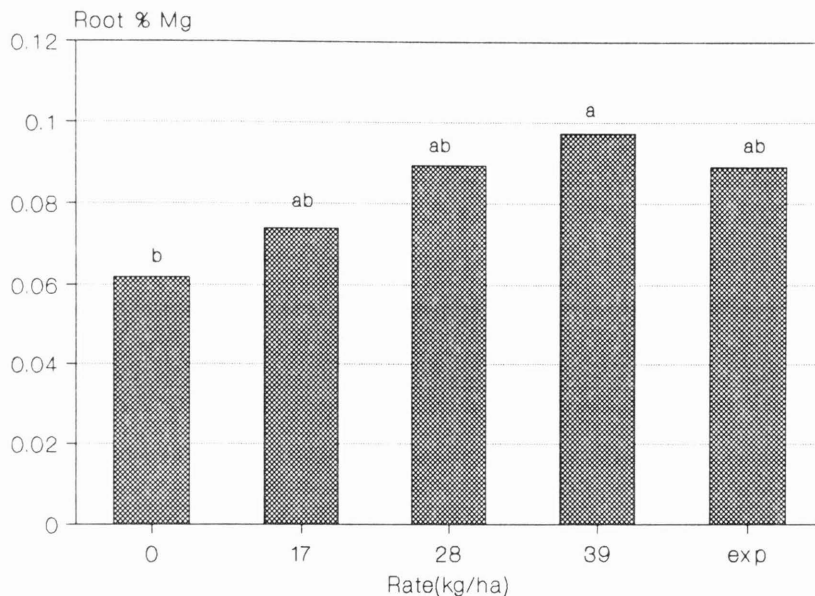


Figure 5. Mean root Mg concentration (weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (January 1989). Columns with same letter not significantly different at the 95% level. N=8.

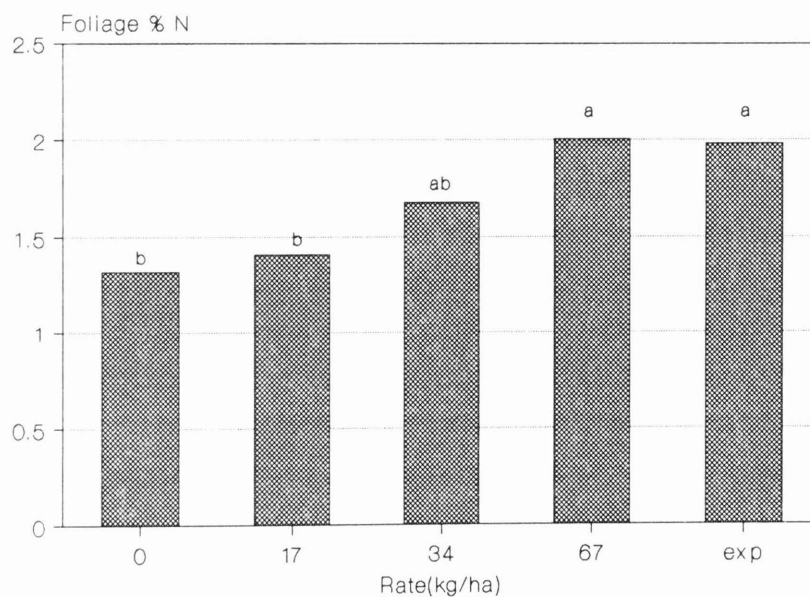


Figure 6. Mean foliar N concentration (weighted %) of bed 3 (compartment 4) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letter not significantly different at the 95% level. N=8.

### Experiment 2 - Top-dress Magnesium

Rate of application was found to cause a significant increase in N concentration in foliar tissue, but not stem or root tissue. No significant effects were detected for concentrations of P, Ca or K in plant tissue. Magnesium concentrations found in foliage, stem and roots all were significantly affected by rate of application with the roots showing a significant block effect (Figs. 3-5). Sulfur foliage concentrations were significantly affected by rate of application. No significant interactions were detected within this experiment.

As noted in the methods, foliar S concentrations were evaluated to determine if fertilization with  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  had a significant impact on foliar S. As shown in Appendix C, foliar S varied from 0.12 to 0.16% depending on Mg fertilizer rate. There were significant differences due to treatment, however, all are well above S critical levels and do not appear to be biologically significant.

Comparisons of the exponential application rate versus the equivalent constant rate showed that constant applications resulted in significantly higher Mg and Ca foliage levels. N, P, and K concentrations remained unaffected. A block effect was observed on both Ca root and Mg foliage concentrations such that two of the four blocks were significantly different for each element.

### Experiment 3 - Top-dress Nitrogen Bed 3(compartment 4)

While nitrogen foliage and stem concentrations were significantly affected by rate of application, root tissue was not (Figs. 6-7). Root and stem concentrations showed an additional block effect. Magnesium concentrations of the roots were also significantly affected by block, whereas P, Ca, and K showed no significant effects due to rate of application or block. There were no significant interactions detected for any nutrients.

Analysis of exponential versus constant equivalent rates of application showed significantly higher K stem and Mg root concentrations when fertilizer was applied exponentially. In addition, there was a significant block effect for stem K concentrations.

### Experiment 3 - Top-dress Nitrogen Bed 4(compartment 7a)

Rate of application was found to significantly increase foliage, stem and root tissue concentrations of N (Figs. 8-10). Foliage N concentrations were also significantly affected by block. Phosphorus foliar concentrations were increased significantly by rate of N application,

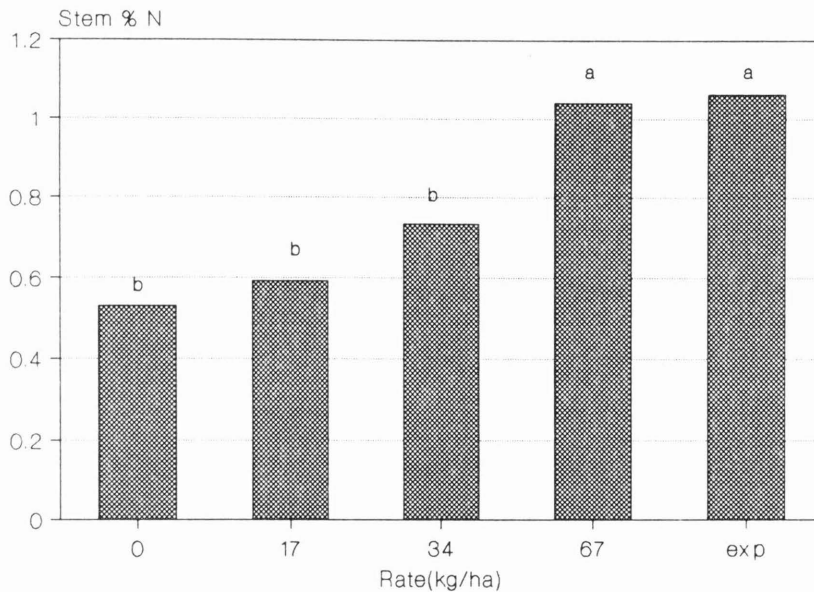


Figure 7. Mean stem N concentration (weighted %) of bed 3 (compartment 4) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letter not significantly different at the 95% level. N=8.

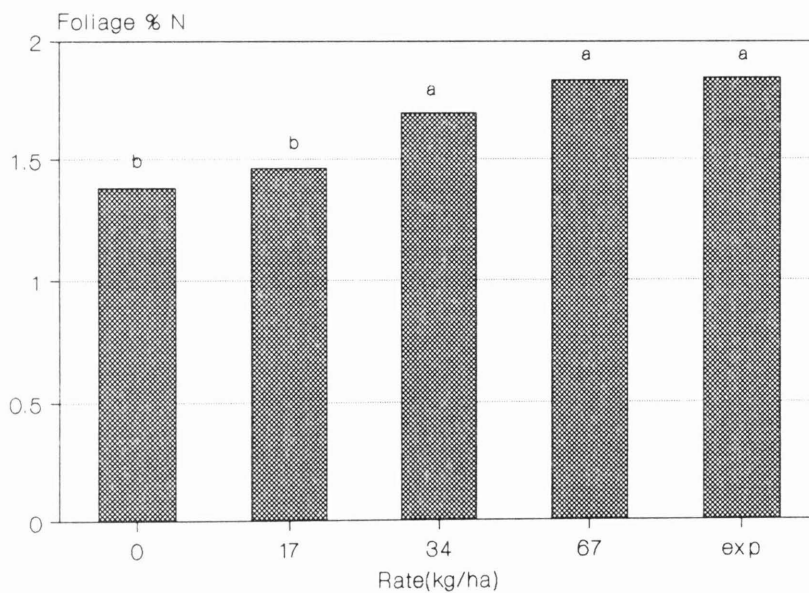


Figure 8. Mean foliar N concentration (weighted %) of bed 4 (compartment 7a) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letters not significant different at the 95% level. N=6.

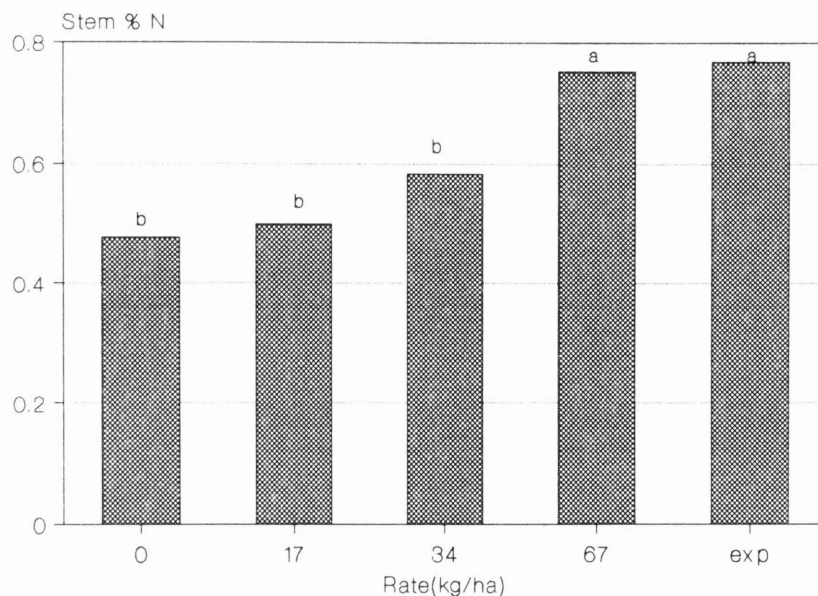


Figure 9. Mean stem N concentration (weighted %) of bed 4 (compartment 7a) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letters not significantly different at the 95% level. N=6.

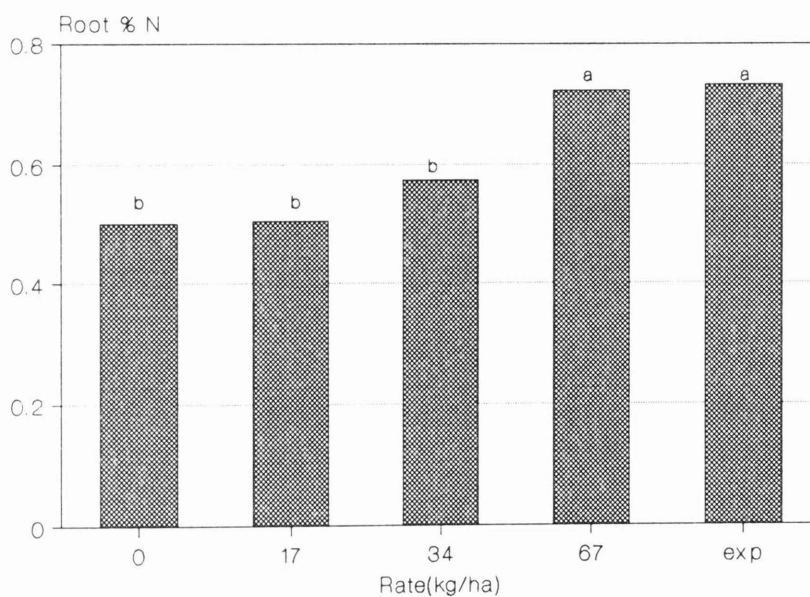


Figure 10. Mean root N concentration (weighted %) of bed 4 (compartment 7a) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letters not significantly different at the 95% level. N=6.

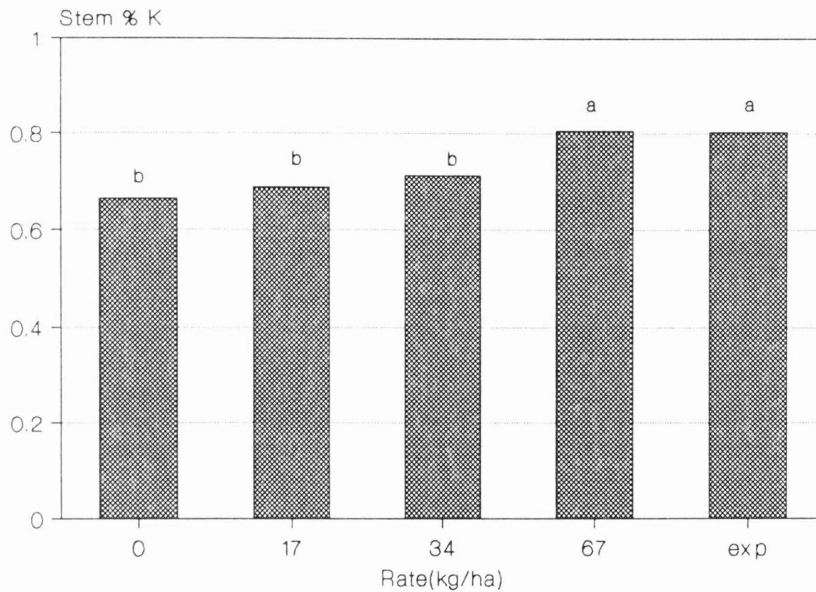


Figure 11. Mean stem K concentration (weighted %) of bed 4 (compartment 7a) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letters not significantly different at the 95% level. N=6.

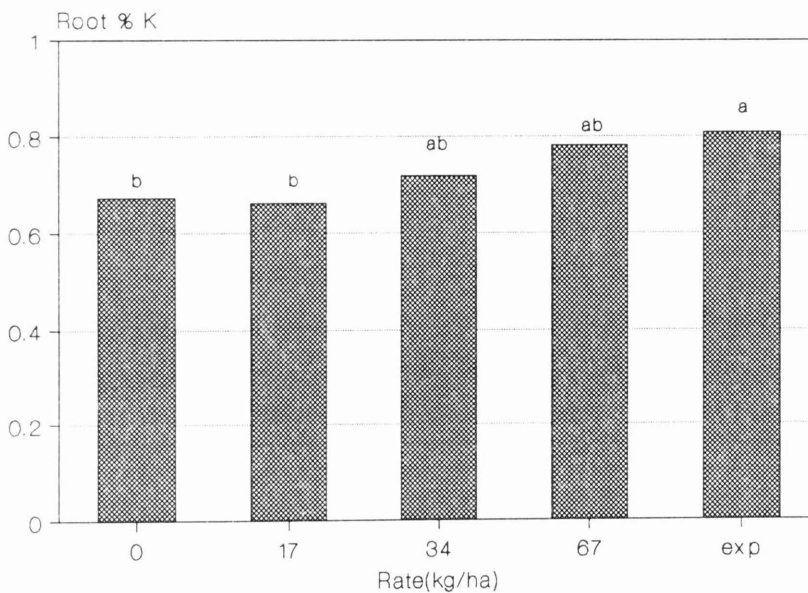


Figure 12. Mean root K concentration (weighted %) of bed 4 (compartment 7a) loblolly pine seedlings receiving varying amounts of top-dress N fertilizer (January 1989). Columns with same letters not significantly different at the 95% level. N=6.

but were also influenced by block placement. Rate of application increased K concentrations in both stem and root tissue (Figs. 11-12), but not foliar tissue. Analysis of Ca and Mg concentrations showed no significant effects due to rate of application or block. No significant interactions were detected within this experiment.

Comparisons of exponential versus constant equivalent rates of application indicate that exponential applications significantly increased foliage, stem and root concentrations of N (Fig. 13). P and Ca showed no significance for any component. Mg root as well as K stem and root concentrations were significantly increased by the exponential applications. Stem K was also significantly affected by block.

### Experiment 3 - Combined Analysis

Analysis showed significant effects that could be attributed to bed location for top-dress N applications of stem and root tissue concentrations of N (Figs. 14-15). Foliage and root tissue concentrations were affected by block within bed. There was a significant bed\*rate interaction on stem N concentrations.

Phosphorus foliage, stem and root concentrations were all significantly affected by block within bed. Stem concentrations of P were the only tissue to be significantly affected by bed location (Fig. 16). Root concentrations were significantly affected by a bed\*rate interaction.

Stem and root, but not foliage, concentrations of K were affected significantly by location of the bed (Figs. 17-18). Root concentrations were also significantly affected by the block within the bed.

No Ca tissue concentrations were significantly affected by bed location. Stem and root concentrations were significantly affected by block within bed.

Root concentrations of Mg were the only tissue significantly affected by bed location (Fig. 19). Foliage and root concentrations were affected by block within bed.

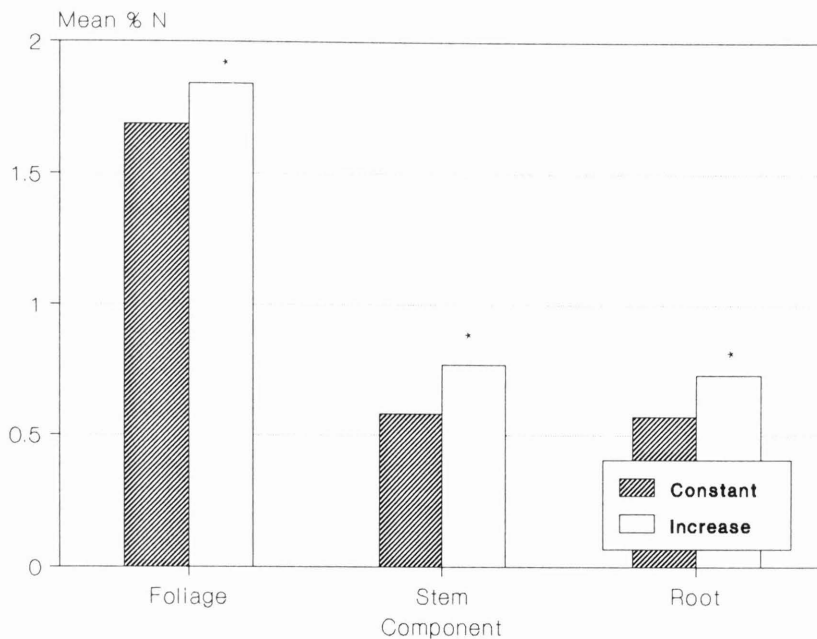


Figure 13. Comparison of mean N concentration(weighted %) of bed 4(compartment 7a) loblolly pine seedlings receiving top-dress N at constant or increasing application rate(January 1989). Constant rate=34 kg/ha elemental N. Increasing rate=5.4,10.8,21.5, 43.0,and 86.1 kg/ha. Significance at the 95% level indicated by \*. N=6.

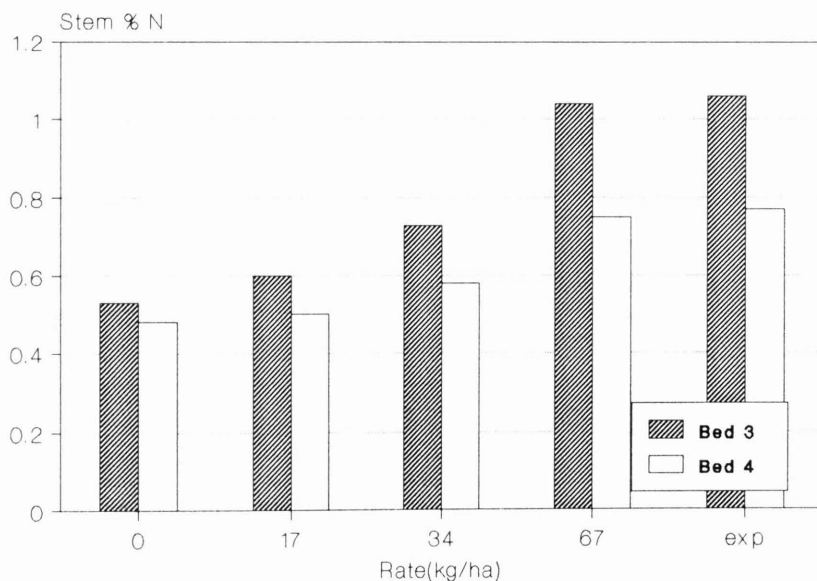


Figure 14. Comparison of mean stem N concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N(January 1989). N=12.

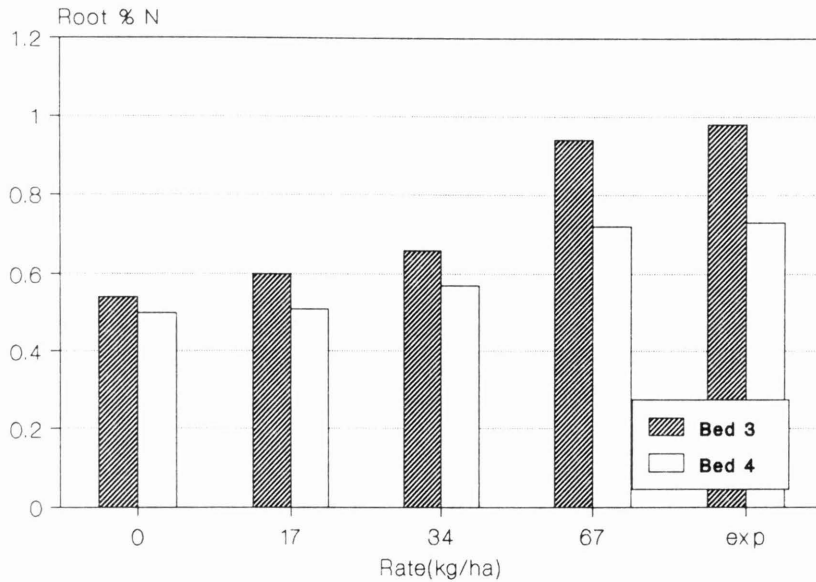


Figure 15. Comparison of mean root N concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress applications of N (January 1989). N=12.

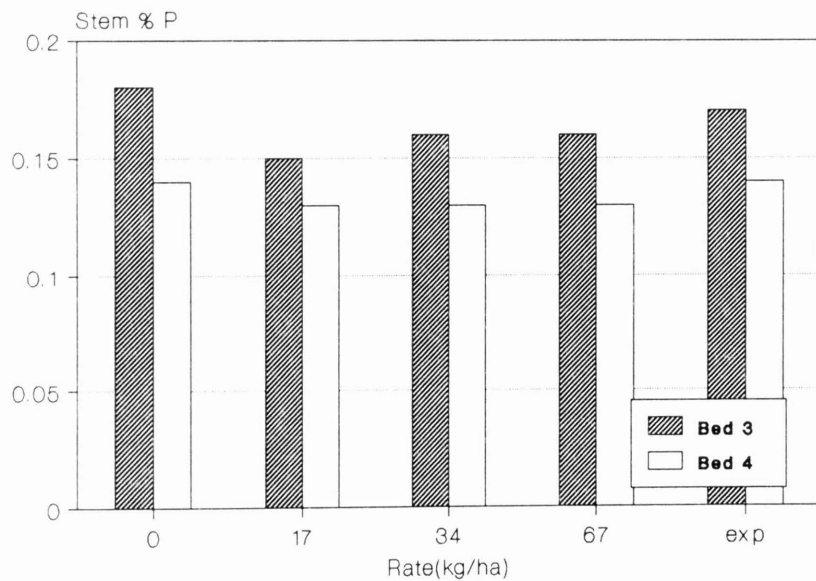


Figure 16. Comparison of mean stem P concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress applications of N (January). N=12.



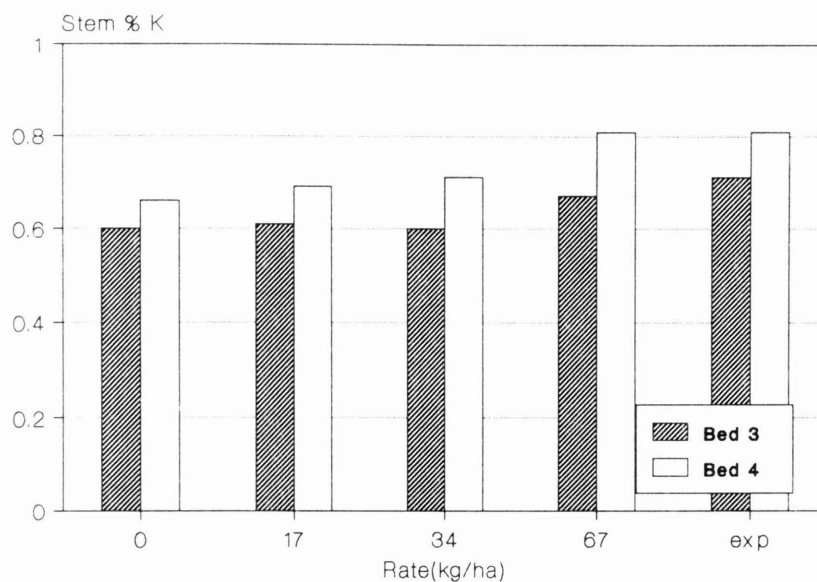


Figure 17. Comparison of mean stem K concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N(January 1989). N=12.

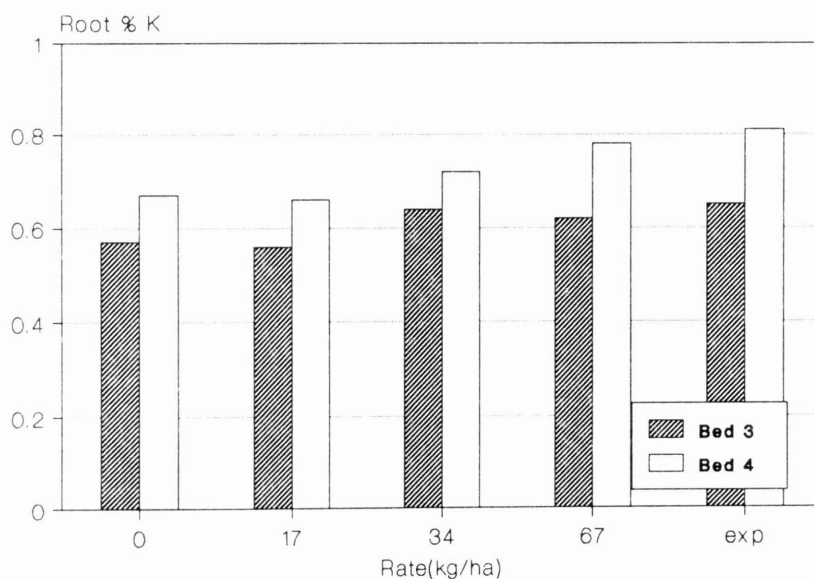


Figure 18. Comparison of mean root K concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N(January 1989). N=12.

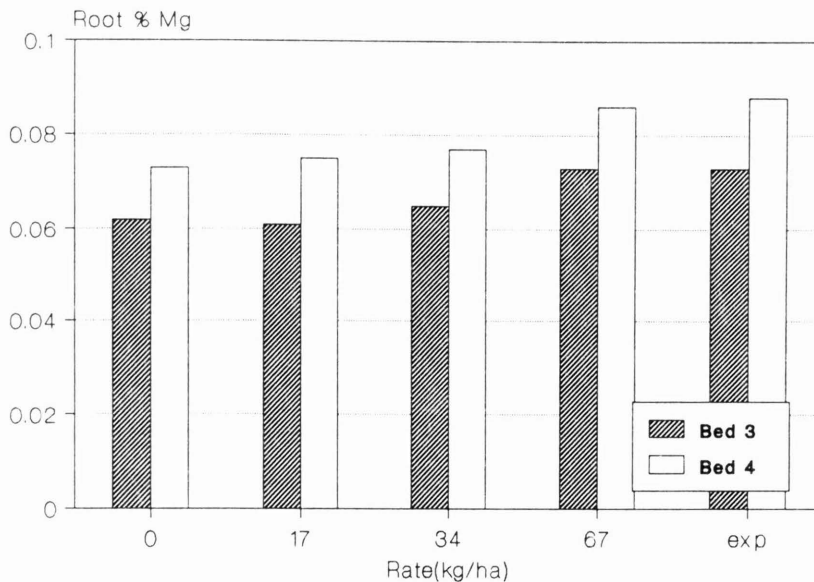


Figure 19. Comparison of mean root Mg concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress applications of N (January 1989). N=12.

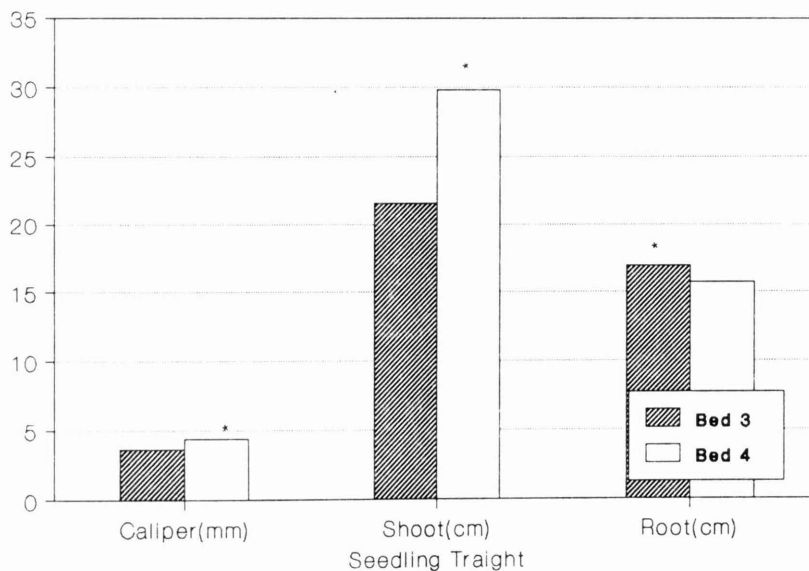


Figure 20. Comparison of mean stem caliper, shoot length, and root length between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress applications of N (January 1989). Significance at the 95% level is indicated by \*. N=6.

## CHAPTER VI

### DISCUSSION

#### MORPHOLOGICAL TRAITS

##### Stem Diameter, Shoot Length, Root Length

In general, this fertility study produced minimal significant morphological results. This would suggest that perhaps the nursery seedlings were at an optimum level of fertility and any additional fertilizer yielded no benefit. Overall, seedling dimensions were within customary target seedling parameters, with the exception of stem diameter, which was smaller than should be according to Brissette & Carlson (1991).

Analyses of experiments one, two, and bed three (compartment 4) of experiment three showed significant block effects, where bed four (compartment 7a) of experiment three showed none. All three of the significant experiments are contained in compartment four. This would suggest that the soil may be a major source of this variability. This variability was evident throughout the study, including mass and nutrient data.

One of the initial reasons for comparing seedlings of experiment three between compartments 4 and 7a was to observe how fumigation with methyl bromide indirectly affects seedling morphology and nutrient status. Observations of a difference seen in growth between seedlings in compartments 4 and 7a were statistically verified. It was clear from statistical analysis that compartment 7a seedlings grew significantly taller and had larger stem diameters than those seedlings growing in compartment 4 (Fig. 20). This was possibly due to increased photosynthesis as a result of the substantial mycorrhizal infection that was evident throughout compartment 7a. Although a physical measure of mycorrhizal infection was never taken, nursery personnel identified the infection of Thelephora species after observing the mycorrhizal collars.

The outcome of the analysis between compartment 4 and 7a did not yield the expected results. Because compartment 7a was most recently fumigated (October 1987), one would expect there to be much less mycorrhizal infection than in compartment 4. Just the opposite occurred, with compartment 7a showing an abundant amount of infection, and bed three showing little or none. Some possible explanations could be inadequate or inappropriate spraying methods, rapid breakdown of the fumigant or more than likely,

reinfection from the surrounding forested areas. Because ectomycorrhizae reproduces by means of an airborne spore, it would take little time to reinfect the area. Bed three is in a much larger, open area with less forested vegetation close by to rapidly reinfect. Another potential factor is the reduction in competing micro-organisms, giving the mycorrhizae a better opportunity to colonize the pine root system. Additionally, mycorrhizae indirectly protects the seedlings against pathogens. Healthy plants would be better able to resist pests and disease.

An additional reason for making comparisons between beds three and four was to look at how the soil would affect seedling growth. Although analysis showed increased stem caliper and shoot length for seedlings grown in bed four, root length was significantly less than bed three. This was more than likely a result of loss of root tissue upon lifting. The heavier soil in bed four made it difficult to remove the seedling's root system in its entirety, and as a result lower root lengths were obtained.

### Component Mass

Comparison of seedling components as ratios would suggest a source of variation within the beds because of the numerous block effects observed in all experiments in compartment four. Again, the most plausible explanation would be variation within the soil, such as drainage, organic matter, fertility level or even variations in pH. Other factors, such as irrigation, radiation amounts, and nursery practices were applied as uniformly as possible, but could nevertheless result in some variations within the bed.

Significant rate effects were observed for needle:stem weight ratios in experiment one (pre-plant N) and needle:stem weight ratio and needle:root weight ratio in experiment three (top-dress N). N fertilization apparently increases foliage mass more than that of stems and roots.

The location of the bed caused a difference in mass measurements of tissue for top-dress N experiments. Weights for needles and stems were significantly higher for seedlings grown in compartment 7a versus those in compartment 4 (Fig. 21). As with component measurements, this could be attributed to the substantial mycorrhizal infection observed in bed four. There have been numerous studies documenting the effect of mycorrhizae on increased photosynthesis, carbon allocation (root to shoot ratios), and nutrient absorption (concentration) (Reid et al. 1983; Ekwevelam 1984; Campagna and White 1973; Ford et al. 1985).

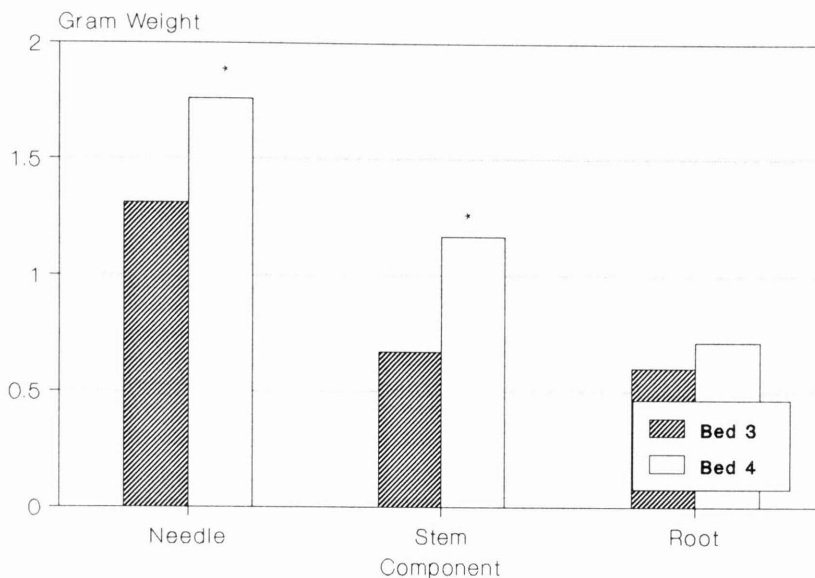


Figure 21. Comparison of mean needle, stem, and root weights between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N (January 1989). Significance at the 95% level is indicated by \*. N=6.

Analysis of ratios between compartments 4 and 7a of experiment three demonstrated that bed location significantly affected six of the seven variables. Two ratios, needle to stem and needle to root were also affected by rate. As with the previous analysis, this can be primarily explained through increased photosynthesis, which in turn increased biomass production. This verifies what Alexander (1977) suggested, that production of mycorrhizae is most vigorous when the roots have a large reserve of carbohydrates, especially following intensive photosynthesis. As more photosynthesizing matter is produced, it increases mycorrhizae which in turn then increases biomass and the cycle continues as long as the conditions are right.

## NUTRITIONAL STATUS

Increases in N concentration of foliage as application of magnesium sulfate increased could be attributed to nutrient translocation from root to stem to foliage. Nitrogen is one of two essential elements absorbed from the soil that is an important constituent of the chlorophyll molecule. As Mg is translocated and chlorophyll is formed, N would be needed to

complete the molecule, and thus be moved up through the shoot and deposited for use. This helps explain the increase in both N foliage and Mg concentrations evident in foliage and stem tissue (Figs. 3&4). Magnesium concentrations were also higher in root tissue as a direct result of applications of magnesium sulfate (Fig. 5).

Applications of magnesium sulfate also had a significant effect on sulfur foliage concentration. This is possibly due to the fact that the S is available in sulfate form, which is readily absorbed from soil. All foliar S levels are well above the deficiency level.

Application of increasing increments of Mg fertilizer yielded no benefit to magnesium foliage concentrations. Just the opposite occurred. The constant rate of application gave consistently higher concentrations for all blocks. This would indicate that higher initial Mg applications allow translocation and accumulation of Mg to occur early on in the seedlings growth cycle. The exponential applications did not permit this to happen, perhaps due to water stress or high soil K levels, which would interfere with Mg uptake.

Increasing N applications in compartment 4 of experiment three increased foliage and stem concentrations of N (Figs. 6&7). This was apparently due to increased uptake and accumulation of N in stem and leaf tissue. Root N and Mg concentrations tended to be affected by block, which could be a manifestation of variations within the soil of bed three 3.

Exponential applications of N in compartment 4 of experiment three increased stem concentrations of K whereas constant rates had no effect. This could be the result of a nitrogen-potassium balance set up through rapid uptake of K prior to uptake of N, that allowed K to accumulate in the stems (uptake tends to increase faster than dry matter production). The accumulated K would help in maintaining structural integrity of cellular components. The constant rate may supply more N than necessary and inhibit K uptake and accumulation in stem tissue (Follett et al. 1981).

Accumulation of Mg in the root tissue increased with exponentially increasing amounts of N fertilizer applied in bed three. Potassium is a monovalent cation that is absorbed in larger quantities by plant roots than any other cation. If K is taken up prior to N (Follett et al 1981), it may inhibit uptake and further translocation of Mg. If Mg is not able to move up the shoot, it will accumulate in the roots. Increases in N will further increase Mg accumulation.

Nitrogen concentrations of foliage, stem and root tissue increased with increasing N applications in compartment 7a of experiment three (Figs. 8-10). The nutrient increase may be due to increased nutrient uptake as a result of mycorrhizal infection. Mycorrhizae are more than likely responsible for increased uptake of P, which increased as N was increased. It's

possible that K was taken up and accumulated in stem and root tissue prior to any major accumulation of N.

Although application of N fertilizer significantly increased tissue N concentrations, exponential additions further increased nutrient levels (Fig.13). This may be due to fertility levels that are better able to match the growth rate of the seedlings. As the seedling increases in biomass, the tissue concentrations are not diluted, but are maintained at the same level.

As was seen in compartment 4, Mg root and K stem and root concentrations were affected by the increasing rate of fertility more so than the constant rate. Stem tissue tends to be a sink for K to establish development of lignin and cellulose for strength and stiffness in plant tissue. Magnesium translocation to other tissue was probably inhibited by K.

Comparisons between seedlings in compartment 4 and 7a revealed that stem P, root and stem N, root Mg, and stem and root K were all significantly affected by bed location (Figs. 14-19). This may be an indication that mycorrhizal infection can have a varied effect on seedling fertility. It has been documented that mycorrhizal development is pronounced in soil low in P and N (Alexander 1977). It may also be a manifestation of the existing condition of the soil. In an analysis performed the previous year, compartment 4 was determined to have higher levels of P than compartment 7a, while compartment 7a had higher Mg and K levels than compartment 4. The most plausible explanation would be the dilution of nutrients in plant tissue of the seedlings in compartment 7a due to increased biomass production. The bed\*rate interactions observed in this analysis emphasizes the difference between the beds. The amount of infection displayed visually was reiterated through nutrient concentration differences verified through statistical analysis.

Overall, tissue nutrient concentrations for all experiments were above recognized critical levels. Nutrient levels of N, P, Mg, and Ca foliage and stems were optimal or higher. K foliage, stem, and roots, P foliage and roots, as well as Ca roots were found to be at the lower acceptable limits for southern pines.

## SURVIVAL AND GROWTH

An additional study to evaluate the effects of varying amounts of N fertilization in the nursery was performed by Texas Forest Service personnel (Barber et al. 1990). Seedlings raised under different pre-plant and top-dress N fertilization regimes were outplanted on a poor site and a good site in east Texas. First year field performance showed that top-dress fertilization significantly increased survival on the poor site, but had little effect on the good site. Survival of pre-plant fertilized seedlings was not affected at either site. Exponential

applications on the poor site resulted in the greatest survival of all the top-dress treatments, but there was no significant difference between the exponential and equivalent constant rate.

First year growth was significantly affected at both sites. Much of the growth performance was attributed to initial size of the seedling. Larger seedlings from the nursery performed better in the field. Exponentially increasing fertilizer applications resulted in better first year performance than equivalent constant applications on the poor site, whereas the equivalent constant rate resulted in slightly better results on the good site. Pre-plant N applications resulted in a slightly negative effect on first year performance with increasing application rates.



## CHAPTER VII

### CONCLUSIONS

This study confirms the positive effects of certain practices affecting loblolly pine seedling growth and development in nursery production. Individually, mycorrhizal infection, higher fertilizer applications, and exponentially increasing fertilizer applications improved many seedling attributes. Mycorrhizal infection combined with elevated N applications increased tissue nutrient levels, but increased biomass production so much that seedling height would not allow for effective planting. Magnesium applied exponentially yielded no increase in Mg tissue concentration, but did increase with exponentially increasing N. There was a connection between Mg and N fertility, but further study is necessary to evaluate or confirm this. There was little response due to fertilization for morphological traits or tissue nutrient levels of P, Ca, and Mg. But, this should not eliminate their consideration in combination with current nursery practices.

Based upon these results, hypotheses two, three and four are accepted, and hypotheses one is rejected. Pre-plant N applications resulted in no statistically significant differences. Levels of pre-plant N fertilizer were low enough to prevent high incidence of damping-off, but may also have been too low to produce any discernable differences in plant tissue. Hence, hypothesis one cannot be accepted. Hypotheses two and three, in which application of top-dress Mg and N resulted in significant increases of numerous plant nutrient levels in all three plant components, are therefore accepted. Comparison of nutrient levels and plant morphological measures between compartments 4 and 7a of experiment three showed some significance, especially with respect to biomass accumulation and hypothesis four was therefore accepted. However, none could be attributed directly to fumigation practices.

In order for an artificial reforestation program to be successful, both the nursery manager and the regeneration forester must consistently strive for parallel goals. Together with other personnel such as geneticists, field foresters, and tree planters, nursery personnel can develop a target seedling designed in detail. After the regeneration forester ascertains the source of seed and the desired seedling characteristics, the nursery manager may then produce the specified seedling. Through studies such as this, the nursery manager would be able to pinpoint individual seedling traits, such as taller seedlings where competitive vegetation would be a factor after outplanting, and modify those seedlings accordingly.

In general, all seedlings benefit from being nutritionally balanced, but in some instances, there may be a need for further manipulation of those nutrients. In cases where the seedlings will be planted into a harsh (droughty) site, for example, potassium levels could be critical. Additionally, other concerns, such as time of planting, soil type, site preparation, and planting technique must be considered.

Altering seedling architecture to improve seedling survival and subsequent growth after outplanting requires input from all levels of production. Implementing an effective program will demand that considerations beyond their control need to be factored into the scheme. Some of the largest considerations are; amount of sunlight, growing degree days, temperature, late springs, early falls, and soil structure. These will all play a role in the formation of seedlings. Longer term experiments may allow scientists to more specifically manipulate seedlings through practices in the nursery.

Specific recommendations to personnel at Texas Forest Service Indian Mound Nursery would include increased application of N(34-67 kg/ha), perhaps exponentially applied. In order to eliminate the dilution effect observed in this study, nursery personnel could manipulate biomass accumulation through reduced irrigation. Seedlings grown in the newer parcels that exhibited tremendous height and diameter growth could also be controlled through moisture management.

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**APPENDIX A**

EXPECTED MEAN SQUARES FOR ALL (JANUARY 1989)

EXPERIMENTS

Expected mean squares for one growing season for weight, weight ratio, component, and nutritional analyses of loblolly pine seedlings from all beds grown at Indian Mound Nursery.

Source of Variation	Degrees of Freedom	Expected Mean Square
Block (B)	b-1	$\sigma^2 + r\sigma_b^2$
Rate (R)	r-1	$\sigma^2 + b\sigma_r^2$
B*R	(b-1)(r-1)	$\sigma^2$

Model statement:

$$Y_{ij} = \mu + \alpha_i + \beta_j + \epsilon_{ij}$$

Where:  $Y_{ij}$  = observation of variable for treatment( $\alpha_i$ ) in block( $\beta_j$ )

$\mu$  = overall mean for all observations

$\alpha$  = effect due to treatment( $\alpha_i$ )

$\beta$  = effect due to block( $\beta_j$ )

$\epsilon$  = random error for  $\alpha_i$  in  $\beta_j$

Variable observed:

Component weights - mean needle wt. mean stem wt. mean root wt.

Weight ratios - mean top wt. mean stem wt. mean needle wt. mean root wt. mean needle:stem wt. mean top:root wt. mean needle:root wt.

Component measures - mean caliper mean shoot length mean root length

Nutrient concentration - mean weight percent

Nutrient concentration - mean weight percent

Expected mean squares for one growing season for weight, weight ratio, component, and nutritional analyses between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N applications.

Source of Variation	Degrees of Freedom	Expected Mean Square
Bed(A)	a-1	$\sigma^2 + r\sigma_{ab}^2 + br\sigma_a^2$
Block(block)(B)	a(b-1)	$\sigma^2 + r\sigma_{ab}^2$
Rate(R)	r-1	$\sigma^2 + ab\sigma_r^2$
Bed*Rate	(a-1)(r-1)	$\sigma^2 + b\sigma_{ar}^2$
Block*Rate(block)	a(b-1)(r-1)	$\sigma^2$

Model statement:

$$Y_{ijk} = \mu + \gamma_i + \beta_{ij} + \alpha_k + \delta_{ik} + \epsilon_{ijk}$$

Where:  $Y_{ijk}$  = observation of variable for bed( $\gamma$ )<sub>i</sub> in block( $\beta$ )<sub>j</sub> of treatment( $\alpha$ )<sub>k</sub>

$\mu$  = overall mean for all observations

$\gamma$  = effect due to bed( $\gamma$ )<sub>i</sub>

$\beta$  = effect due to block( $\beta$ )<sub>j</sub>

$\alpha$  = effect due to treatment( $\alpha$ )<sub>k</sub>

$\delta$  = effect due to interaction of bed( $\gamma$ )<sub>i</sub> and treatment( $\alpha$ )<sub>k</sub>

$\epsilon$  = random error for  $\gamma_i$  in  $\beta_j$  of  $\alpha_k$

Variable observed:

Component weights - mean needle wt. mean stem wt. mean root wt.

Weight ratios - mean top wt. mean stem wt. mean needle wt. mean root wt. mean needle:stem wt. mean top:root wt. mean needle:root wt.

Component measures - mean stem caliper mean shoot length mean root length

Nutrient concentration - mean weight percent

**APPENDIX B**

STATISTICAL SIGNIFICANCE FOR ALL (JANUARY 1989)  
EXPERIMENTS

Summary of statistical significance for loblolly pine seedlings(January 1989) receiving pre-plant applications of N(bed 1). Significance at the 95% level is indicated by \*. ns = not significant.

Attribute	Block	Rate	Block*Rate
N foliage	ns	ns	ns
N stem	ns	ns	ns
N root	ns	ns	ns
P foliage	ns	ns	ns
P stem	ns	ns	ns
P root	ns	ns	ns
Ca foliage	ns	ns	ns
Ca stem	ns	ns	ns
Ca root	ns	ns	ns
Mg foliage	ns	ns	ns
Mg stem	ns	ns	ns
Mg root	ns	ns	ns
K foliage	ns	ns	ns
K stem	ns	ns	ns
K root	ns	ns	ns
Shoot length	ns	ns	ns
Stem caliper	ns	ns	ns
Root length	*	ns	ns
Needle weight	ns	ns	ns
Stem weight	ns	ns	ns
Root weight	ns	ns	ns
Shoot:root	ns	ns	ns
Needle:stem	ns	*	ns
Needle:root	ns	ns	ns

Summary of statistical significance for loblolly pine seedlings(January 1989) receiving top-dress applications of Mg(bed 2). Significance at the 95% level is indicated by \*. ns = not significant.

Attribute	Block	Rate	Block*Rate
N foliage	*	*	ns
N stem	*	ns	ns
N root	ns	ns	ns
P foliage	ns	ns	ns
P stem	ns	ns	ns
P root	ns	ns	ns
Ca foliage	ns	ns	ns
Ca stem	ns	ns	ns
Ca root	ns	ns	ns
Mg foliage	ns	*	ns
Mg stem	ns	*	ns
Mg root	*	*	ns
K foliage	ns	ns	ns
K stem	ns	ns	ns
K root	ns	ns	ns
S foliage	ns	*	ns
Shoot length	*	ns	ns
Stem caliper	ns	ns	ns
Root length	ns	ns	ns
Shoot weight	ns	ns	ns
Needle weight	ns	ns	ns
Stem weight	ns	ns	ns
Root weight	ns	ns	ns
Shoot:root	*	ns	ns
Needle:stem	ns	ns	ns
Needle:root	*	ns	ns

Summary of statistical significance for loblolly pine seedlings(January 1989) receiving top-dress applications of N(bed 3). Significance at the 95% level is indicated by \*. ns = not significant.

Attribute	Block	Rate	Block*Rate
N foliage	ns	*	ns
N stem	*	*	ns
N root	*	ns	ns
P foliage	ns	ns	ns
P stem	ns	ns	ns
P root	ns	ns	ns
Ca foliage	ns	ns	ns
Ca stem	ns	ns	ns
Ca root	ns	ns	ns
Mg foliage	ns	ns	ns
Mg stem	ns	ns	ns
Mg root	*	ns	ns
K foliage	ns	ns	ns
K stem	ns	ns	ns
K root	ns	ns	ns
Shoot length	*	ns	ns
Stem caliper	*	ns	ns
Root length	*	ns	ns
Needle weight	*	ns	ns
Shoot weight	*	ns	ns
Stem weight	*	ns	ns
Root weight	ns	ns	ns
Shoot:root	ns	ns	ns
Needle:stem	ns	*	ns
Needle:root	ns	*	ns

Summary of statistical significance for loblolly pine seedlings(January 1989) receiving top-dress applications of N(bed 4). Significance at the 95% level is indicated by \*. ns = not significant.

Attribute	Block	Rate	Block*Rate
N foliage	*	*	ns
N stem	ns	*	ns
N root	ns	*	ns
P foliage	*	*	ns
P stem	ns	ns	ns
P root	ns	ns	ns
Ca foliage	ns	ns	ns
Ca stem	ns	ns	ns
Ca root	ns	ns	ns
Mg foliage	ns	ns	ns
Mg stem	ns	ns	ns
Mg root	ns	ns	ns
K foliage	ns	ns	ns
K stem	ns	*	ns
K root	ns	*	ns
Shoot length	ns	ns	ns
Stem caliper	ns	ns	ns
Root length	ns	ns	ns
Needle weight	ns	*	ns
Shoot weight	ns	ns	ns
Stem weight	ns	ns	ns
Root weight	ns	ns	ns
Shoot:root	ns	ns	ns
Needle:stem	ns	ns	ns
Needle:root	ns	ns	ns

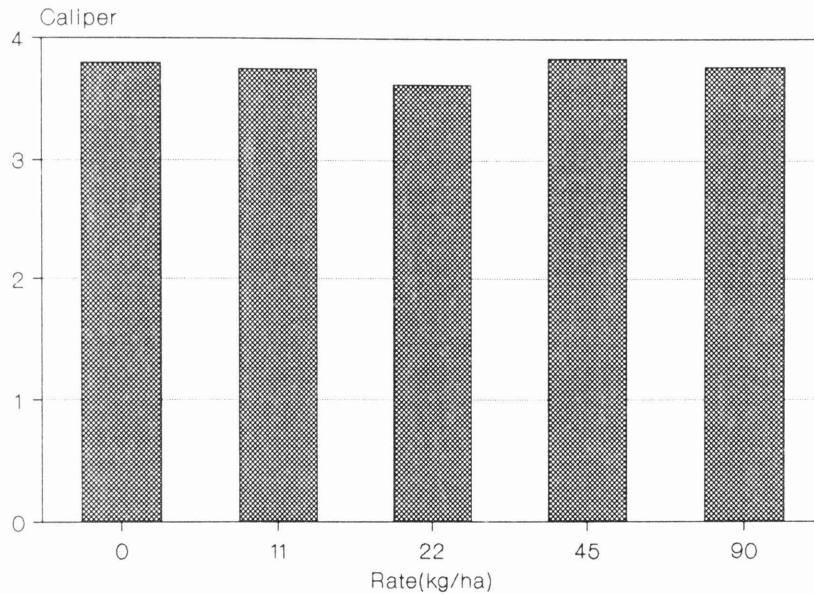


Summary of statistical significance between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings(January 1989) receiving top-dress applications of N. Significance at the 95% level is indicated by \*. ns = not significant.

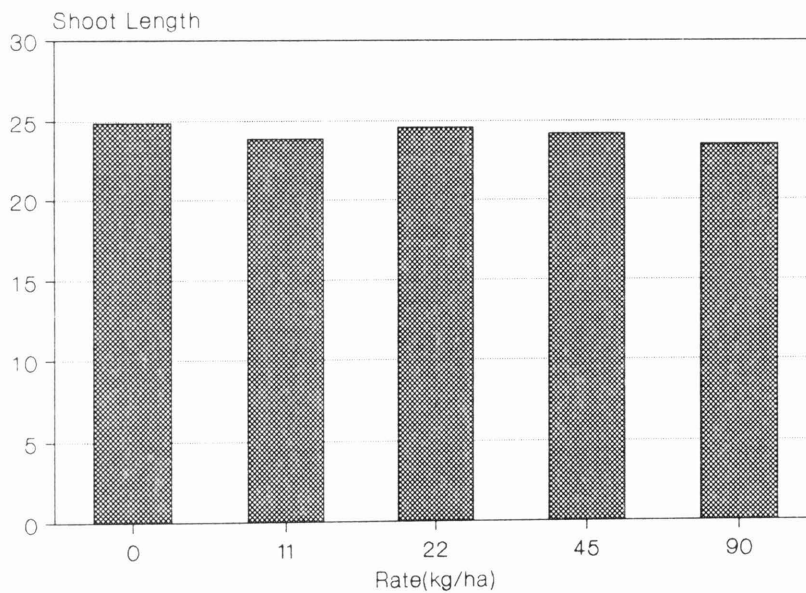
Attribute	Block(bed)	Bed	Bed*Rate
N foliage	*	ns	ns
N stem	ns	*	*
N root	*	*	ns
P foliage	*	ns	ns
P stem	*	*	ns
P root	*	ns	*
Ca foliage	ns	ns	ns
Ca stem	*	ns	ns
Ca root	*	ns	ns
Mg foliage	*	ns	ns
Mg stem	ns	ns	ns
Mg root	*	*	ns
K foliage	ns	ns	ns
K stem	ns	*	ns
K root	*	*	ns
Shoot length	ns	*	ns
Stem caliper	*	*	ns
Root length	*	ns	ns
Needle weight	*	*	ns
Shoot weight	*	*	ns
Stem weight	*	*	ns
Root weight	*	ns	ns
Shoot:root	ns	*	ns
Needle:stem	ns	*	ns
Needle:root	ns	*	ns

**APPENDIX C**

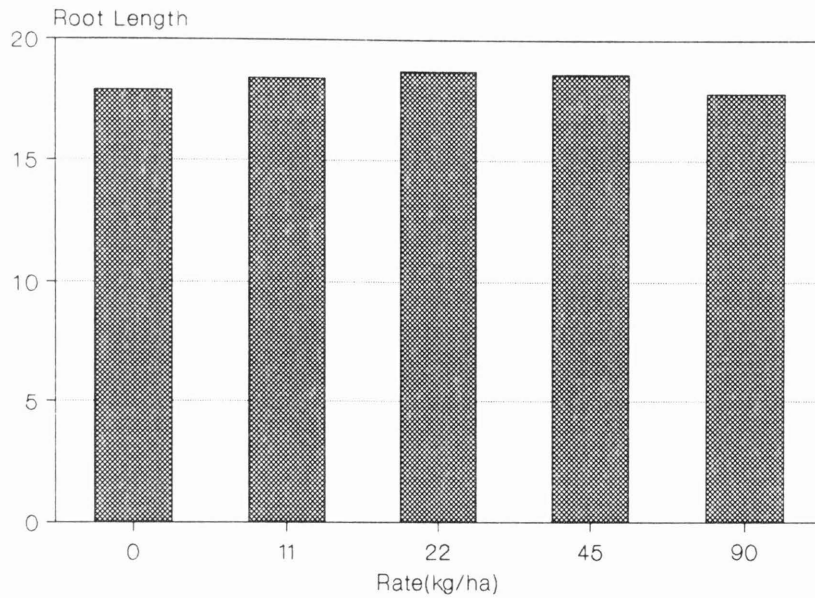
GRAPHICAL REPRESENTATION OF JANUARY 1989 DATA  
FROM ALL EXPERIMENTS



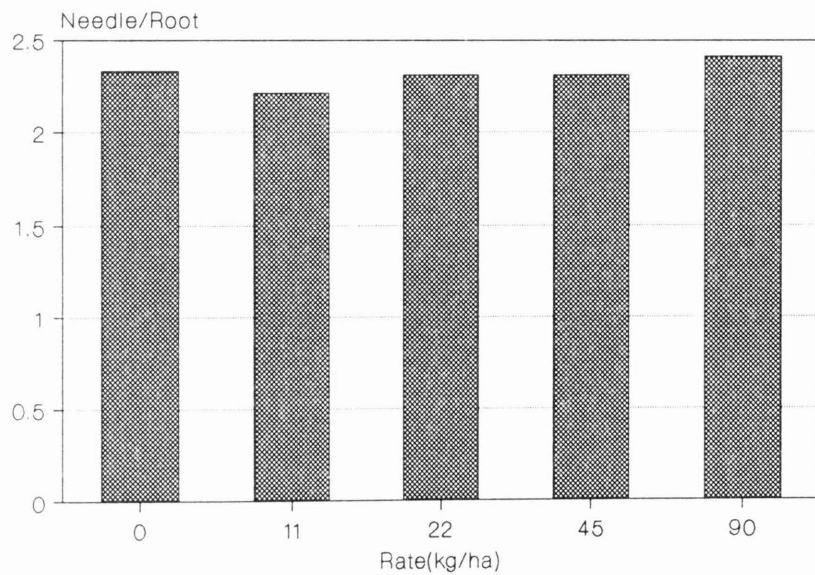
Mean stem caliper(mm) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



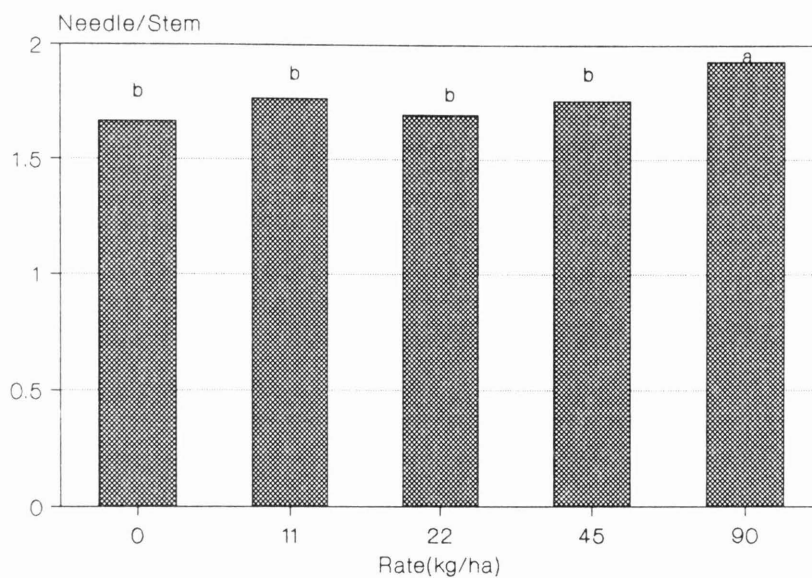
Mean shoot length(cm) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



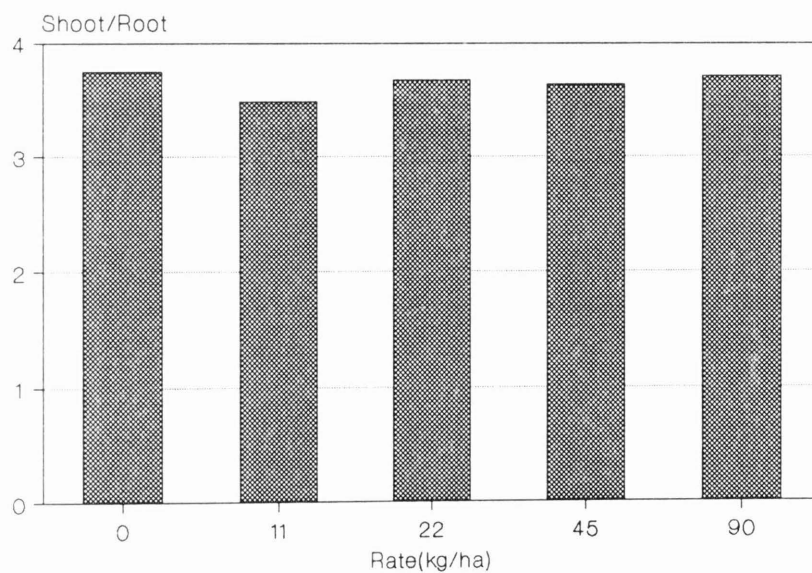
Mean root length(cm) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer (bed 1). N=4.



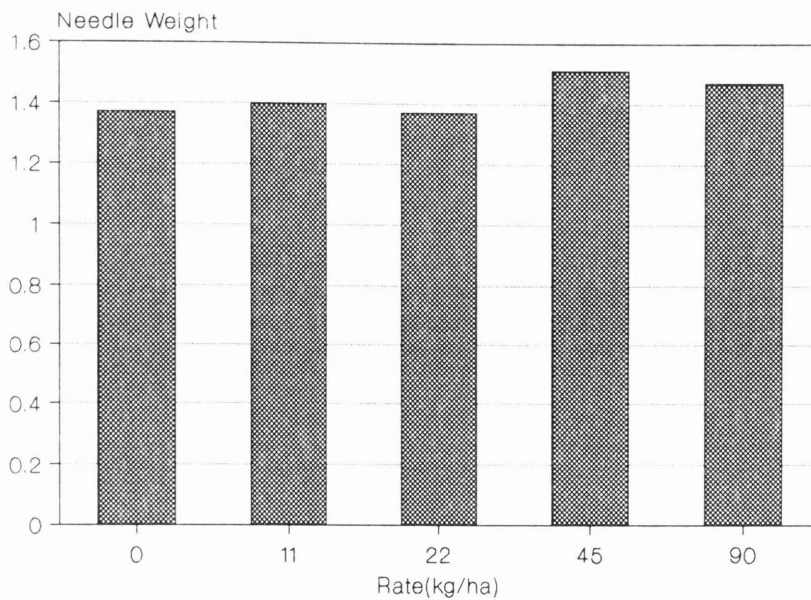
Mean needle:root ratio of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer (bed 1). N=4.



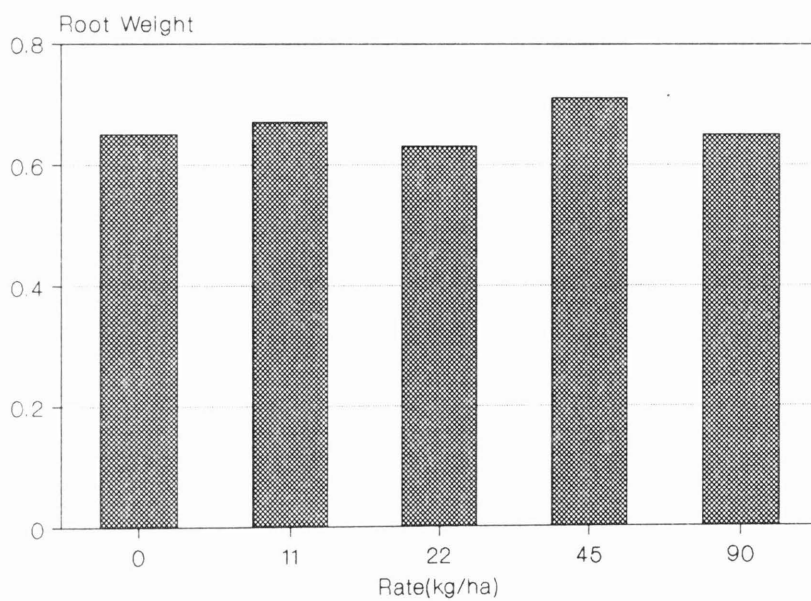
Mean needle:stem ratio of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



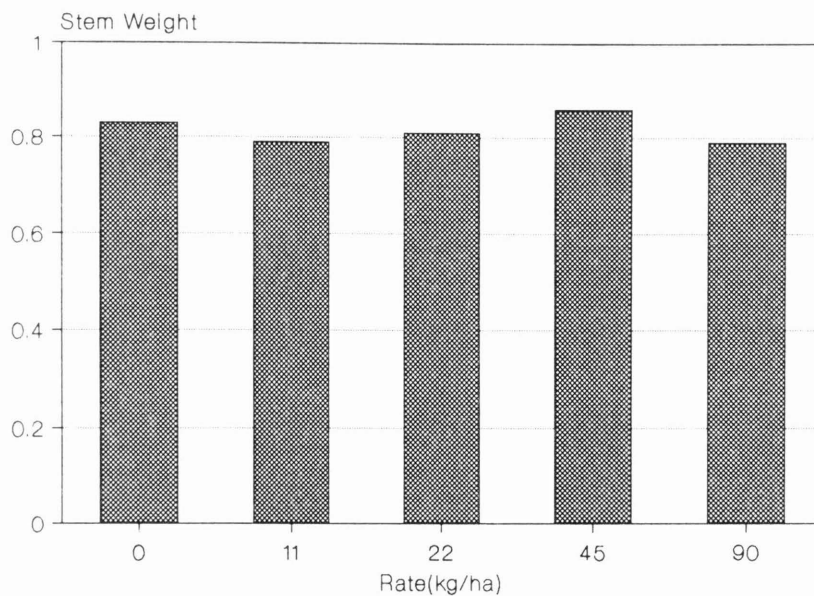
Mean shoot:root ratio of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



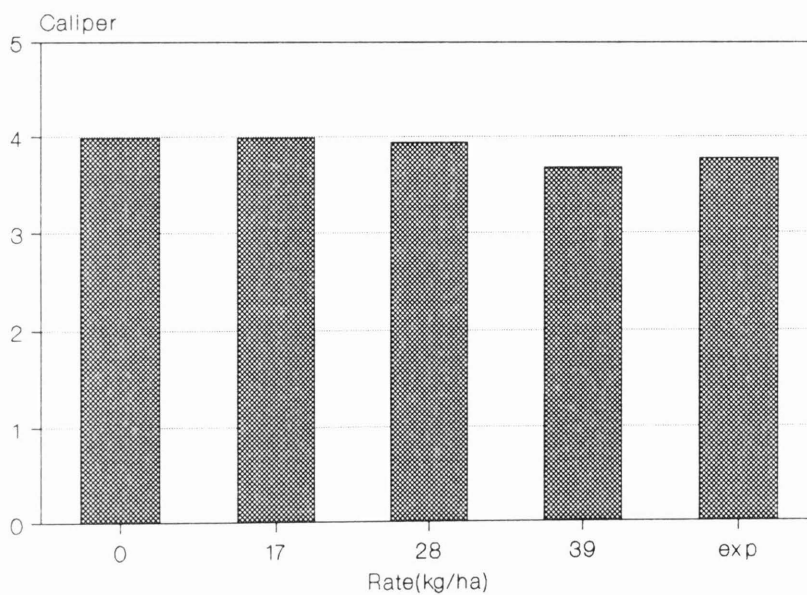
Mean needle weight(gm) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



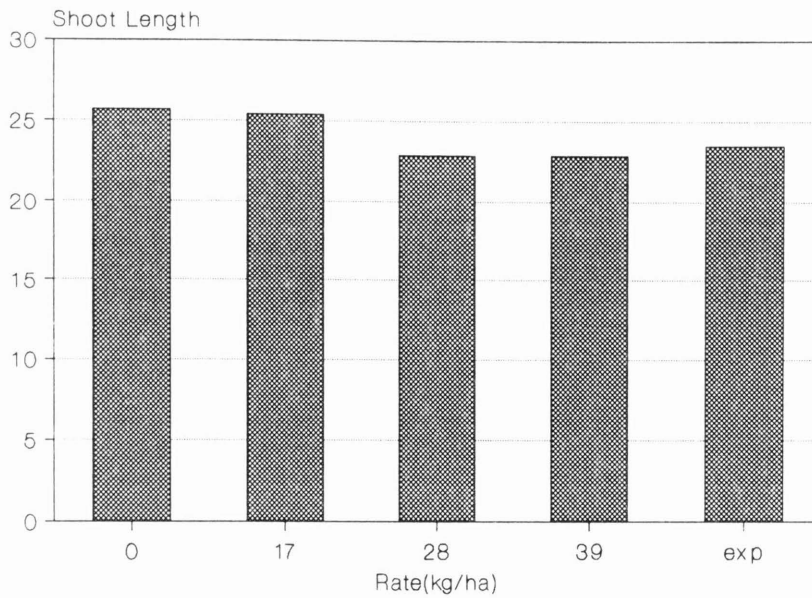
Mean root weight(gm) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



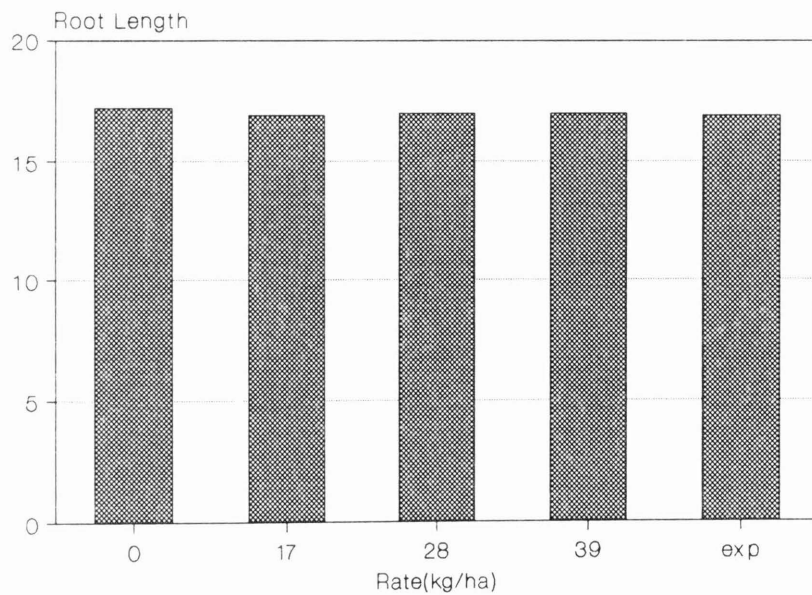
Mean stem weight(g) of loblolly pine seedlings receiving varying amounts of pre-plant N fertilizer(bed 1). N=4.



Mean stem caliper(mm) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer(bed 2). N=4.

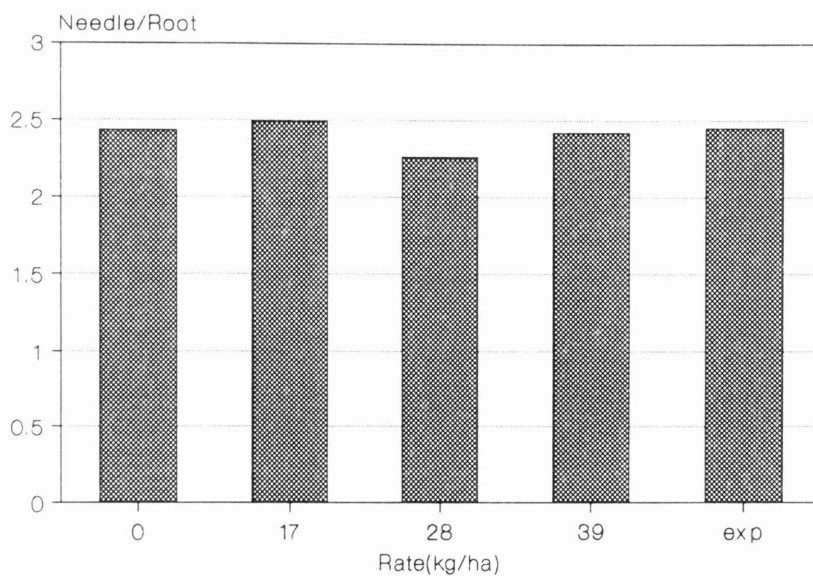


Mean shoot length(cm) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.

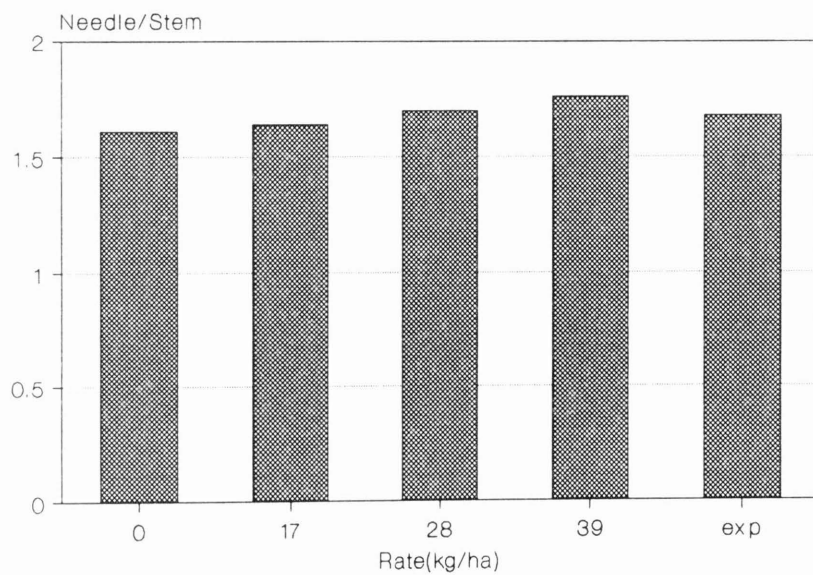


Mean root length(cm) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.

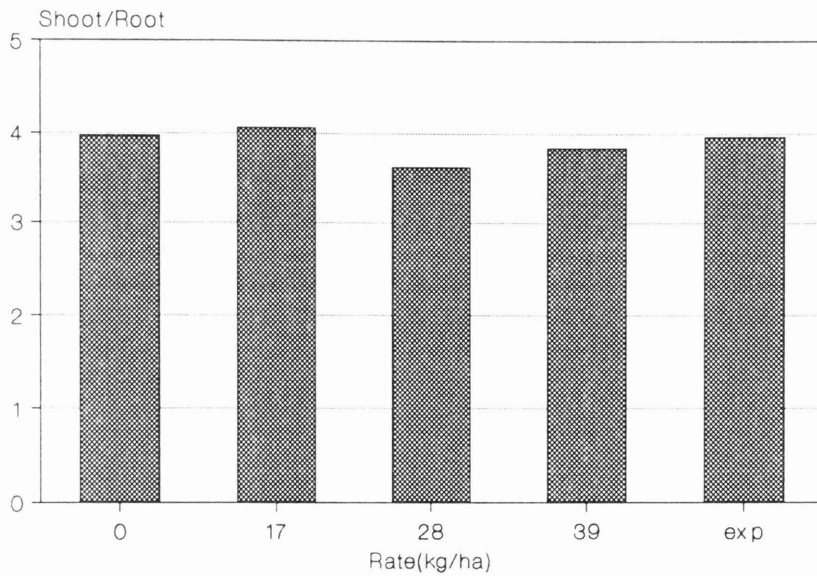




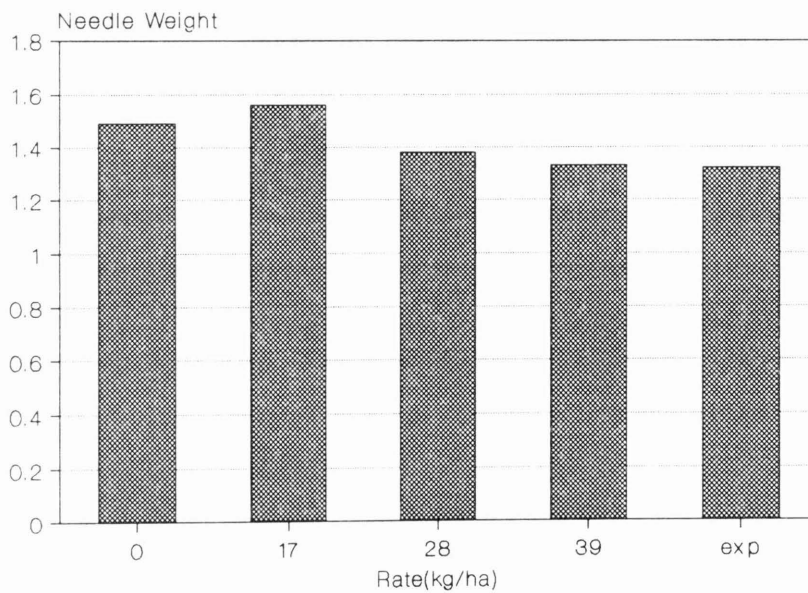
Mean needle:root ratio of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer(bed 2). N=4.



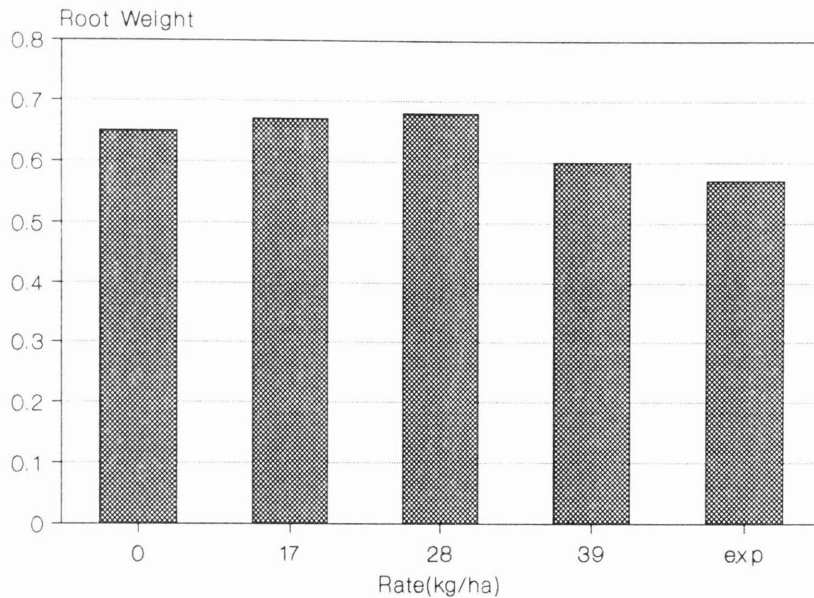
Mean needle:stem ratio of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer(bed 2). N=4.



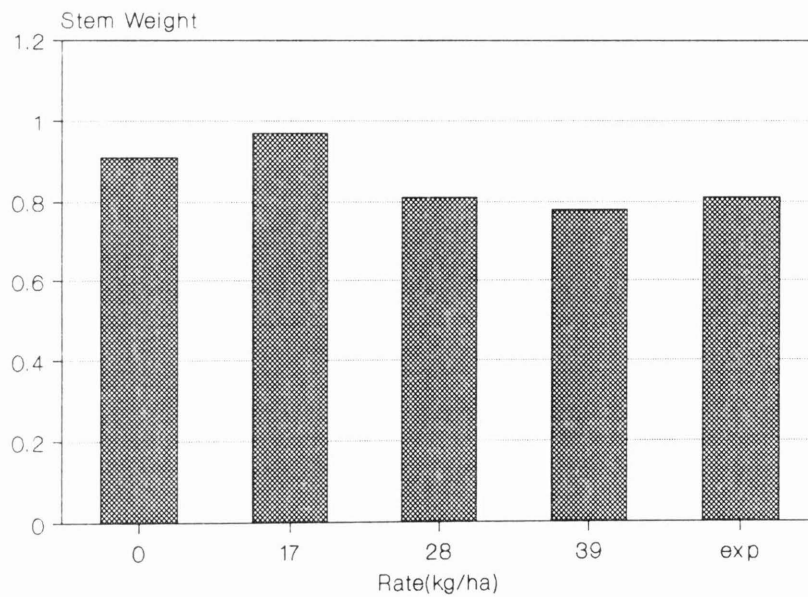
Mean shoot:root ratio of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.



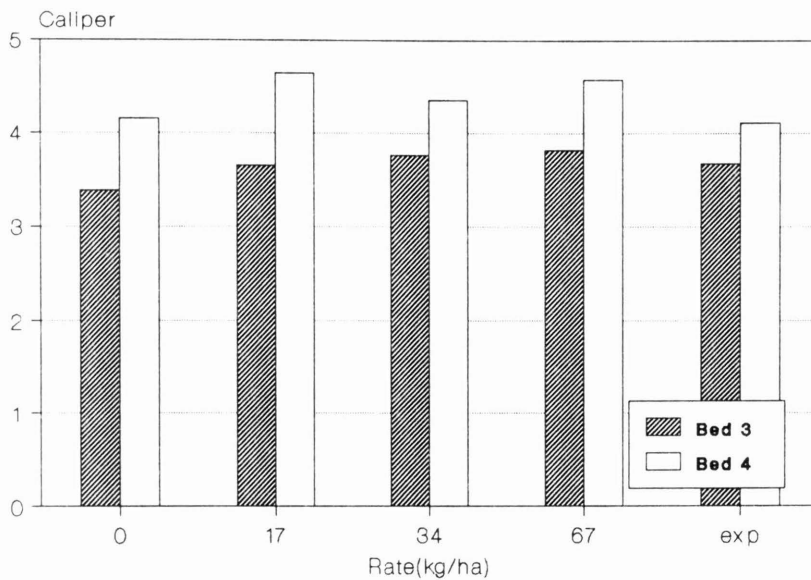
Mean needle weight (g) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.



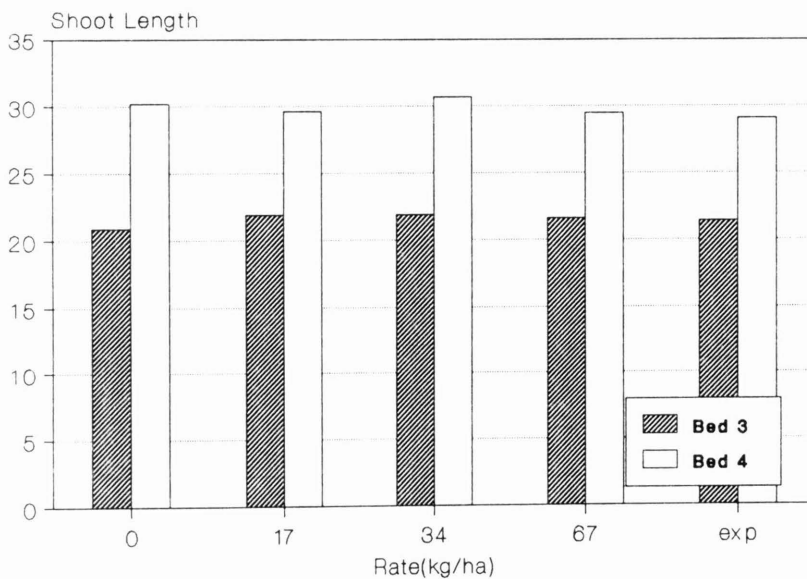
Mean root weight(g) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.



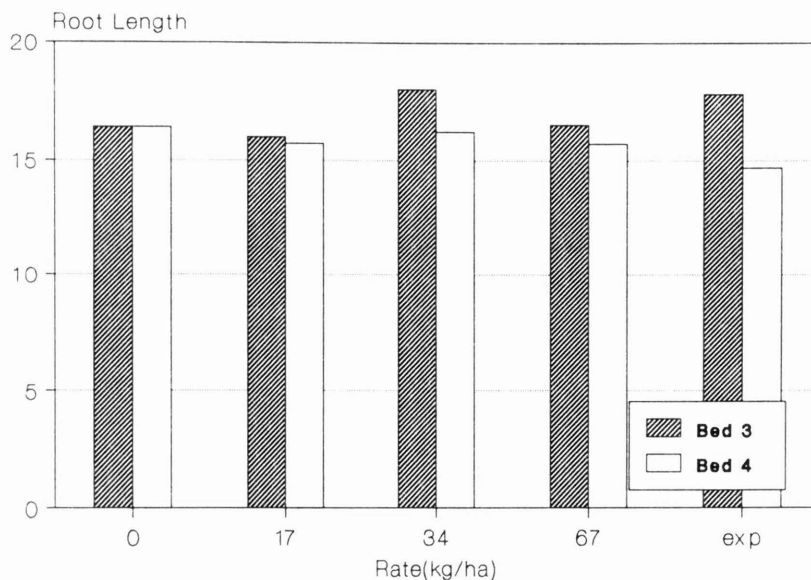
Mean stem weight(g) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer (bed 2). N=4.



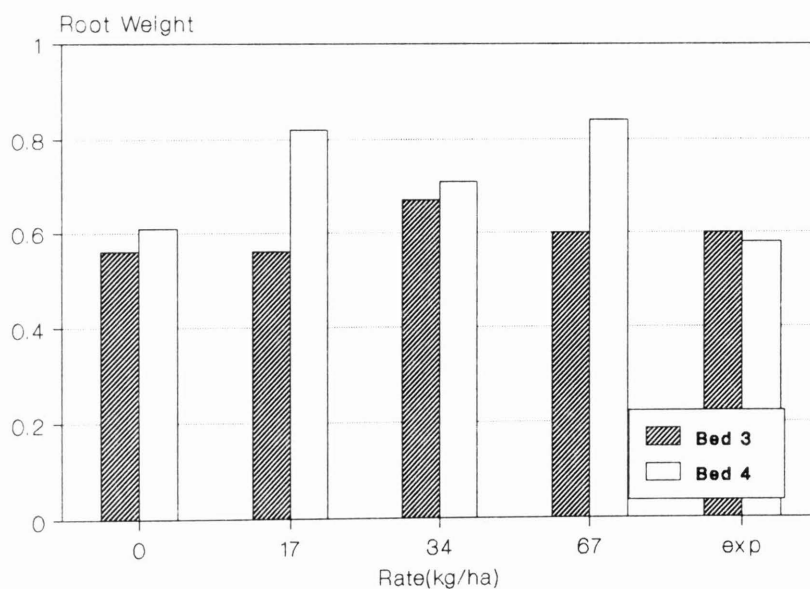
Comparison of mean stem caliper(mm) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



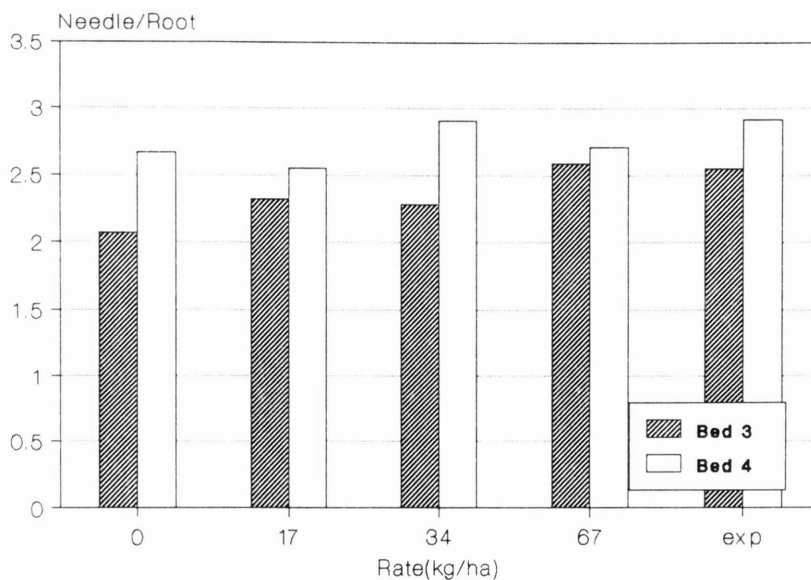
Comparison of mean shoot length(cm) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



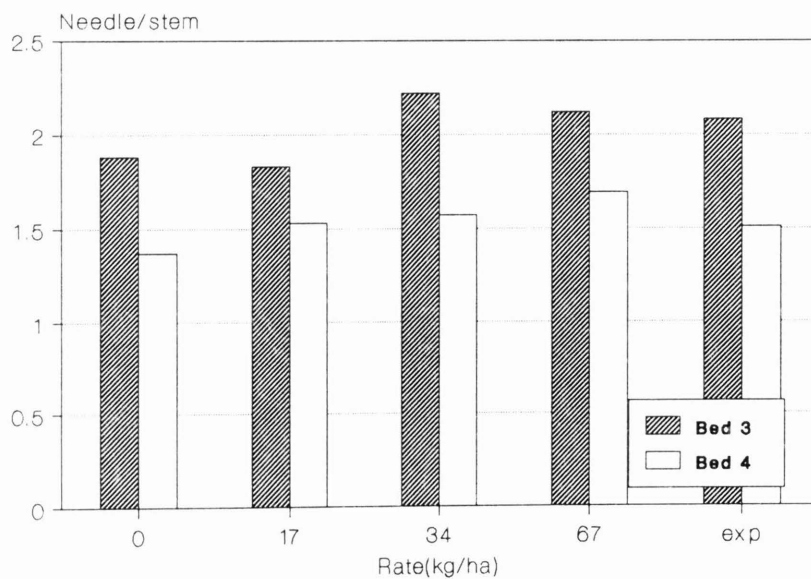
Comparison of mean root length(cm) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



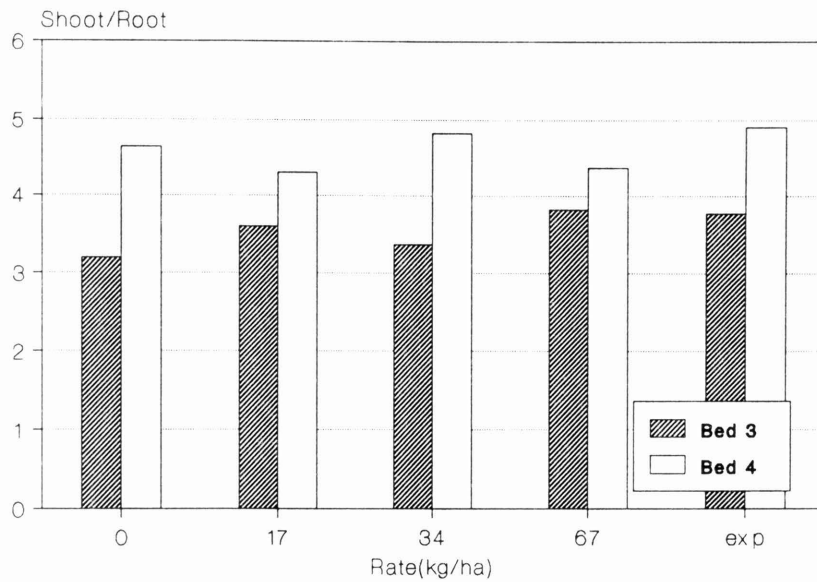
Comparison of mean root weight(g) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



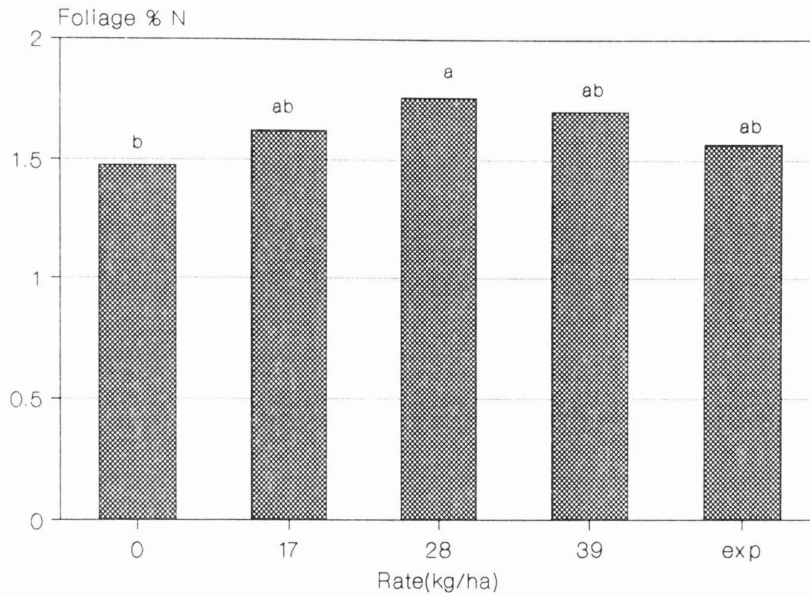
Comparison of mean needle:root ratio between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



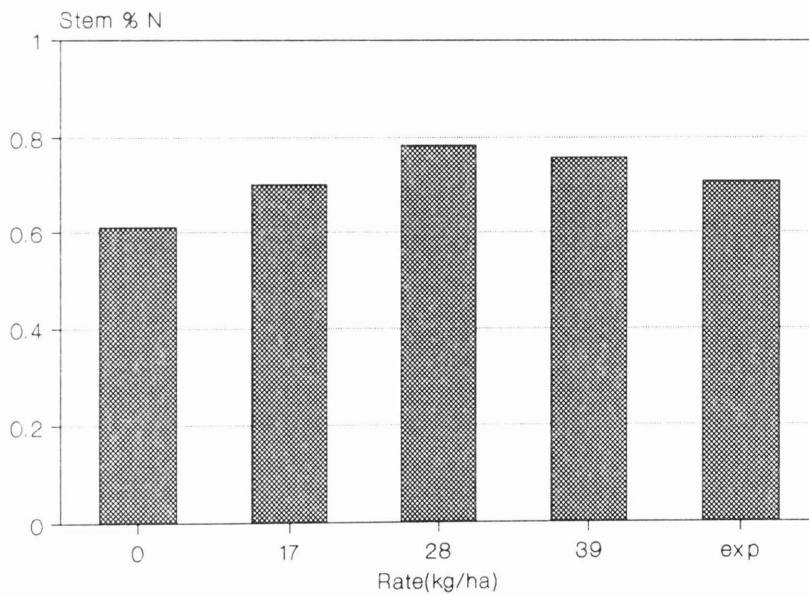
Comparison of mean needle:stem ratio between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.



Comparison of mean shoot:root ratio between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress applications of N. N=6.

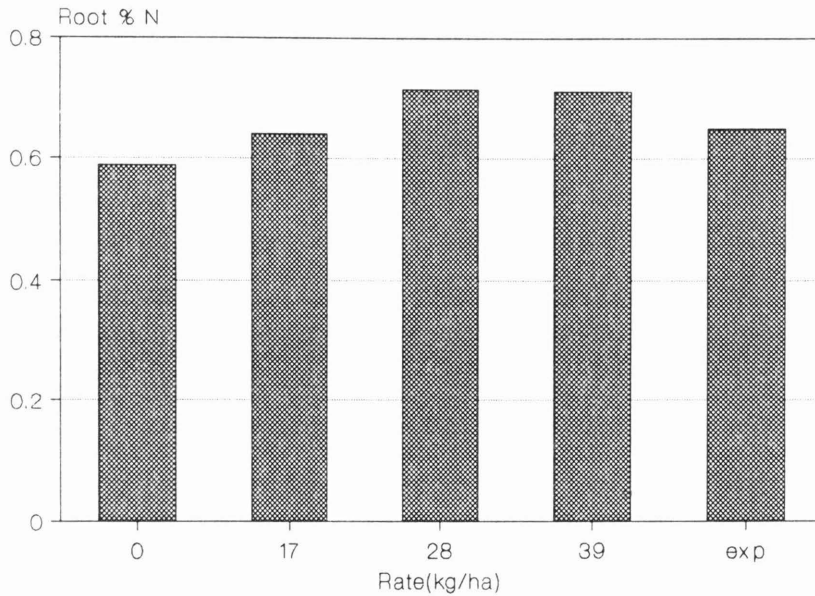


Mean foliar N concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. Columns with same letter not significantly different at the .05 level. N=8.

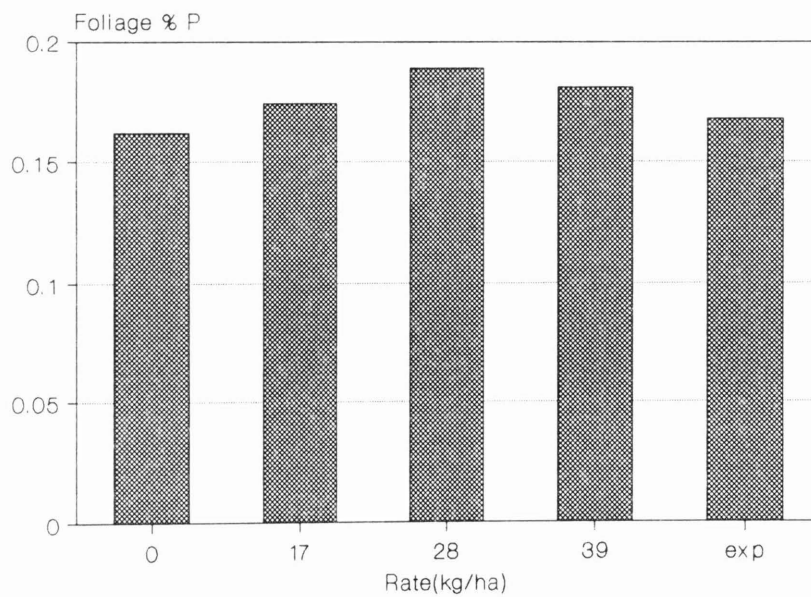


Mean stem N concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. N=8.

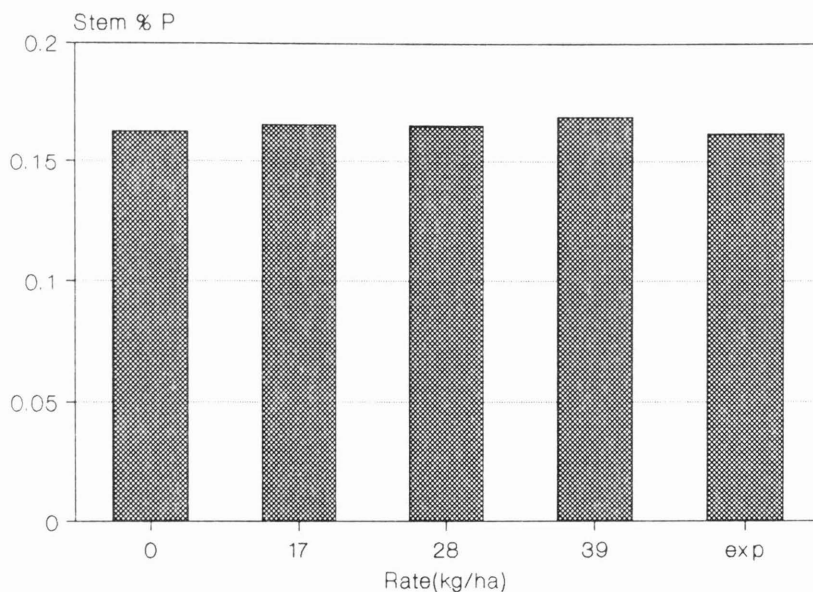




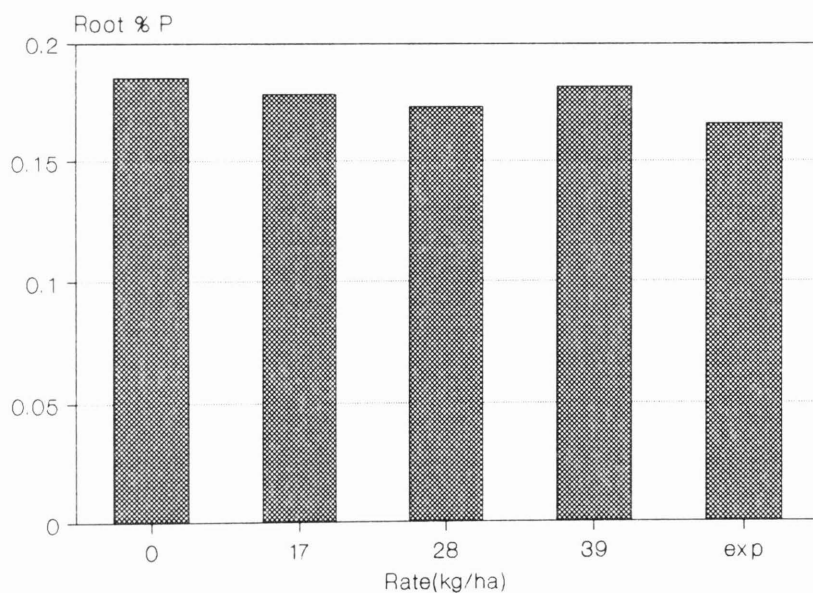
Mean root N concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. N=8.



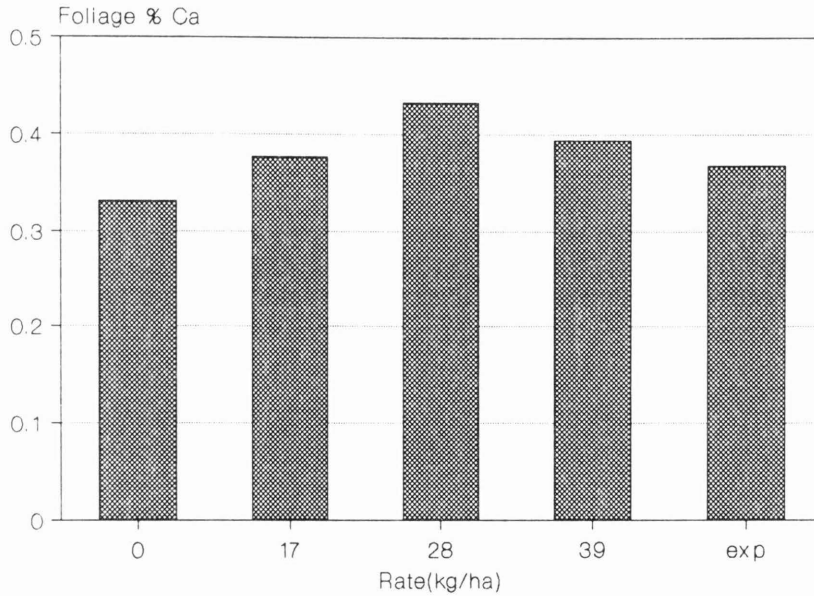
Mean foliar P concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. N=8.



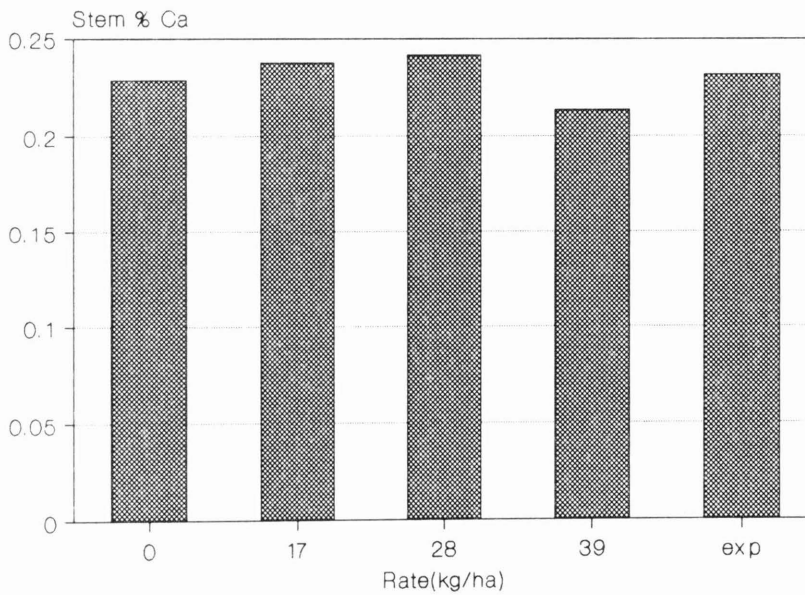
Mean stem P concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. N=8.



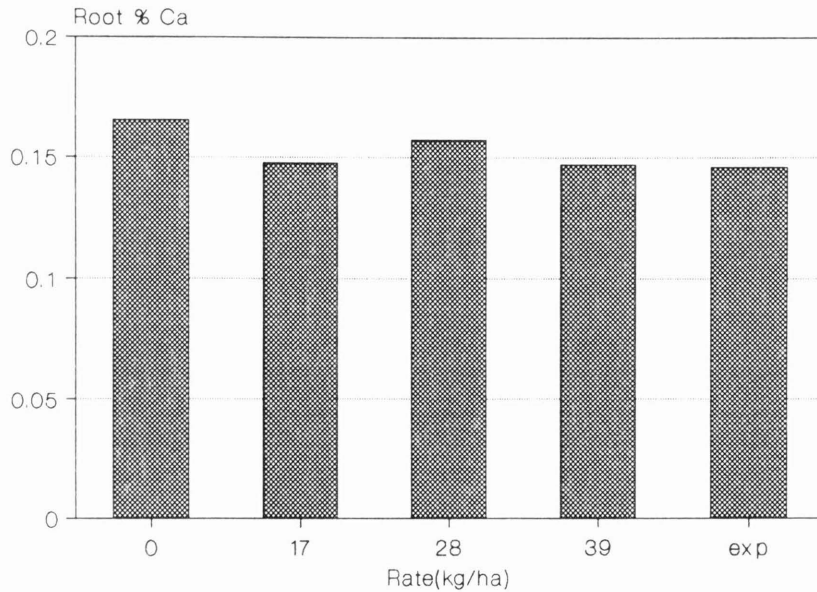
Mean root P concentration(weighted %) of loblolly pine seedlings receiving varying amounts of top-dress Mg fertilizer. N=8.



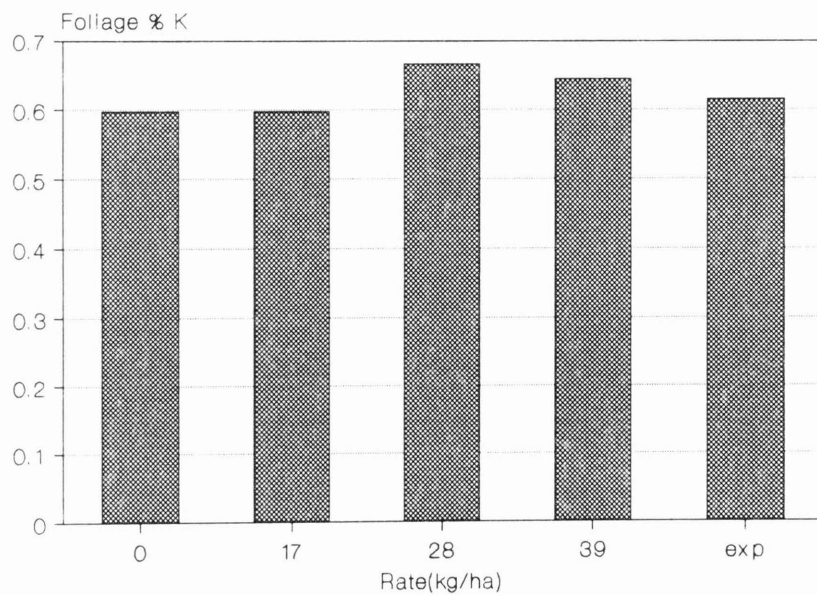
Mean foliar Ca concentration (weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



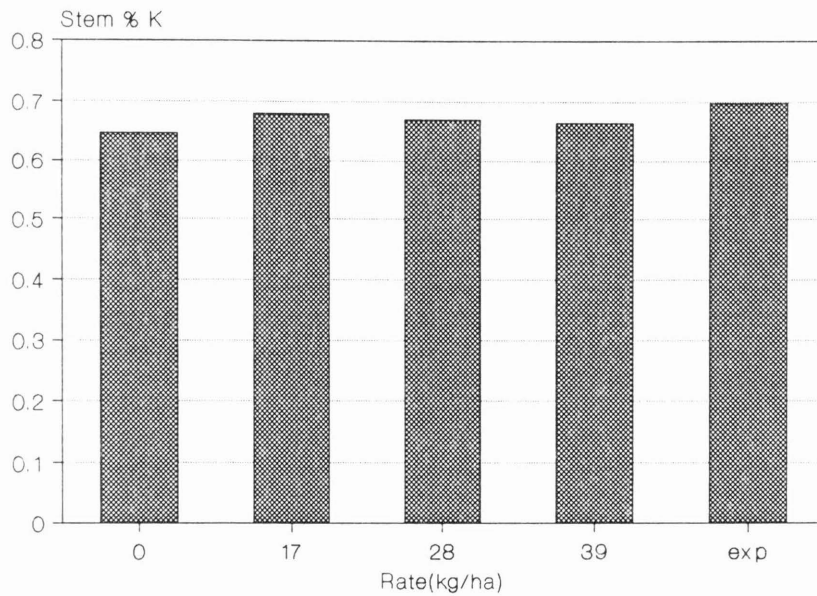
Mean stem Ca concentration (weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



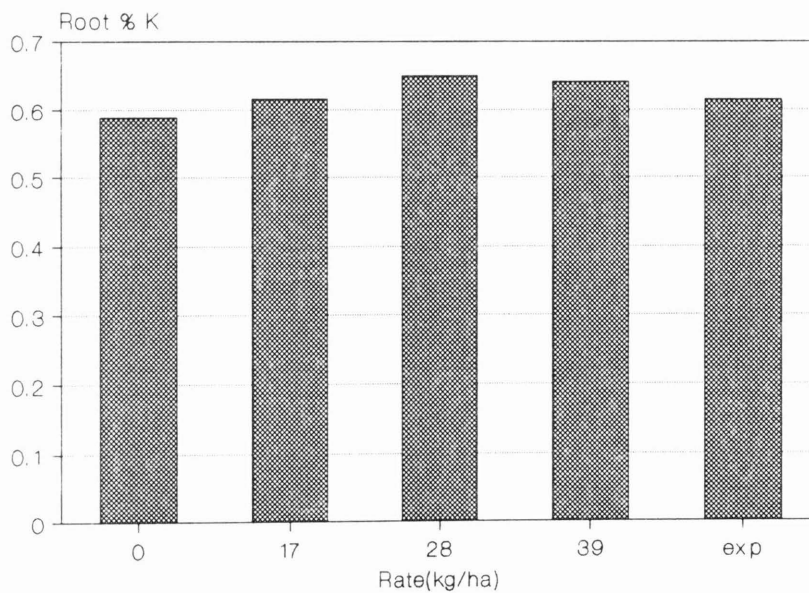
Mean root Ca concentration(weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



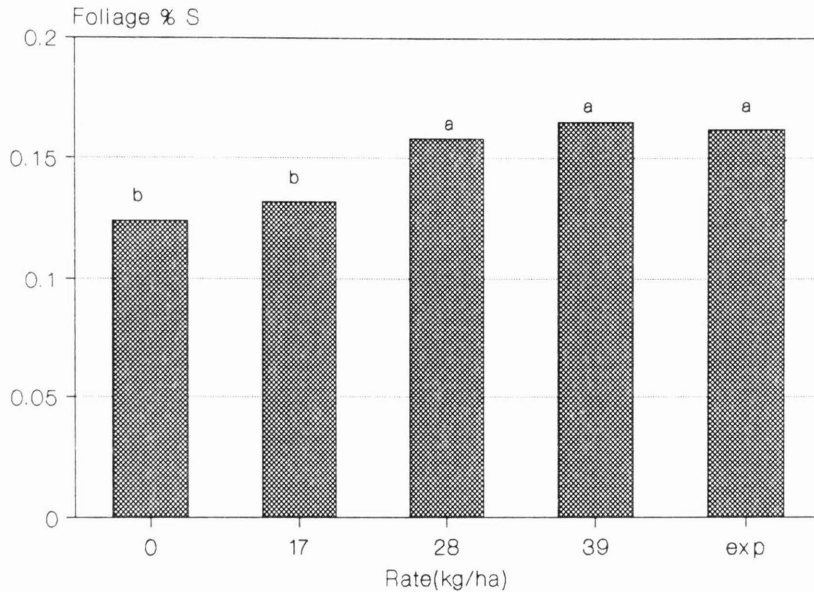
Mean foliar K concentration(weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



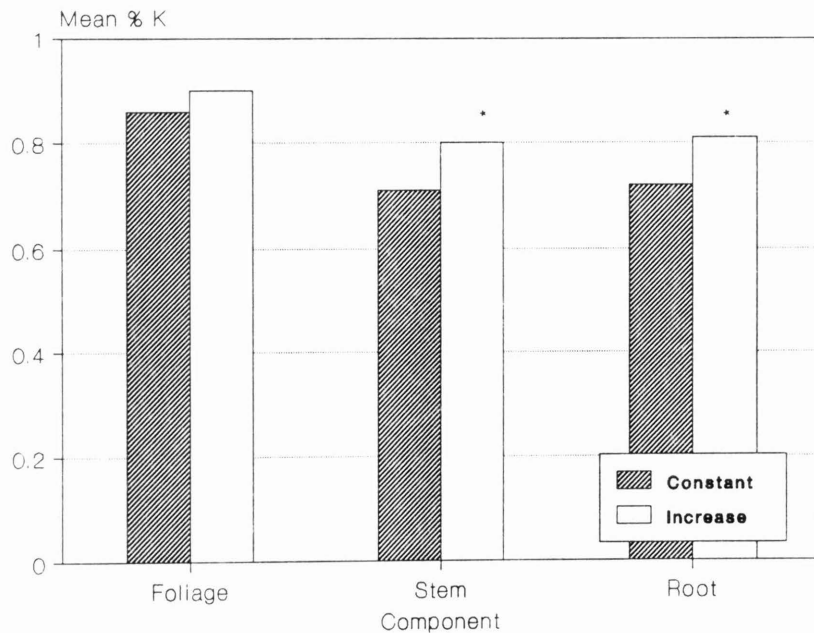
Mean stem K concentration(weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



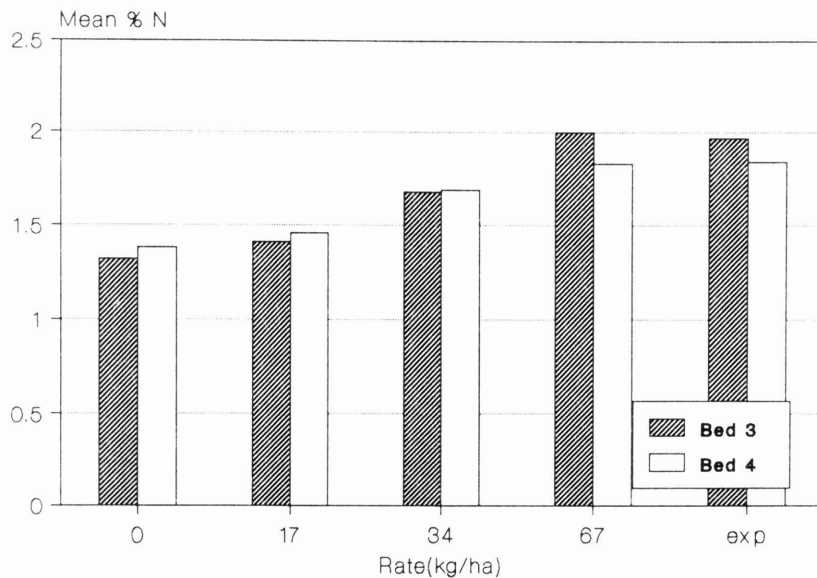
Mean root K concentration(weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. N=8.



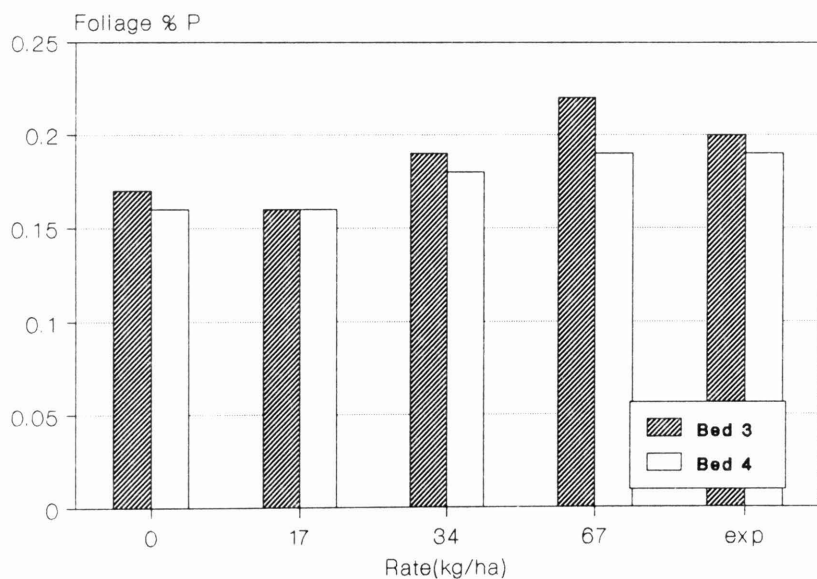
Mean foliar S concentration (weighted %) of loblolly pine seedling receiving varying amounts of top-dress Mg fertilizer. Columns with same letter not significantly different at the 95% level. N=8.



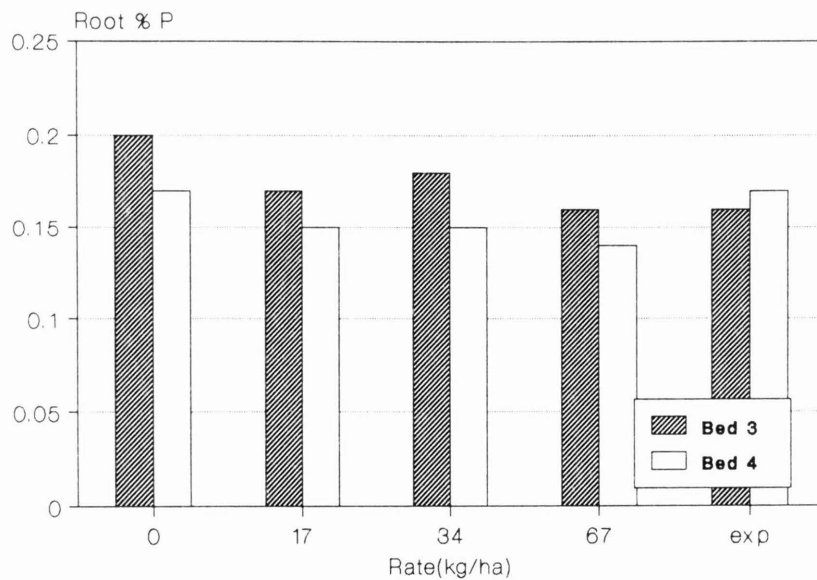
Comparison of mean foliar K concentration (weighted %) of loblolly pine seedlings in bed 4 (compartment 7a) receiving top-dress N at a constant or increasing application rate. Constant = 34 kg/ha elemental N. Increasing rate = 5.4, 10.8, 21.5, 43.0, and 86.1 kg/ha elemental N/application. Seedlings received a total of 5 treatments. Significance at the 95% level indicated by \*. N=4.



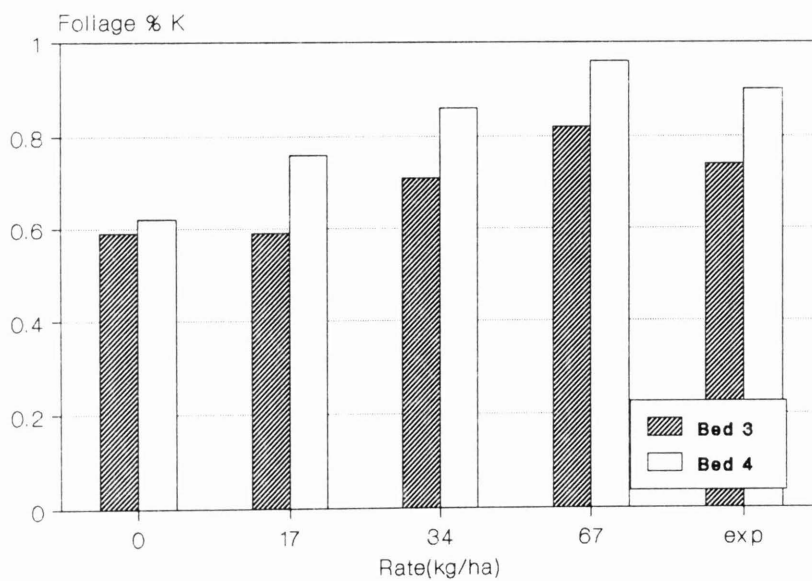
Comparison of mean foliar N concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.



Comparison of mean foliar P concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.

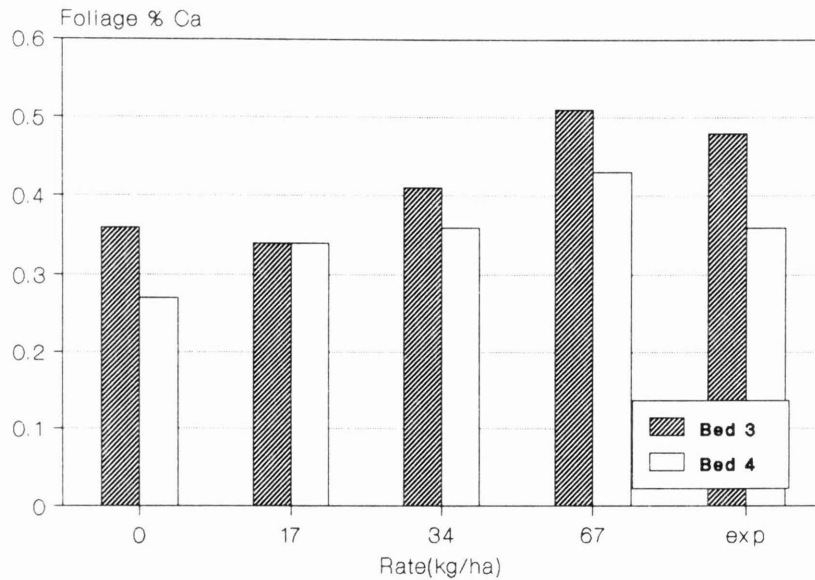


Comparison of mean root P concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.

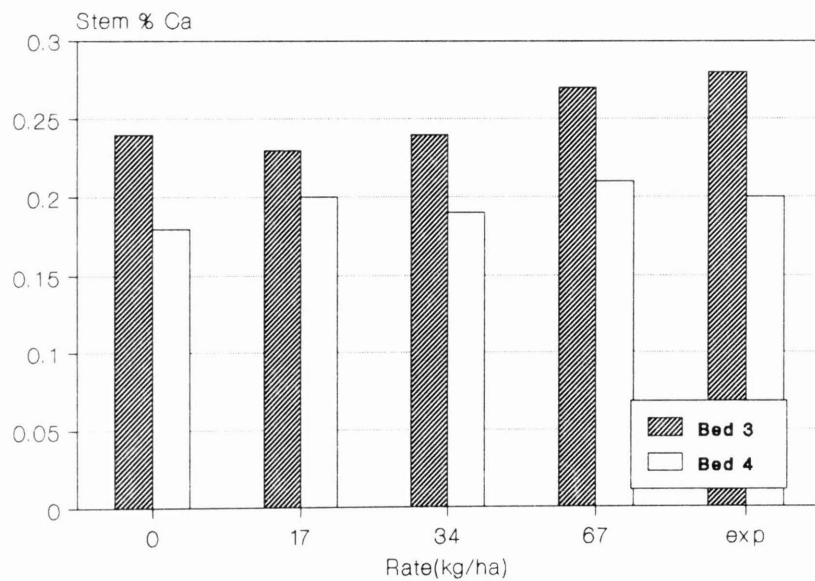


Comparison of mean foliar K concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.

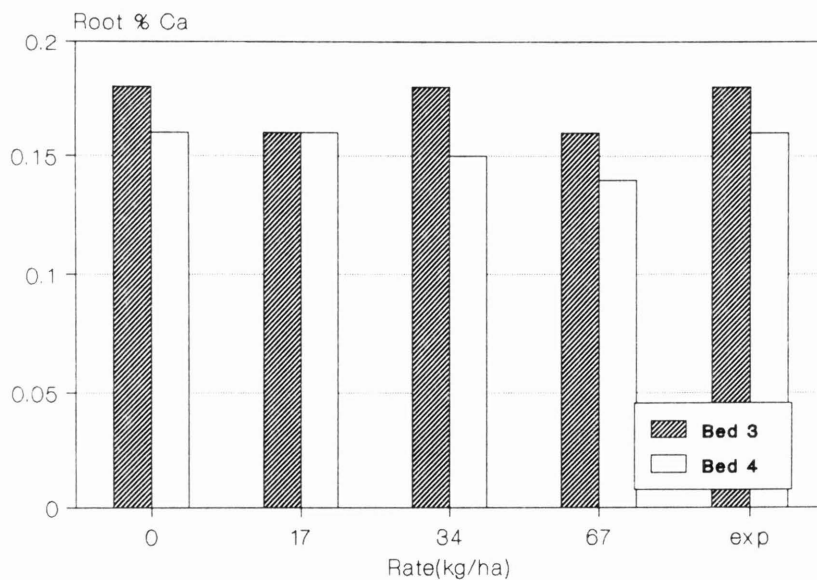




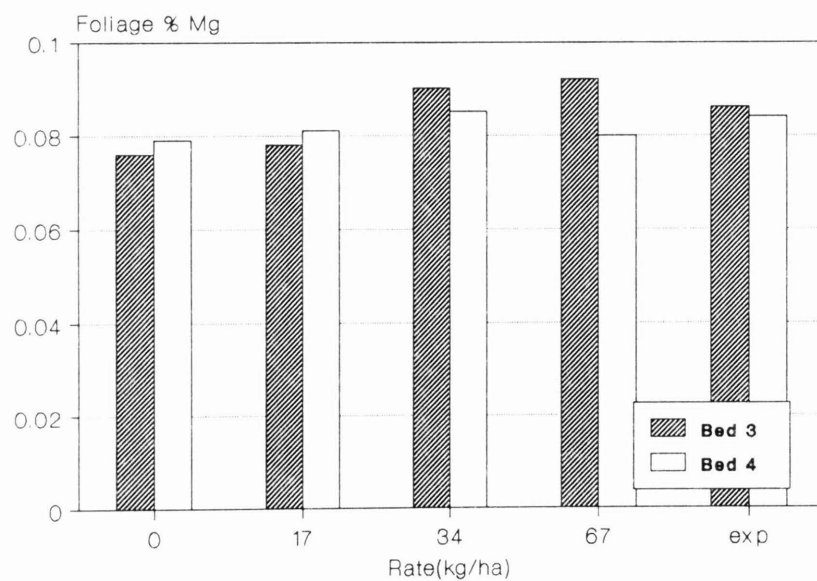
Comparison of mean foliar Ca concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.



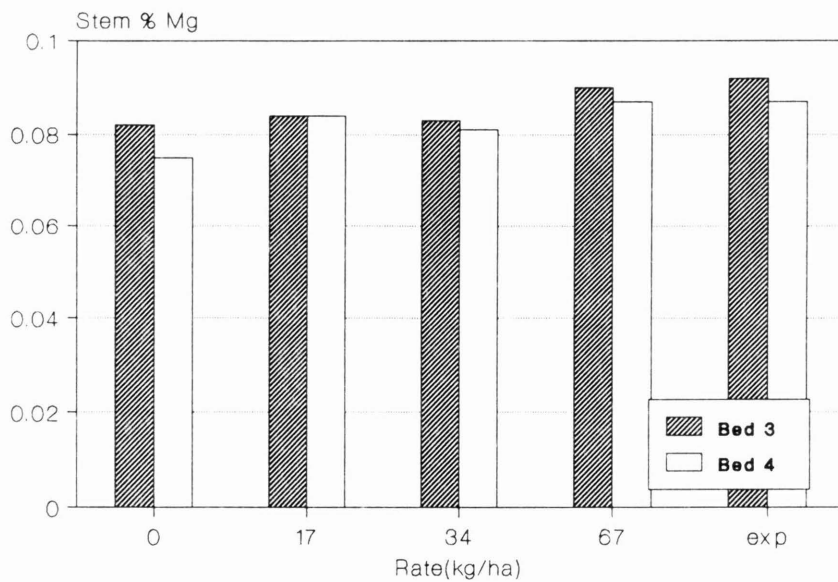
Comparison of mean stem Ca concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.



Comparison of mean root Ca concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.



Comparison of mean foliar Mg concentration(weighted %) between bed 3(compartment 4) and bed 4(compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.



Comparison of mean stem Mg concentration (weighted %) between bed 3 (compartment 4) and bed 4 (compartment 7a) loblolly pine seedlings receiving top-dress N fertilizer. N=12.

**APPENDIX D****JULY SAMPLING SUMMARY**

Average shoot weight, root weight, total weight, and shoot to root ratio of eight week old (July sampling) loblolly pine seedlings receiving pre-plant applications of nitrogen.

Effect	Component (g)			
	Total weight	Shoot Weight	Root Weight	Shoot:root
<u>Block</u>				
A	.51	.456	.054	8.52
B	.463	.415	.048	8.59
C	.471	.422	.049	8.69
D	.447	.40	.047	8.49
<u>Rate</u>				
0 kg N/ha	.486	.435	.051	8.53
11 kg N/ha	.423	.378	.045	8.47
22 kg N/ha	.475	.424	.050	8.42
45 kg N/ha	.521	.467	.054	8.65
90 kg N/ha	.458	.411	.047	8.78

Average shoot weight, root weight, total weight, and shoot to root ratio of eight week old (July sampling) loblolly pine seedlings receiving top-dress applications of magnesium.

Effect	Component (g)			
	Total weight	Shoot Weight	Root Weight	Shoot:root
<u>Block</u>				
A	.538	.481	.057	8.48
B	.470	.417	.053	7.80
C	.475	.423	.052	8.17
D	.459	.410	.049	8.29
<u>Rate</u>				
0 kg Mg/ha	.464	.415	.050	8.35
17 kg Mg/ha	.467	.416	.051	8.11
28 kg Mg/ha	.474	.420	.053	7.87
39 kg Mg/ha	.494	.439	.055	8.03
exp kg Mg/ha	.528	.473	.055	8.55

Average shoot weight, root weight, total weight, and shoot to root ratio of eight week old (July sampling) loblolly pine seedlings receiving top-dress applications of nitrogen(compartment 4).

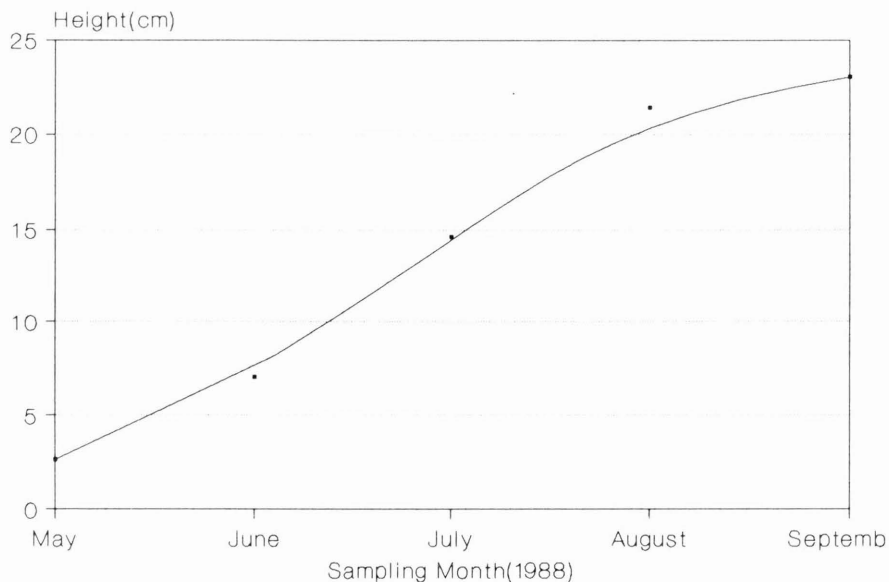
Effect	Component (g)			
	Total weight	Shoot Weight	Root Weight	Shoot:root
<u>Block</u>				
A	.495	.441	.054	8.08
B	.411	.365	.046	7.89
C	.447	.399	.048	8.39
<u>Rate</u>				
0 kg N/ha	.451	.400	.051	7.75
17 kg N/ha	.493	.441	.052	8.55
34 kg N/ha	.450	.400	.049	8.10
67 kg N/ha	.404	.360	.044	8.13
exp kg N/ha	.456	.405	.050	8.08

Average shoot weight, root weight, total weight, and shoot to root ratio of eight week old (July sampling) loblolly pine seedlings receiving top-dress applications of nitrogen(compartment 7a).

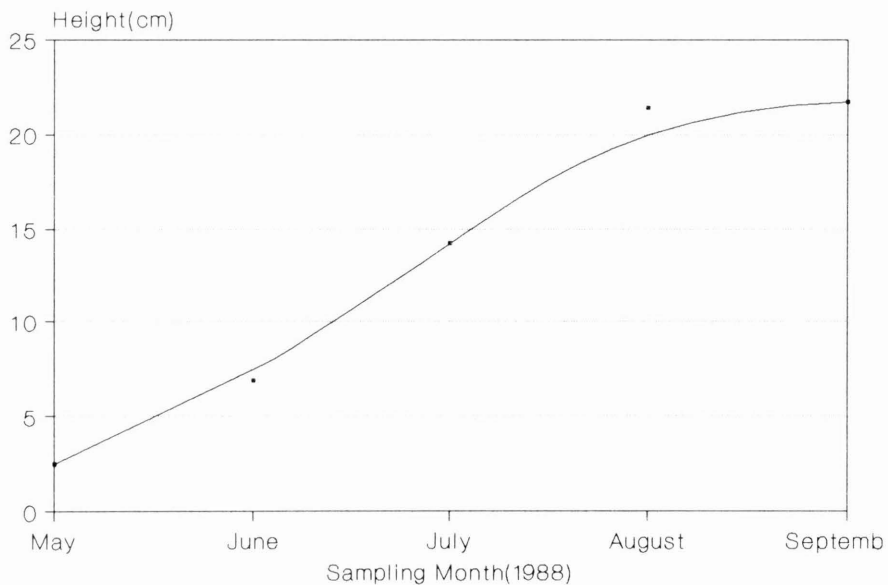
Effect	Component (g)			
	Total weight	Shoot Weight	Root Weight	Shoot:root
<u>Block</u>				
A	.458	.410	.048	8.63
B	.423	.376	.048	7.87
C	.431	.385	.046	8.39
<u>Rate</u>				
0 kg N/ha	.424	.377	.046	8.19
17 kg N/ha	.407	.364	.043	8.40
34 kg N/ha	.447	.401	.045	8.97
67 kg N/ha	.442	.377	.050	7.90
exp kg N/ha	.468	.416	.052	8.04



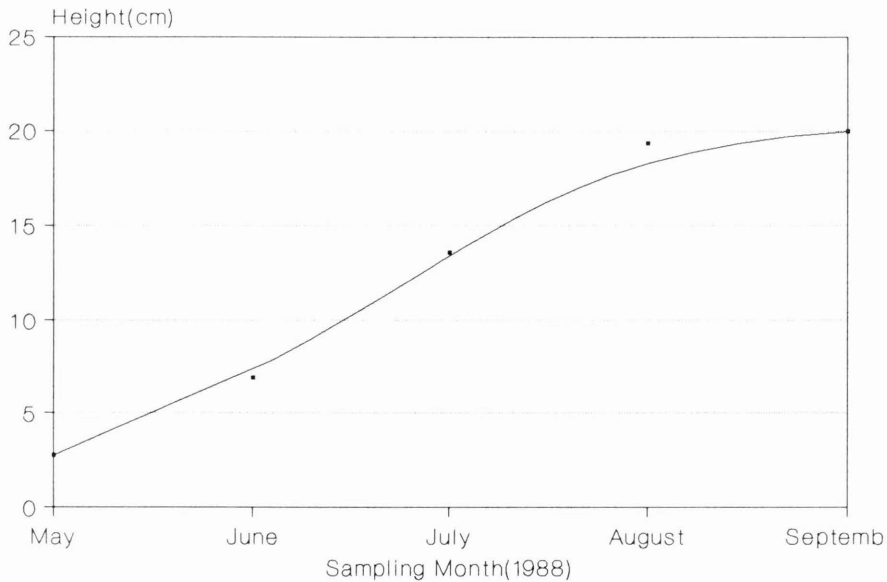
**APPENDIX E****HEIGHT GROWTH CURVES**



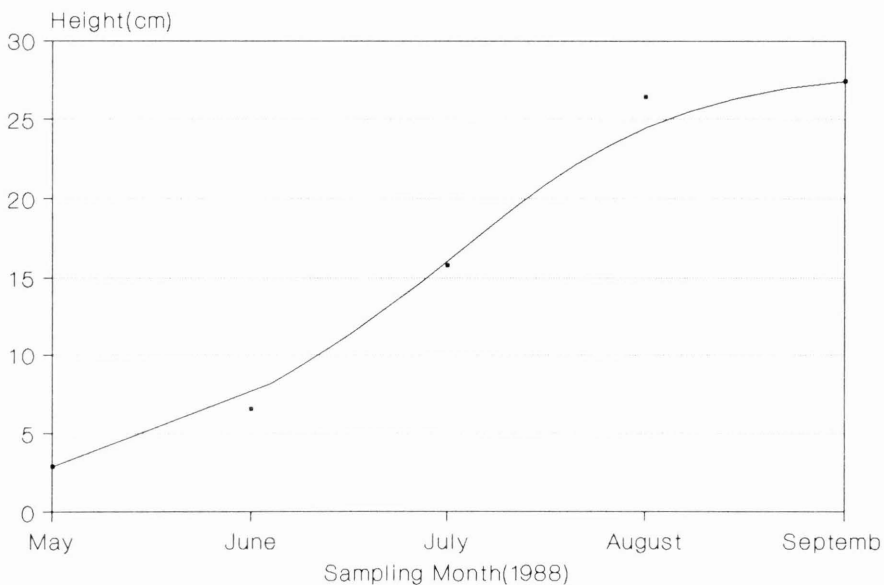
Mean monthly height increments of loblolly pine seedlings receiving pre-plant applications of N (bed 1). N=20. Height measures reported only through September because plots inadvertently disturbed in late September.



Mean monthly height increment of loblolly pine seedlings receiving top-dress applications of Mg (bed 2). N=20. Height measures reported only through September because plots inadvertently disturbed in late September.



Mean monthly height increment of loblolly pine seedlings receiving top-dress applications of N (bed 3, compartment 4). N=15. Height measures reported only through September because plots inadvertently disturbed in late September.



Mean monthly height increment of loblolly pine seedlings receiving top-dress applications of N (bed 4, compartment 7a). N=15. Height measures reported only through September because plots inadvertently disturbed in late September.