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### Fertilizer trials for bareroot nurseries in North America

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### Abstract

In North America, most tree nursery nutrition publications during the past two decades involved either container-grown stock or stock grown in greenhouses. In contrast, most bareroot nursery fertility trials in North America were published during the last century. As a result, some bareroot fertilization recommendations have remained the same since 1980 and some practices continue to be based on myths and assumptions. The bareroot nursery industry in the USA might benefit if the next generation of graduate students will consider testing old and new theories about nursery fertilization. Hopefully, they will discover new facts so that future fertilization regimes will be based on science. This paper provides various fertilizer trials that should be established in bareroot nurseries.

### Keywords

Fertilization; Hypothesis testing; Bareroot nursery; Seedlings; Myths

### Contents

1	Introduction					
2	Macr	onutrients	55			
	2.1	Nitrogen	55			
	2.2	Phosphorus	58			
	2.3	Potassium	59			
	2.4	Calcium	62			
	2.5	Magnesium	63			
	2.6	Sulfur	64			
3	Micro	onutrients	64			
	3.1	Boron	64			
	3.2	Copper	65			
	3.3	Iron	66			
	3.4	Manganese	67			
	3.5	Sodium	67			
	3.6	Zinc	67			
4	Reco	mmendations	68			
5	Acknowledgments					
6	References					

### **1** Introduction

If fertility research in North American nurseries ceases, then myths, mistakes and stagnation will prevail. Fertility research in bareroot nurseries peaked before 1990 and has been on a decline ever since. In North America, the "enthusiasm gap" (Haase and Davis 2017) for bareroot fertility research is rather large. As a result, some 40-year old practices (Sweetland 1978) are still used in bareroot nurseries. For example, 55% of bareroot nurseries in the southern USA still apply K in the fall (Starkey et al. 2015) even though this practice has little benefit (Andivia et al. 2012; Benzian et al. 1974; Birchler et al. 2001; Bryan 1954; Dierauf 1982; Hinesley and Maki 1980; Khan et al. 2013; Rowan 1987; South and Donald 2002; South et al. 1993). I do not know of any physiologists who has data to show that fertilizing pines in the fall with K will "hardenoff" seedlings. Likewise, some nursery managers still apply calcium nitrate to "hardenoff" seedlings. Unfortunately, 20<sup>th</sup> Century research is of little use when it is ignored or forgotten or was never published.

In the absence of 21<sup>st</sup> Century research, myths and opinions will contribute to deciding when, how much and what type of fertilizer to purchase. It is easy to start myths, especially when using the precautionary principle. Just write it down in a publication and show the publication to your friends. Fortunately, myths can be relatively easy to stop by graduate students who follow the scientific method. If the next generation will test hypotheses in bareroot nurseries, they can help to improve our fertilizer regimes. The objective of this paper is to provide a list of hypotheses for those graduate students who choose to test fertilizers in bareroot nurseries in North America.

[Note: Nutrient levels mentioned in this paper were determined using the Mehlich 3 extraction procedure. B = boron. Ca = calcium. Cu = copper. Fe = iron. K = potassium. Mg = magnesium. Mn = manganese. N = nitrogen. Na = sodium. P = phosphorus. S = sulfur. Zn = zinc. ppm = parts per million. Cation exchange capacity = CEC. H0= null hypothesis. LSD = least significant difference.].

### 2 Macronutrients

#### 2.1 Nitrogen

Urea is the second most popular N source for farmers in the United States and is sometimes used as the primary N source for growing tree seedlings (Jacobs et al. 2005; Knight 1978; McNabb and Heser 1997; Sing et al. 2017). Urea may be taken up quicker than other forms of N (Coker et al. 1987; McNabb and Heser 1997). However, when too much granular urea is applied to young seedlings, growth is reduced (Faustino et al. 2015; Sander 1966; South 1975; Villarrubia 1980) and in some cases seedlings may die (Winston 1974). When seedlings are older, they become tolerant of liquid formulations of urea applied at appropriate rates (Sander 1966). Some mangers apply liquid urea (23-0-0) to grow both pine and hardwood seedlings.

H0: When liquid urea (23-0-0) is applied to six-week old pine seedlings (< 11 cm tall), seedling tolerance is not affected by either the amount of irrigation after application (zero, 1 cm) or the rate of application (0, 10, 20, 40, 80, 160 kg ha<sup>-1</sup>).

When applied correctly, certain N fertilizers can increase freeze tolerance of seedlings (Andivia et al. 2012; Islam et al. 2009; Rikala and Repo 1997; Taulavuori et al. 2014; Toca et al. 2017; Triebwasser and Altsuler 1995; Villar-Salvador et al. 2013). For example, "Needles from seedlings with 0.64% N (dry mass basis) before hardening did not harden. Those with 0.87% N showed a lesser degree of hardiness than those with

1.28% N" (Bigras et al. 1996). Likewise, for longleaf pine (*Pinus palustris*), seedlings with 0.9% N were less cold tolerant than seedlings with 1.8% N (Davis et al. 2011). Although ammonium nitrate can increase freeze tolerance of several pines (Toca et al. 2017), it is not known if other forms of N will also be as effective. One study suggests that applying urea to pine seedlings in the fall does not increase freeze tolerance (Montville and Wenny 1990).

## H0: Increasing foliar N concentrations in the fall with either nitric acid or urea (0, 33, 66, 99 kg N ha<sup>-1</sup>) does not affect freeze tolerance of ponderosa pine (Pinus ponderosa) seedlings.

Managers often apply liquid fertilizers to improve uniformity of application and to improve the growth and color of seedlings (Sander 1966). But do the nutrients penetrate through the needle wax? Although there are several advantages of applying liquid fertilizers to seedlings (Gagnon and DeBlois 2017), some people do not claim any benefit from "foliar feeding" of conifers. In most cases "the fertilizer is washed off the foliage and into the soil with irrigation immediately after spraying" (Triebwasser and Altsuler 1995). Although foliar feeding through needles may occur in greenhousegrown seedlings (Eberhardt and Pritchett 1971), I expect a thick layer of epicuticular wax prevents most nutrients from penetrating the needles of outside-grown loblolly pine (Pinus taeda) seedlings (Coker et al. 1987). Typically, micronutrients that are applied in July are washed into the soil (during rainfall and irrigations) and afterwards a portion is taken up through the roots. Dr. Sam Lyle (1972) had to cover pine seedlings with a plastic bag (for 1 or 2 days) to get the foliage to take up nutrients. Since many hardwood species have a thinner epicuticular wax, they might respond to foliar applications without the use of plastic bags (Lyle 1972). The thickness of the epicuticular wax increases with leaf age and this can reduce uptake of foliar applied N (Bondada et al. 2001).

## H0: When loblolly pine seedlings are grown outside in full sun, then applying urea (23-0-0) to older needles (using a paint brush) does not correct the N deficiency of younger needles near the shoot apex.

A few professors have said that nitrate-based fertilizers (e.g. nitric acid, Ca-, Mg-, K-, Na-nitrate) are not good fertilizer sources for bareroot conifers. These claims are likely based on certain hydroponic and greenhouse studies (Bedell et al. 1999; Christersson 1972; Davey 1988; van den Driessche 1978) while disregarding contrary research from greenhouse studies (Addoms 1937; Everett et al. 2010; Pharis et al. 1964), nurseries (Iyer et al. 2002; Lyle and Pearce 1968; Ogbonnaya and Kinako 1993; Radwan et al. 1971), shade-houses (Radwan and DeBell 1980) and plantations (Fisher and Pritchett 1982; Kais et al. 1984; Knight 1963; Radwan et al. 1984; Weetman and Fournier 1984). Some hardwoods grow better when fertilized with a 3:1 nitrate:ammonium ratio (Kim et al. 2002). Just because some hardwoods are sensitive to fertilizers that contain Na (Thorton et al. 1988), this does not mean that nitric acid and calcium nitrate are not good sources of N. In fact, some favor nitrate-based fertilizers when mixing up stock fertilizer solutions (Barker 2010; Donald 1991; Murison 1960; Tinus 1984).

# H0: When fertilizing bareroot northern red oak (Quercus rubra) seedlings with equal rates of N and S, growth is the same when applying either 6% nitric acid (+ S) or ammonium sulfate.

In the past, some N was applied prior to sowing pine seed (Hinesley and Maki 1980; Marx et al. 1984; Leach and Gresham 1983; May 1985; Rowan 1971; van den Driessche 1984). Some managers now accept the view that most of the N applied before sowing may be leached before seedlings can benefit from the application (Knight 1978). In addition, to reduce the chance of damping-off, some recommend delaying applying N fertilizers until after germination is complete (Tinus and McDonald 1979). However, pre-sowing fertilization in Petri dishes can produce some positive effects on germination of pine seed (Henig-Sever et al. 2000) and applying urea before sowing might suppress damping-off of slash pine (*Pinus elliottii*) seedlings (Huang and Kahlman 1991). Is there any benefit from applying urea before sowing pine seed?

### H0: Applying urea to the soil before sowing has no effect on seed efficiency of either pines or hardwoods.

Applying too much urea before (or after) sowing can reduce the production of hardwood seedlings (South 1975). What concentrations of urea are toxic to young hardwood seedlings? Some say the solution should not exceed 0.5% (urea mass/solution mass) for some hardwoods (Knight 1978) but solutions as high as 16% have been applied in a greenhouse (Bondada et al. 2001). Some conifers are tolerant of solutions containing 14% urea (Gagnon et al. 2017). Applying 23-0-0 at 10 kg N ha<sup>-1</sup> (at 310 l ha<sup>-1</sup>) would amount to a 7% urea solution.

H0: When applying 310 l  $ha^{-1}$  of solution to young sycamore (Platanus occidentalis) seedlings, there are no phytotoxic effects from applying 23-0-0 (urea) at 0, 10, 20, 40, 80 kg N  $ha^{-1}$  (with no irrigation after application).



Figure 1. Photos of *Pinus taeda* seedlings at the Great Southern Nursery in Georgia were taken by Gary Cannon on March 3, 1982. Seedlings in the right photo were fertilized in September 25, 1981 with diammonium phosphate (140 kg ha<sup>-1</sup>) while those in the left photo were not treated. The fall-fertilized seedlings broke-bud sooner in the spring indicating nutrition affected seedling performance.

In some trials, a seedling's field performance is increased by N fertilization (Figure 1), even when the fertilizer treatment did not affect seedling morphology. This is because root-growth potential and height growth in the field can be related to foliar

N concentration (Autry 1972; Barber et al. 1991; Grossnickle 2012; Irwin et al. 1988; Jackson et al. 2012; Larsen et al. 1988; Oliet et al. 2013).

# H0: There is no benefit (e.g. foliar N concentration, root-growth potential, early bud-break in the spring) from applying N (0, 100 kg ha<sup>-1</sup>) to conifer seedlings after September 21.

A 21-week delay in fertilizing container-grown pines might reduce seedling mass by 70% (Mexal et al. 1995). How much of a decrease in mass would result from a 2-week delay in N fertilization of bareroot seedlings?

H0: There is no difference in growth of bareroot loblolly pine seedlings when initiating a fertilization program (e.g. 23-0-0 @ 10 kg ha<sup>-1</sup> per application) at three different times after sowing (5-, 7-, 9-weeks). Total application rate for each treatment would be 150 kg ha<sup>-1</sup>.

Some mangers add the herbicide oxyfluorfen to the liquid fertilizer solution because this reduces fuel costs. Does liquid urea increase the phytotoxicity associated with oxyfluorfen?

H0: Phytotoxicity to 6-week old whitebark pine (Pinus albicaulus) seedlings is not increased when applying a liquid urea (23-0-0) solution to oxyfluorfen.

#### 2.2 Phosphorus

What happens when soil P in a bareroot nursery gets too high? Applying 488 kg P ha<sup>-1</sup> before sowing can cause chlorosis and stunting of loblolly pine (Steinbeck et al. 1966). May (1964) said that high P may encourage a deficiency of Zn, Fe, Cu and N. Fertilization with diammonium phosphate can cause a Cu deficiency in loblolly pine plantations (South et al. 2004). In one trial with slash pine, applying high rates of P lowered foliar K concentration to below 0.25% (Brendemuehl 1968).

Soil P levels may be increasing at some bareroot nurseries. In 1977 (Marx et al. 1984), the highest soil level reported in nurseries was 155 ppm (double acid extraction). At that time some managers were applying 217 kg P ha<sup>-1</sup> to seedbeds (Brendemuehl and Mizell 1978). Four decades later four nurseries had fields where the levels exceeded 160 ppm (Mehlich 3) and the level in one field exceeded 300 ppm P. If healthy seedlings are produced at 300 ppm P, at what level would seedlings appear abnormal?

H0: The level of soil P (100, 200, 400, 800 ppm) before sowing has no effect on foliar nutrient values in loblolly pine seedlings in November.

Experts differ on the minimum desired level for soil P in pine nurseries but do these minimums have any practical value? Soils at many nurseries test high for P and, therefore, most soil agronomists do not recommend fertilizing seedbeds with P. However, even a soil with 100 ppm P will not prevent a P deficiency when mycorrhizae are absent (South et al. 2018). In contrast, when mycorrhizae are present, adequate growth of seedlings can be achieved in soil with 11 ppm available soil P (Wall 1994). Therefore, where are the data to show fertilizer should be applied when soil has 20 ppm P? Soils low in mycorrhizal inoculum should be fertilized with P but testing soil chemistry says nothing about the status of soil biology. Therefore, when mycorrhizal spores are present, and the soil contains 25 ppm, will adding P before sowing make any difference?

### H0: The level of soil P (20, 40, 80, 160 ppm) before sowing has no effect on the growth of mycorrhizal bareroot pine seedlings.

When spring fumigation eliminates ectomycorrhizal soil inoculum, nursery managers may apply P to increase the level of P in the foliage (South et al. 1988) but the timing of application is important. Waiting too late to correct the P deficiency can results in a variable seedling crop (South et al. 2018). The next generation of researchers could determine the most effective rates and timing of application.

## H0: The date of application (week 5, 7, 9 after sowing) and rate of liquid P fertilization (0, 20, 40, 80 kg P ha<sup>-1</sup>) have no effect on the growth of non-mycorrhizal loblolly pine seedlings in bareroot seedbeds.

Some experts say that applying ammonium phosphate before sowing may be beneficial, but is this a myth or a fact? This practice might be beneficial when soil does not contain mycorrhizal inoculum, but will it increase seed efficiency in mycorrhizal soil when the soil P level is high? Applying 59 kg P ha<sup>-1</sup> (0-46-0) before sowing had no significant effect on shortleaf pine (*Pinus echinata*) (Brissette and Carlson 1987).

# H0: In sandy soils with adequate mycorrhizal inoculum and P level >100 ppm, applying ammonium phosphate or phosphoric acid before sowing has no effect on seed efficiency of pine or hardwoods.

Some experts say that container-grown pines should have twice the concentration of P in the foliage as bareroot pines. Is this a myth or a fact? In one survey (Starkey and Enebak 2012), the median values for loblolly pine foliage in January were about the same (0.13% and 0.15% P for container and bareroot stock, respectively). I suspect the twice-as-much assumption was based upon sampling stock of different sizes that were grown under different environments and sampled at different ages. When grown outside using the same seed, sowing date, fertilization program and testing lab, is there a difference in foliar P levels when seedlings of the same size are sampled in October?

H0: When pine seedlings are grown to the same size in the same soil using the same fertilizer practice, there is no difference in foliar nutrient levels (October) between container stock and bareroot stock.

### 2.3 Potasium

Some researchers and nursery managers apply about 60% more N than K to produce bareroot pine seedlings (Table 1; van den Driessche 1984). However, when soil K values are low, some managers are told to apply 33% more K than N. Is the practice of applying more K than N based on greenhouse research (Brix and van den Driessche 1974)? Some container-grown pine seedlings will grow slower when fertilized with more K than N (Del Campo et al. 2011; Oliet et al. 2004). If a 2 N/K ratio is suitable for container stock, why fertilize bareroot seedlings with a 0.75 N/K ratio?

H0: The ratio of N (180 kg ha<sup>-1</sup>) to K (0, 60, 120, 180, 240 kg ha<sup>-1</sup>) has no measurable effect on either the morphology or the freeze tolerance of bareroot loblolly pine seedlings.

Species	Ν	К	N/K Ratio	Reference
	Kg ha⁻¹	Kg ha⁻¹		
Pinus taeda	185	24	7.7	Greene and Britt 1998
Pinus elliottii	106	41	2.6	Marx et al. 1989
Pinus elliottii	179	84	2.1	Marx et al. 1986
Pinus taeda	153	79	1.9	Leach and Gresham 1983
Pinus taeda	110	60	1.8	VanderSchaaf and McNabb 2004
Pinus palustris	352	227	1.6	Rodríguez-Trejo et al. 2003
Pinus caribaea	188	120	1.6	Ward and Johnson 1985
Pinus taeda	179	108	1.6	South et al. 2017
Pinus taeda	171	112	1.5	South and Donald 2002
Pinus taeda	157	156	1	South et al. 2015
Pinus clausa	60	60	1	Brendemuehl and Mizell 1978

Table 1. Selected examples of the ratio of nitrogen (N) and potassium (K) used when fertilizing bareroot pine seedlings. Median N/K ratio = 1.6.

Why are K fertilizers applied to soil before shaping seedbeds? Brendemuehl and Mizell (1978) said that K fertilization is usually not needed prior to sowing and sometimes applying 279 kg ha<sup>-1</sup> before sowing had no effect on seedling growth (Switzer and Nelson 1956). Davey (2002) said that it is not wise to apply all the K before sowing; perhaps because (1) foliar levels may fall to 0.5% K by December (Steinbeck et al. 1966; Sung et al. 1997), (2) applying too much KCl before sowing can kill germinating seedlings (Andrews 1941), and (3) KCl easily leaches from sandy soil (Bengston and Voigt 1962). Since rainfall leaches K, why apply KCl a month before roots emerge?

# H0: In soils with less than 30 ppm K, applying KCl (150 kg ha<sup>-1</sup>) before sowing pine has no effect on seed emergence or seedling morphology when seedlings are sampled eight weeks after sowing.

When soil contains at least 25 ppm K, can the first application of K be made in July? Sometimes the first K application is made four or five weeks after sowing (Davis et al. 2011; Jackson et al. 2007; Sampson 1973). K deficiency symptoms can occur when pine is grown in solutions (Hobbs 1944) or in sand with little or no K. When K levels are near zero, symptoms may first appear when the stem above the cotyledons is 2 cm long (Sucoff 1961). When grown in soil with sufficient K, no symptoms appeared when slash pine received no K fertilizers (Figure 2). Can good quality seedlings be produced when all the K fertilizer is applied after June?

H0: In soils with less than 30 ppm K, applying K fertilizers after July 1 (50 kg ha<sup>-1</sup> in July and 50 kg ha<sup>-1</sup> in August) has no effect on seedling morphology, foliar nutrition, or freeze tolerance of seedlings lifted in December.



Figure 2. *Pinus elliottii* seedlings at the Page Nursery in Georgia (photo taken by Jack May August 8, 1961). Seedlings were growing in a loamy sand (33 ppm K) without any potassium fertilizer (Steinbeck et al. 1966). Nitrogen (112 kg ha<sup>-1</sup>) and phosphorus (244 kg ha<sup>-1</sup>) were mechanically incorporated into the soil two weeks before sowing (April 17, 1961) with no fertilization after sowing. In September, seedlings sampled from this treatment were 18 cm tall and foliar nutrient levels were; N-0.9%, P-0.13% and K-0.57%.

In some soils, seedlings can be grown without any K fertilization (Brissette et al. 1989; Khan et al. 2013; Mexal et al. 2002; South 1975; Wall 1994). Greenhouse studies indicated that K fertilization had little or no effect on sand pine (*Pinus clausa*) and slash pine seedlings (Brendemuehl 1968; Steinbeck et al. 1966). K deficiencies can occur when pines are grown in pure sand in greenhouses (McGee 1963) or perhaps in nursery soils with only 4 ppm K (Leach and Gresham 1983). A single liquid application of 28 kg ha<sup>-1</sup> of K may not be enough to prevent a K deficiency when applied as a top-dressing (Leach and Gresham 1983). Applying 10-0-6 (10% N and 5% K) as a liquid spray during the growing season might apply 180 kg N ha<sup>-1</sup> and 90 kg ha<sup>-1</sup> of K. When soil levels before sowing are less than 30 ppm, would this rate be sufficient to prevent a K deficiency?

### H0: In soils with less than 30 ppm, applying K (0, 30, 60, 120 kg ha<sup>-1</sup>) in a liquid formulation (10-0-6) has no effect on growth or foliar content of loblolly pine seedlings. All treatments receive a total of 240 kg N ha<sup>-1</sup>.

KCl is a common fertilizer in pine nurseries since it is cheaper than sulfate of potash (SOP). In contrast, some container nurseries prefer SOP since it is not as toxic to conifers as KCl. Toxicity of KCl to loblolly pine was first reported in North Carolina where 672 kg ha<sup>-1</sup> of KCl (applied before sowing) reduced seedling dry mass (Rosendahl and Korstian 1945). Since then, managers learned how to apply Cl without damaging conifers. Many managers have applied 168 kg ha<sup>-1</sup> of KCl before sowing and a few applied more than 260 kg ha<sup>-1</sup> (Knight 1978; Marx et al. 1984). During germination, is pine injured by S or Cl?

## H0: When nursery soil is < 40 ppm K, the rate of KCl and SOP (0, 200, 400, 600 kg K ha<sup>-1</sup>) applied a week before sowing has no effect on germination and seedling biomass.

Langbeinite (a.k.a K-mag and sul-po-mag) originates from mines near Carlsbad, New Mexico and contains K (18%), Mg (11%) and S (22%). When an increase in soil pH is not desired, langbeinite is often the preferred source of Mg. It is sometimes applied before sowing at rates of 224 kg ha<sup>-1</sup> to 560 kg ha<sup>-1</sup> (Rees et al. 1990; Rodríguez-Trejo and Duryea 2003) and is also applied as a top-dressing (Davey 2002; Triebwasser and Altsuler 1995).

H0: When nursery soil contains less than 20 ppm Mg, the rate of langbeinite (0, 200, 400, 600 kg ha<sup>-1</sup>) before sowing has no effect on germination and seedling biomass.

### 2.4 Calcium

Harvesting 10 Mg ha<sup>-1</sup> of pine seedlings might remove 30 kg of Ca from the topsoil (Table 2). Although some agronomists recommend no Ca when the topsoil in March contains 105 ppm Ca, others apply Ca when soil values drop below 200 ppm (Davey 1991) or 300 ppm (Kormanik et al. 1994)?

H0: When nursery soil contains 100 ppm Ca, incorporating gypsum (0, 200, 400, 800 kg ha<sup>-1</sup>) before sowing has no effect on the growth of pine by November.

Table 2. Estimates of nutrient removals from harvesting a crop of 1-0 loblolly pine seedlings depend on seedling size and seedbed density (e.g. 200 seedlings m<sup>-2</sup>).

Seedling dry mass	Harvested dry mass	Ν	к	Ca	Ρ	Mg	S	Fe	Mn	Zn	В	Cu		
g	Mg ha⁻¹	%	%	%	%	%	%	%	%	%	%	%		
		1.1	0.8	0.3	0.15	0.11	0.1	0.02	0.04	0.005	0.002	0.002		
		Kg ha <sup>-1</sup>							a <sup>-1</sup> of seedbeds					
1	2	22	16	6	3	2	2	0.4	0.8	0.1	0.04	0.04		
2	4	44	32	12	6	4	4	0.8	1.6	0.2	0.08	0.08		
3	6	66	48	18	9	7	6	1.2	2.4	0.3	0.12	0.12		
5	10	110	80	30	15	11	10	2.0	4.0	0.5	0.20	0.20		
7	14	154	112	42	21	15	14	2.8	5.6	0.7	0.28	0.28		
10	20	220	160	60	30	22	20	4.0	8.0	1.0	0.40	0.40		
13	26	286	208	78	39	28	26	5.2	10.4	1.3	0.52	0.52		
Boyei	111	94	22	16	7.9	6.7	5.4	3.4	0.47	0.17	0.07			
Nelson	90	58	21	10	9	-	-	-	-	-	-			
	200	72	42	22	18	-	-	-	-	-	-			
S	-	23	19	11	3	0	-	-	-	-	-			

Due to the higher cost, calcium nitrate is rarely applied to seedbeds in the southern United States. However, calcium nitrate and calcium ammonium nitrate are sometimes used to increase growth of seedlings (Dumroese and Wenny 1997). In Oregon, 2-0 seedlings fertilized with urea survived stress better than seedlings

fertilized with calcium nitrate (Radwan et al. 1971). Calcium chloride can decrease drought tolerance of pine seedlings (Christersson 1976) and in one trial calcium nitrate did not increase freeze tolerance of pine seedlings (Montville et al. 1996). Even so, some people still believe the myth that applying Ca will increase freeze tolerance.

H0: Applying Ca (0, 40, 80, 160 kg ha<sup>-1</sup>) to pine seedlings in September has no effect on freeze tolerance, drought tolerance, stem sinuosity or storability in November.

### 2.5 Magnesium

Why fertilize bareroot pine seedlings with Mg when soil tests indicate more than 25 ppm Mg? Fertilizing with Epsom salts had no positive effect on growth of loblolly pine when soil contained 31 ppm Mg (Wall 1994). Likewise, fertilizing 1-0 seedlings had no effect on growth when they were transplanted into soil with 15 ppm Mg (Edwards et al. 1990). Why would young loblolly pine seedlings need soil with more than 25 ppm Mg?

H0: Applying a pre-sow application of Epsom salt (0, 30, 60, 120 kg ha<sup>-1</sup> to a sandy soil with 25 ppm Mg) has no measurable effect on the morphology, root-growth potential and freeze tolerance of bareroot loblolly pine seedlings.



Figure 3. *Pinus taeda* seedlings showing tip chlorosis in July (photo taken by Chase Weatherly, Arborgen, 2014). Foliar nutrients were: 2.0% N, 0.24% P, 1.04% K, 0.08% Mg, 0.19% S, 0.2% Ca, 77 ppm B, 8 ppm Cu, 949 ppm Mn, and 236 ppm Al. Symptoms of Mg deficiency include chlorosis on tips of needles.

When Mg deficiencies occur on pine seedlings, needle tips turn yellow (Hobbs 1944; Leaf 1968; Landis et al. 1989; May 1984; van Goor 1970; Will 1961b). Some pine species exhibit Mg deficiency symptoms when foliage contains 0.08% Mg (Leaf 1968; Sucoff 1961, 1962). At one nursery, loblolly pine seedlings growing in soil with 16 ppm Mg exhibited deficiency symptoms (Figure 3), but no symptoms occurred at another nursery where seedlings were growing in soil with 11 ppm Mg (South et al. 2017). Growth in that soil was good and when sampled in February, the needles contained 0.1% Mg which is typical for bareroot loblolly pine (Boyer and South 1985). Will

chlorotic tips occur when too much Ca is applied to the soil? If so, will applying too much Ca to seedbeds induce a Mg deficiency?

### H0: Adding gypsum (500, 1,000, 2,000 kg ha<sup>-1</sup>) to seedlings in June (< 20 ppm soil Mg) will not induce a Mg deficiency in pine seedlings sampled in July.

Mg deficiencies can occur when too much K fertilizer is applied (Boynton and Burrell 1944; Knight 1978; Stone 1953). When the K/Mg ratio in young loblolly pine seedlings is less than 18, needle color is often normal but when the ratio exceeds 20, a Mg deficiency can occur. Will applying too much K induce a Mg deficiency in pine seedlings?

### H0: Adding K (50, 100, 200 kg ha<sup>-1</sup>) to seedlings in June (< 20 ppm soil Mg) will not induce a Mg deficiency in pine seedlings sampled in July.

Epsom salts may be used as a top-dressing, but how much is too much? Will applying five applications (over a 15-week period) reduce height growth? Thus far, the following null hypothesis has not been rejected.

H0: Applying a top-dressing application of Epsom salt (170, 280, 390 kg ha<sup>-1</sup>) has no measurable effect on height growth of loblolly pine seedlings.

### 2.6 Sulfur

Research on sulfur requirements for pine and hardwood seedlings is scarce (Browder et al. 2005; Salifu and Jacobs 2006, Leaf 1968) perhaps because some fertilizers contain sulfur. Even so, sulfur deficiencies have occurred in a few bareroot seedbeds (Lyle and Pearce 1968; South and Davey 1983). Some managers reduce the risk of a deficiency by applying sulfur (< 80 kg ha<sup>-1</sup>) before sowing (Marx et al. 1984, van den Driessche 1984). At one nursery, adding more than 62 kg S ha<sup>-1</sup> to the soil before sowing did not affect seedling growth (South et al. 2017). Some managers do not apply any sulfur to the soil when soil tests indicate more than 9 ppm S.

H0: When soil contains 7 ppm S, adding  $H_2SO_4$  (100, 200, 400 kg ha<sup>-1</sup>) before sowing has no effect on the growth of pines by November.

Although applying too much elemental sulfur just before sowing can cause chlorosis to loblolly pine seedlings during a dry year (Carey et al. 2002), no chlorosis occurs when a sufficient amount of rainfall occurs before seeds germinate (South et al. 2017). A greenhouse study could better define this sulfur-rainfall interaction.

H0: When nursery soil is treated with 800 kg S ha<sup>-1</sup> (before sowing), the amount of irrigation in a greenhouse has no effect on the growth or chlorosis of loblolly pine seedlings.

### **3** Micronutrients

#### 3.1 Boron

Applying too much B before sowing can injure pine seedlings (Figure 4). Greenhouse trials can be useful in determining what levels are toxic to seedlings (Khan et al. 2012).

H0: When nursery soils are tested in a greenhouse, the rate of B (0, 8, 16, 32, 64 kg ha<sup>-1</sup>) before sowing has no effect on germination and growth of pine seedlings.

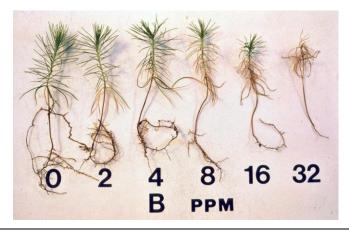


Figure 4. Applying too much B prior to sowing can injure Pinus taeda seedlings (photo taken by David South).

When soil Ca is high (> 600 ppm) and the pH is > 6.0, a B deficiency might occur in sandy soils (Stone et al. 1982). Even applying 0.26 kg ha<sup>-1</sup> of B before sowing loblolly pine seed may not be enough to overcome a deficiency. In November, foliage near deficient terminals ranged from 1.8 ppm to 4.2 ppm B (Stone et al. 1982). At sandy nurseries, some managers apply 2.7 kg ha<sup>-1</sup> of B before sowing pine seed and, therefore, rarely do levels in the foliage fall below 6 ppm. In contrast, some suggest a lower rate when soil levels are less than 0.4 ppm. Can a B deficiency occur when nursery soil pH is below 5.0?

H0: When nursery soil is at 0.1 ppm, the rate of a liquid top-dressing of B (0, 0.5, 1 kg ha<sup>-1</sup>) in July has no effect on the foliar content of washed loblolly pine foliage sampled in September.

#### 3.2 Copper

Although Cu deficiencies can occur in research greenhouses (Majid 1984; van den Burg 1983), on certain sandy nursery soils (Benzian and Warren 1956; Knight 1975) and in plantations (van Goor 1970; South et al. 2004), I have never seen a Cu deficiency at a sandy, low organic matter nursery, even when the soil contains less than 0.2 ppm (Klimek et al. 2008). Rarely does loblolly pine foliage in bareroot nurseries contain less than 6 ppm. When Cu in the topsoil is less than 0.7 ppm, some will recommend applying 3.3 kg ha<sup>-1</sup> of Cu to the soil before sowing pines. When research is rare, recommended rates will vary.

H0: When nursery soil is at 0.1 ppm, the rate of Cu (0, 1, 2, 4 kg ha<sup>-1</sup>) before sowing has no effect on the foliar content of loblolly pine foliage sampled in July.

The cost of Cu may be \$20, \$46 and \$55 kg<sup>-1</sup> when applied as a sulfate, a liquid or as a fertilizer coating, respectively. When a fertilizer distributor adds a coating of Cu to a granular fertilizer, this eliminates the need for an additional application. Tests with different methods of applications will likely have an effect only on an economic spreadsheet. It seems the major reason we apply Cu to sandy nurseries is because we can test the soil for this element.

H0: Application method (before sowing, coated fertilizer and liquid spray in July) has no effect on Cu content of loblolly pine foliage sampled in November.

Fertilization with P can lower Cu levels in conifer and hardwood foliage (Ladiges 2003; Saur et al. 1995; Teng and Timmer 1990, 1994). Will applying too much phosphoric acid induce a Cu deficiency in pine seedlings?

H0: When the soil is at 0.4 ppm Cu or less, the rate of phosphoric acid (0, 200, 400 kg P ha<sup>-1</sup> over time) has no effect on deficiency symptoms of loblolly pine seedlings.

3.3 Iron

Chlorosis can occur when pine seedlings are grown at pH 7.5 (Backmon 1969; Chapman 1941; Richards 1965). In the past when loblolly pine seedlings turned chlorotic in the summer (Figure 5), nursery managers applied chelated Fe in hopes of turning seedlings green (Davey 2002). Several nurseries now grow seedlings at lower soil pH (South 2017) and they no longer experience summer chlorosis. As a result, several managers no longer apply Fe to pine seedlings.

H0: When nursery soil is at less than pH 5.5, the rate of Fe  $(0, 1, 2, 4 \text{ kg ha}^{-1})$  applied to chlorotic seedlings in July has no effect on the color of loblolly pine foliage sampled one and two weeks after treatment.



Figure 5. It has been estimated that 400 seedlings out of every million seedlings produced in southern nurseries are stunted due to a nutrient deficiency (Starkey et al. 2017). Most of these seedlings likely suffer from a phosphorus deficiency (South et al. 2018) but other deficiencies include Ca, Fe, Mg and S. On slightly acidic soils, high doses of N (and root injury from nematodes) induced chlorosis of *Pinus taeda* needles (Photo taken by David South on July 11, 2007).

Does Fe chlorosis affect growth of pine seedlings? It is well known that pH 7.5 soil can reduce the growth of pine seedlings (Chapman 1941; Marx 1990; Richards 1965; South 2017), but how much does Fe chlorosis affect growth of pines? Richards (1965) reported that the growth response to applying chelated Fe depends on soil pH, but an application of Fe did not affect growth at pH 6.7. When pine is growing in pH 6.5, is Fe chlorosis just a temporary, cosmetic effect?

H0: When nursery soil is at pH 6.5, the rate of Fe (0, 1, 2, 4 kg ha<sup>-1</sup>) applied to chlorotic seedlings in a greenhouse has no effect on the growth of pine seedlings.

#### 3.4 Manganese

High levels of Mn in nursery soils may contribute to Ca and Cu deficiencies (South 2017; Turvey et al. 1992). Visual symptoms of Mn toxicity were not observed when pine foliage had more than 1,000 ppm Mn (Adams and Walker 1975; Boyer and South 1985; St.Clair and Lynch 2005). Although little is known about toxic levels of Mn in foliage of pines, applying too much Mn-sulfate can injure some nursery seedlings (Slaton and Iyer 1974). Research conducted with nursery soils found a high level of Mn tolerance in Douglas-fir (*Pseudotsuga menziesii*) (Radwan et al. 1979).

H0: Fertilizing soil with manganese sulfate (0, 200, 400, 800 kg ha<sup>-1</sup>) before sowing has no effect on the germination or foliar content of Douglas-fir foliage sampled in July.

As soil acidity increases, available Mn can increase and when seedlings take up too much Mn, a Ca deficiency can occur. In sandy soils, Mn availability at pH 4.5 may not be great enough to interfere with Ca uptake (South et al. 2017). However, Mn availability is greater in other soils and the risk of a Ca deficiency increases when the soils become more acidic. Research could help determine which soils have a high risk of Ca deficiency when soil pH is less than 5.

## H0: Fertilizing different soil textures with manganese sulfate (0, 200, 400, 800 kg ha<sup>-1</sup>) has no effect on chlorosis and foliar content of loblolly pine seedlings growing in pH 4.5 soil.

Harvesting 10 Mg ha<sup>-1</sup> of seedlings might remove 4 kg Mn ha<sup>-1</sup> from the nursery (Table 2). Therefore, a topsoil with 4 ppm Mn may contain the equivalent of two crops of seedlings. Even so, manganese sulfate is rarely applied to nursery soils that have 5 ppm Mn. Is a threshold of 5 ppm Mn too low for bareroot pine seedlings?

H0: When soil contains 5 ppm Mn, incorporating manganese sulfate (0, 10, 20, 40 kg ha-1) before sowing has no effect on the growth of pine seedlings by November.

#### 3.5 Sodium

Growth of young hardwood seedlings was decreased after fertilizing with Na2SO4 in a growth chamber (Thorton et al. 1988). This might also explain why fertilizers containing Na have reduced the growth of hardwoods in bareroot seedbeds (Villarrubia 1980).

H0: When fertilizing green ash (Fraxinus pennsylvanica) with 120 kg/ha of Na and equal amounts of N, the type of anion (Na<sub>2</sub>SO<sub>4</sub>, NaCl, and NaNO<sub>3</sub>) has no effect on height growth and seedling mass.

### 3.6 Zinc

I have never seen a Zn deficiency in bareroot loblolly pine seedlings. Therefore, when soils contain 1.4 ppm Zn, why do some experts recommend 2.2 kg ha<sup>-1</sup> when others see no need to add Zn? Is a threshold value of 3 ppm Zn too high for loblolly pine?

H0: When soil contains 1 ppm Zn, incorporating zinc sulfate (0, 10, 20, 40 kg ha<sup>-1</sup>) before sowing has no effect on the growth of loblolly pine by November.

### 4 Recommendations

Graduate students who choose to test the above hypotheses should be aware of the most common statistical errors (Fowler 1990; Haase 2014) and then consult with an experienced statistician before designing a fertilizer trial. Ask for an experimental design with enough statistical power to detect an 8% difference in seedbed density and a 7% first-year height increase. The statistical power of some fertilizer trials is sometimes low (South and VanderSchaaf 2017) and, therefore, variability might not be able to reject a null hypothesis even when a treatment caused a 100% increase in a seedling trait (e.g. Figure 6). Likewise, the least significant difference value (LSD) should be reported for each response variable as this allows the reader some idea of the statistical power (South and VanderSchaaf 2017). This will be especially informative when the null hypothesis is not rejected. If you do not already know, ask how to use contrast tests to examine linear and quadratic effects because these tests should be used for almost all fertilizer rate trials. In fertilizer toxicity trials where all we need to know is if the treatment reduces growth, use a one-sided t-test where appropriate (South and VanderSchaaf 2017).

Please describe in detail all soil parameters. Far too often, the data presented omit important detail that affect the outcome of the results. Also, provide the cost of fertilizers being tested. In some cases, the cost of a chelated fertilizer can cost 90 times more than a commonly used formulation. It can be a waste of time to conduct research on fertilizers that are cost prohibitive. Also, realize that fertilizers typically represent a small percentage of the total growing costs. Even when fertilizers cost \$1,800 ha<sup>-1</sup>, the cost per seedling (@ 190 m<sup>-1</sup>) is less than 0.1 cent which equates to a small percentage (e.g. 2%) of the total cost of production. Regeneration economics should also be considered. In some cases, spending extra money on nursery fertilization can lower the overall cost of reforestation. For example, spending 0.01 cent per seedling for fall fertilizers in Spain lowered the cost per living seedling (Puértolas et al. 2012).

When writing a study proposal, state the null hypotheses you wish to test. This might avoid embarrassment when the assumed outcome (i.e. alternative hypothesis) does not occur. Finally, when writing a thesis or dissertation, provide all the data (i.e. individual seedling measurements) in appendices. This will allow others the opportunity to collaborate by asking different questions and perhaps providing additional insights (South and Duke 2010).

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