GENETIC ASPECTS OF NURSERY MANAGEMENT FOR SEEDLING UNIFORMITY

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Abstract

Several experiments were designed to evaluate various genetic and environmental aspects of sweetgum (Liquidambar styraciflua L.) seedling production. Open-pollinated seed lots of 12 clones were sorted into three density classes. Family and seed density class affected early seedling height but became unimportant by mid-summer. Removal of a particular density class from a bulked seed lot altered the genetic composition of the lot. Year of collection also affected growth and uniformity of the seedling crops. Experimentation further revealed that (1) sweetgum seedlings require elemental soil phosphorus levels greater than 50 kg/ha in sandy soils, (2) amendments of soluble phosphates or mycorrhizal inocula to fumigated nursery soils have greatest impact early in the growing season, (3) mycorrhizal inoculation is not a substitute for good soil fertility, (4) family-by mycorrhiza interaction is nonsignificant, and (5) elemental phosphorus levels as high as 130 kg/ha do not inhibit mycorrhizal development.

Additional keywords: seed sizing, mycorrhizae, Liquidambar styraciflua L., seedling growth.

INTRODUCTION

Successful establishment of sweetgum plantations requires uniformly tall, large calipered seedlings. Unfortunately, nursery managers have had difficulty in consistently producing seedling crops of acceptable quality. Traditional factors at the disposal of nursery managers to improve seedling crops are seed quality and soil fertility. Important components of seed quality include seed size, seed year, and genetic composition. Soil fertility management traditionally includes maintenance of macro-and micronutrients and their combination at specified levels for good seedling growth. Use of mycorrhizae has recently been determined to have the same type of beneficial effect on seedling growth as fertilizer supplements. This paper summarizes several studies conducted by the Hardwood Research Cooperative at North Carolina State University to evaluate nursery practices which would improve seedling uniformity and quality in sweetgum.

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EFFECT OF SEED SIZE

In a study of seed size by year of collection bulked seed lots were collected from Weyerhaeuser Company's select sweetgum seed orchard in Washington, N. C. in 1975 and 1976. Seeds from each collection year were separated into four density classes using a gravity feed table in a manner described by Bonner (1979) for sycamore. Seed fractions were *sawn* in a greenhouse using a replicated complete block design in July, 1978. Each treatment plot of nine square dm contained ten seedlings after the last thinning was completed in October, 1978. The December 1978 measurements reflected a trend (not statistically significant) of larger seedlings corresponding to heavier seeds (Table 1). Year of collection significantly affected height growth, but not root collar diameter (CD).

Table 1. Effect of seed density anu year of collection on seedling height and root-collar diameter

HEIGHT

Collection Year	Seed Density 1 (lightest)	2	3	4 (heaviest)	$\bar{X}$
	cm				
1975	25.4	26.6	26.8	29.4	27.0
1976	27.5	29.6	30.5	30.0	29.4
$\bar{X}$	26.5	28.1	28.6	29.7	28.2

ROOT COLLAR DIAMETER

Collection Year	Seed Density 1 (lightest)	2	3	4 (heaviest)	$\bar{X}$
	mm				
1975	2.9	2.9	3.0	3.3	3.0
1976	3.0	3.0	3.3	3.0	3.1
$\bar{X}$	3.0	3.0	3.1	3.1	3.1

In a study of seed size by half-sib family (HSF), seed lots were collected from twelve clones in Weyerhaeuser's orchard in the fall, 1978. The seeds from each lot were extracted, floated to remove debris and unsound seeds, and stored in a cold roan until spring, 1979. Each lot was then separated by placing it in water and adding sodium tartrate (an inert salt) until the density caused one-third of the seeds to float. After these seeds were removed and rinsed, more salt was added to the solution to separate the remaining seeds in half. A portion of each seed lot was left unsorted. To prevent differences in water absorption rates from influencing density, seed lots were soaked in water for three days prior to separation.

All lots were sown in April, 1979 in the Federal Paperboard Company Nursery at Lumberton, N. C. Plots were 30 an by 140 an, the bed width, and were separated by pieces of heavy plastic-covered cardboard buried 13 an deep to restrict lateral root competition among plots. The nursery bed was fumigated two weeks before planting and was fertilized before and after planting to maintain soil nutrients at specified levels. Final thinning brought the spacing to 1.1 seedling per square dm.

Analysis of seedling height measured in June, at 12 weeks of age, revealed seedling size was significantly affected by both HSF and seed density within HSF (Table 2). Heavier seed density classes generally produced larger seedlings. Family and seed size within family accounted for 40% of the variation in plot means.

Table 2. Mean height of sweetgum seedlings by clone and seed density class in June at 12 weeks of age

Clone	Seed Density Light	Medium	Heavy	$\overline{X}$	Unsorted
	cm				
1	8.6	7.9	10.9	9.13	11.3
2	10.1	10.4	10.9	10.47	10.6
3	8.6	9.6	10.9	9.67	9.9
4	8.5	10.2	10.1	9.57	12.8
7	8.3	7.8	9.0	8.38	9.2
9	9.7	14.1	12.3	12.04	13.4
10	10.5	7.9	9.5	9.29	11.0
13	10.7	9.8	9.0	9.82	10.0
23	8.9	9.1	10.0	9.33	11.6
29	8.8	8.7	10.9	9.46	7.5
39	10.7	11.4	10.2	10.76	9.5
42	8.7	10.4	10.5	9.89	8.0
$\overline{X}$	9.33	9.78	10.35	9.82	10.40

By August neither HSF nor seed density within HSF had a significant effect on seedling height growth. Only two families still showed a consistent trend of larger seedlings being produced from heavier seeds. By October, two families showed larger seedlings being produced from heavier seeds, but these were not the same two families. The mean height of seedlings grown from non-sorted seed was not significantly different than the mean of those from the sized seed lots, indicating no competitive advantage by growing seeds from sized seed lots. Root collar diameter was also not significantly affected by HSF or density within HSF, further suggesting that the nursery environment and seed to seed variation were more influential in affecting seedling size than were the effects of HSF or density class.

These findings were also supported by a study to determine whether seedlings grew better when sown by family or by density class (Johnson, 1980). No height growth advantage was obtained in segregating seeds by family or by density class.

The density distribution of seeds within families was subsequently examined. Fifty seeds were selected from each of the non-sorted seed lots and floated using known concentrations of sodium chloride (Table 3). Both the mean density and degree of variability required to float seeds differed among families. Such variation alters the genetic composition of bulked seed lots when they are density sorted. The heavier families remain in larger proportions after removal of the lighter seeds. This same effect has been found in white spruce (Hellum, 1976) and Douglas-fir (Silen and Osterhaus, 1979).

Table 3. Sweetgum seed density distribution by clone

Density	Clone 1	2	3	4	7	9	10	13	23	29	39	NaCl per 100 ml water
g/cc	Percent of seeds that float											gm
1.084	0	0	0	2	0	2	0	0	4	2	2	12
1.098	0	0	0	2	2	2	2	4	4	2	2	14
1.112	0	6	2	2	2	4	6	4	4	2	2	16
1.126	0	6	4	4	6	4	8	4	6	4	2	18
1.140	2	6	4	6	6	10	10	4	12	16	4	20
1.154	6	6	4	6	14	28	16	4	24	38	20	22
1.168	32	20	8	16	40	32	28	8	50	80	64	24
1.182	32	60	22	44	66	61	54	26	72	96	84	26
1.196	62	88	38	88	94	80	74	50	88	100	96	28
1.210	94	94	62	90	100	82	90	66	90	100	100	30
1.224	100	100	90	100	100	86	100	76	100	100	100	32

EFFECT OF PHOSPHORUS AND MYCORRHIZAE ON SEEDLING SIZE

Since sorting sweetgum seed by density class and family did not improve growth or uniformity, emphasis was shifted to nursery fertility factors. Conventional wisdom implies that the presence of mycorrhizal fungi, as well as minimum nutrient levels, must be maintained for the genetic potentials of seeds to be expressed.

Since sweetgum is reported to be an obligate endomycorrhizal species (i.e. it does not grow past the primary leaf stage without endomycorrhizae), the use of methyl bromide fumigation could contribute to inconsistent seedling crops (Kormanik et al. 1977). In an experiment evaluating one endomycorrhizal fungus, Glomus fasciculatus, and three open pollinated sweetgum families collected in 1974 from the Weyerhaeuser orchard, Paschke et al. (1979), found that: (1) sweetgum seedling height was negatively affected by fumigation, (2) amendments of mycorrhizal inocula or water-soluble phosphates (surface broadcast) equally increased seedling heights, (3) family effect was significant but the family by mycorrhizal interaction was always nonsignificant, and (4) background soil phosphorus determined the significance of the mycorrhizal effect, i.e., when elemental soil phosphorus (P) was above 50 kg/ha, the mycorrhizal effect was barely significant, whereas the mycorrhizal effect was highly significant when soil phosphorus was below 50 kg/ha.

To better evaluate the phosphorus fertility, mycorrhizal, and family relationships, three mycorrhizal fungi (Glomus etinucatus, Glomus fasciculatus, and Gigaspora margarita) plus a control, and six randomly selected families from the 1978 seed collection were chosen for further investigation. A standardized inoculum density of 110 spores per square decimeter was applied. Two replications were planted in soil with an elemental soil P content, as determined by the double acid extractant, of 30 kg/ha (low P), and the remaining two replications were planted into a soil having 67 kg/ha of P (high P). The soils were fumigated with over 600 kg/ha of methyl bromide (MC2) before planting, and seedling density by the fourth month was reduced to 1.1 per sq. dm.

The combined analysis utilized data from both fertility levels (Table 4). Mycorrhizal and family effects were significant for seedling height at all measurement tines and RCD measured at month seven. Foliage dry weight was not affected by either family or mycorrhizal inoculation, although control seedling means were significantly smaller than inoculated seedling means for height and CD. No statistical differences existed among the mycorrhizal fungi. Family mean rankings changed over time but only family 7 made any important rank change after the fifth month. The family by mycorrhizae interaction was nonsignificant for all measured traits regardless of soil P content.

The high P replications exceeded the soil P level of 50 kg/ha recommended for hardwood seedlings (Davey, 1980). In this case the mycorrhizal effect on seedling height was significant for months three and four but became nonsignificant after month five (Table 4). By the seventh month the control seedlings, though still the shortest, were not significantly different from the inoculated seedlings. Seedling RCD was affected by the fungal treatments. Seedlings inoculated with Glomus etinucatus and Gigaspora margarita were significantly larger than seedlings treated with Glomus fasciculatis, and all inoculated seedlings were significantly larger in RCD than the control seedlings. Under high P conditions no family differences were observed.

For seedlings grown with low soil P, the mycorrhizal effect for seedling height was significant at the ten percent level for months three, four and five (Table 4). For these months control seedlings were significantly shorter than the inoculated seedlings, though no differences developed

Table 4. Rankings of height over time, root collar diameter, and foliage dry weight means for mycorrhizae and family effects by Duncan's Multiple Range Test, and Analysis of Variance significances

			Combined		Low P		High P	
Main Effect	Trait	Measurement Time	ANOVA Significance	Mean Rankings	ANOVA Significance	Mean Rankings	ANOVA Significance	Mean Rankings
Mycorrhizae								
	Height	3 months	**	1,2,3,4	+	1,2,3,4	*	3,2,1,4
		4 months	**	1,2,3,4	+	1,2,3,4	*	3,2,1,4
		5 months	*	2,1,3,4	+	2,1,3,4	NS	2,3,1,4
		7 months	*	2,1,3,4	+	2,1,3,4	NS	3,2,1,4
	Root Collar Diameter	7 Months	*	2,3,1,4	**	2,1,3,4	**	2,3,1,4
	Foliage Dry Wt.	7 months	NS	2,3,1,4	**	2,1,3,4	NS	3,4,1,2
Family	Height	3 months	**	9,13,28,20,24,7	**	9,13,28,24,7,20	**	13,9,28,20,24,7
		4 months	*	9,13,28,24,7,20	+	9,28,24,7,13,20	+	13,9,28,20,24,7
		5 months	*	28,13,7,9,24,20	**	28,7,9,24,13,20	NS	13,24,9,28,7,20
		7 months	**	28,13,9,24,7,20	**	28,7,9,24,13,20	+	13,28,24,9,7,20
	Root Collar Diameter	7 months	NS	13,28,9,24,7,20	NS	28,9,24,7,13,20	NS	13,9,28,24,7,20
	Foliage Dry Wt.	7 months	NS	28,13,24,9,7,20	**	28,24,9,7,13,20	NS	13,28,24,9,7,20

a) Mycorrhizal Treatments:

1= *Glomus etinucatus*
2= *Glomus fasciculatus*
3= *Gigaspora margarita*
4= control

Any two mean rankings with the same line in common are not significantly different by Duncan's multiple range test procedure which used the ANOVA significance as the probability level.

and + means significant at 10 percent probability level.
* means significant at 5 percent probability level.
** means significant at 1 percent probability level.

among the fungal treatments. At seven months, the mycorrhizal effect upon height had increased in significance to the five percent level with Glomus species superior to Gigaspora. Foliage dry weights were highly significant with the control plots being inferior to the amended plots. Ranking of the fungi was the same for height and foliage dry weight. Mycorrhizal effects on RCD were significant at the ten percent level. Control seedlings were again significantly smaller than the inoculated seedlings. No family differences were found for RCD in low P, but the family effect for height was highly significant at months three, five and seven. Foliage dry weight also had a highly significant family effect. Family mean rankings for height stabilized after month five with ranking of foliage dry weight mirroring these results.

Soil P content had a major impact upon the significance of the mycorrhizal treatments. Yet, the practical outcome of this experiment was that mycorrhizal inoculation was not a substitute for good soil fertility because the shortest seedlings in the high P replications were larger than the tallest seedlings in the low P replications. The diminished significance caused by the high soil P level can be explained by the effect mycorrhizal fungi have upon root absorption area of developing seedlings. Bieleski (1973) calculated that four connections per nun of root, each extending 20 mm from the root surface, could increase phosphorus uptake from ten times if uptake were proportional to root surface area to about 60 times if diffusion were limiting.

Considering the small root system of developing seedlings, the additional root absorption area caused by mycorrhizal infections can explain observed significant effects. Eventually, even the non-inoculated seedlings would become mycorrhizal as the root systems extended beyond the. fumigation zone, although this development would be delayed by law P levels. Infection and subsequent improved growth of control seedlings reduced the significance of the inoculation treatments in the high P soil. Seedlings in law P soil could not grow until a mycorrhizal relationship was established supporting findings of Kormanik et al. (1977).

Family differences were also influenced by soil P fertility with differences evident under high P exaggerated by law P conditions. Family 13 was most sensitive to the P fertility differences moving from next last rank when P was limiting to second best under high P.

Since most nursery soils contain soil P levels above 50 kg/ha, the relatively uniform family results reported for high P are more likely consistent with nursery expectations as well as with the seed size trial mentioned earlier. Other sweetgum studies have found the family component to be highly significant (Weir and Sprague, 1975, Kormanik et al. 1979), Since the families for the reported studies were collected from a seed orchard, the uniformity could have been caused by panmixia. Seed year has played a role in family differences because the 1974 seed collection (Paschke et al. 1979) had highly significant family differences. Nevertheless, the uniform family growth would be desired by nurserymen and plantation establishers.

CONCLUSIONS

Sowing seeds by density class or family should not increase sweetgum seedling growth or uniformity since family and seed density did not affect seedling size after three months. However, removal of a particular density class from bulked seed lots will affect the genetic composition. Year of collection affects both growth and uniformity of a seedling crop. Seed orchard seed appeared to produce seedlings superior to natural stand collections.

Mycorrhizal inoculation in fumigated nursery soils has its greatest effect early in the growing season because of the small root absorption area of the seedling. Mycorrhizal inoculation was not found to be a substitute for soil fertility. Soil P levels significantly affected seedling growth and should be maintained above 50 kg/ha. Amendments of mycorrhizal inoculation or soluble phosphates (surface broadcast, 5-10 kg/ha) were found to equally increase sweeten seedling height. Family differences for the 1978 seed year were masked by soil P fertility above 50 kg/ha but the family by mycorrhizae interaction was always unimportant. RCD was increased by inoculation and differences among the fungi existed at the high P level. This may prove important after further evaluation. Soil P levels as high as 130 kg/ha did not inhibit mycorrhizal development but could affect uptake of other important nutrients.

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