Genetics / Tree Improvement

SURVIVAL AND GROWTH OF A *QUERCUS RUBRA* REGENERATION COHORT DURING FIVE YEARS FOLLOWING MASTING

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Post-harvest regeneration of oak in the mid-Atlantic region is often limited by an insufficiency of numbers and size of advance reproduction seedlings. In Pennsylvania, this problem is sometimes (but not always) associated with a dense ground cover of ferns that appears to inhibit tree seedling growth, both before and after harvest. An unnaturally dense population of white-tailed deer is at least partly to blame for weak levels of oak advance reproduction, and the paucity of low woody vegetation occasioned by deer browsing may contribute to the abundance of fern growth in some stands. This suggests the hypothesis that, in many mature hardwood stands, deer browsing has tipped the competitive balance between ferns and understory woody plants (including tree regeneration), with the result that the scarcity and small size of woody plant regeneration is both a cause and an effect of profuse fern growth.

If this hypothesis is correct, then what happens to seedlings that germinate under a fern cover and are protected from browsing? Can they survive and grow sufficiently to eventually dominate the fern, or do both deer browsing and fern competition need to be controlled to permit tree seedlings to establish? In this study we followed two “reproduction cohorts” for five years, from the time of masting in early autumn of 1990 until early autumn 1995. The objectives were 1) to document losses at various stages of the reproduction process and 2) to determine the ability of unbrowsed seedlings to grow out of dense fern ground cover.

The study was performed in two stands located 2.6 miles from one another on Moshannon State Forest in central Pennsylvania. Both stands were about 75 to 80 years of age at the beginning of the study. In the “Smith Road” stand northern red oak (NRO) represented 68 percent of the 96 ft^2/acre basal area, and the forest floor was 43 percent covered with herbaceous vegetation (bracken fern, *Pteridium aquilinum,* and hayscented fern, *Dennstaedtia punctilobula*) and 19 percent covered with woody vegetation (predominantly *Vaccinium* spp.). In the “Firebreak” stand NRO represented 48 percent of the 139 ft^2/acre basal area, and the forest floor was 65 percent covered with herbaceous vegetation (predominantly hayscented fern) with negligible woody ground cover. Site indices of the two stands are 63 and 73 ft, respectively. Advance reproduction of NRO for these stands in any given year has been one to a few thousand, with no seedlings observed in excess of 30 cm in height. More information on these stands may be found in Steiner and others (1993).

The study reported here was prompted by heavy mast crops in both stands in 1990. Acorn production, autumn removals by vertebrates, and viability loss caused by insect or disease injury were determined by sampling as described by Steiner (1995). Winter removals by vertebrates were determined indirectly by the difference between potential germinants in November and actual germinants in May of the following year. Immediate post-germination losses (to both insects and vertebrate predation) were determined by successive visits in May and June. In June of 1991, 400 (200 per stand) first-year NRO seedlings were located, mapped, labeled, and measured for height. Study seedlings were fairly evenly scattered over an area of about 4 acres at each site but all were located within fern ground cover. To permit an examination of growth and survival in the presence and absence of deer browsing, half (100) of the study seedlings in each stand were protected by means of a 3.25 x 36-inch tubular plastic net (sold as a “Rigid Seedling Protector Tube”) supported by two bamboo stakes. This material casts a measured 11 percent shade. Seedling heights and survival were determined 1.0, 1.5, 2.0, and 4.5 years after germination (up to 5.0 years following masting). Stand canopies remained essentially intact over the course of the study.

Survival of the reproduction cohort from fall 1990 until fall 1995 is shown in Table 1, and height growth of protected and unprotected seedlings is shown in Table 2. In general, the two stands were very similar in their overall pattern of acorn loss and seedling mortality, so only mean values will be presented. Of the average regeneration cohort of 141,075 filled acorns per acre, 90 percent were destroyed or consumed before germination in spring 1991 (as quantified in May), and 99 percent were lost by the end of the fifth growing season. Pre-germination losses to insects or disease (not tabulated separately in Table 1) accounted for an average of only 2.2 percent of the acorn crop. Some unquantified losses to insects occurred immediately after germination, but total cohort mortality attributable to insects or disease was no more than 11.2 percent and probably less.

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Table 1—Survival over 5 years of NRO reproduction cohorts in two Pennsylvania stands, beginning at masting in 1990

		Smith Road		Firebreak		Mean	
	Year	*Number*	*Percent*	*Number*	*Percent*	*Number*	*Percent*
Fall 1990[a]	0	198,510	100	83,640	100	141,075	100
May 1991[b]	0.5	23,390	11.78	5,130	6.13	14,260	10.11
June 1991[c]		2,548	1.28	667	0.80	1,608	1.14
Spring 1992	1.5	1,631	0.82	360	0.43	995	0.71
Fall 1992	2.0	1,554	0.78	340	0.41	947	0.67
Spring 1993	2.5	1,249	0.63	240	0.004	360	0.26

[a] Production of filled acorns (with naturally abscised caps), representing initial size of the reproduction cohort.
[b] Percentage germination after losses to insects, disease, and consumption by vertebrates.
[c] Seedling survival after immediate post-germination losses to insects and disease, and consumption by vertebrates. Subsequent survival figures are based on marked, unprotected seedlings selected in June 1991.

Table 2—Heights (cm) of advance reproduction of NRO in two Pennsylvania stands in June of first growing season (when protectors were installed) and after 1, 2, and 5 growing seasons, protected and unprotected against browsing by white-tailed deer

	Smith Road		Firebreak	
Seedling age	Unprotected	Protected	Unprotected	Protected
Years				
0	10.40 b	9.04 a	12.67 c	13.33 c
1	10.48 a	10.69 a	11.38 a	14.93 b
2	11.61 a	12.36 a	12.44 a	15.88 b
5	10.85 a	12.72 a	10.00 a	15.28 b

Means within a row not sharing the letter notation are significantly different at $P<0.05$.

Fall and winter consumption (or removals) of acorns by vertebrates accounted for an average of 87.7 percent of the cohort. Among the animals known to consume NRO acorns in Pennsylvania are white-tailed deer, black bear, gray squirrel, white-footed mice, wild turkey, and ruffed grouse. However, deer were probably the major consumer of acorns in these stands for reasons discussed by Steiner (1995). Of 14,260 germinated seedlings per acre, only 995 (7.0 percent) were still alive after 12 months and only 360 (2.5 percent) were surviving after five growing seasons.

From June 1991 (when seedling protectors were installed) until the termination of the study in September 1995, survival was 72.3 percent for protected seedlings and 15.8 percent for unprotected seedlings (a significant difference at $P<0.05$), indicating approximately 28 percent mortality over five growing seasons from causes other than deer browsing and approximately 56 percent additional mortality from deer browsing. Seedlings exposed to deer browsing exhibited no net height growth between 1991 and 1995 and even became significantly shorter at Firebreak. Seedlings protected from deer browsing grew a small (but statistically significant) amount before age 2 years, but the seedlings were no taller at age 5 than at age 2. Of 400 marked seedlings, the tallest after five growing seasons was only 28 cm in height, and no seedling emerged from the fern cover.

The following are our conclusions:

- Despite an unusually large crop of acorns in both stands, advance reproduction after five years was inadequate in terms of quantity and size of seedlings.
- Survival of the reproduction cohort as a function of time since masting resembles a negative power function: only 10.1 percent of the cohort still survived at approximately the time of germination (7 months after masting), 0.7 percent survived at seedling age 1 (19 months after masting), and 0.3 percent survived at the end of the study (60 months after masting). In other words, losses occurred very rapidly at first and more and more slowly as time went on.
- The failure of advance reproduction to develop in these stands is overwhelmingly attributable to mammal and bird consumption of *acorns*, since the majority (87.7 percent) of the reproduction cohort was lost in this manner. Circumstantial evidence indicates that deer are the major single factor in this loss. By contrast, only a small fraction of the entire reproduction cohort (0.6 percent in this study) was lost because of browsing of s*eedlings* after mid-summer of the first year.
- Although seedlings protected from deer had 72 percent survival after five growing seasons, they exhibited little

increase in height after June of the first year and none after age 2. In oak stands with a heavy ground cover of fern, deer fencing alone will not permit the development of strong advance reproduction. At a minimum, control of the ferns in addition to deer fencing will be necessary if the goal is to establish advance reproduction of NRO. A light thinning of the canopy may be necessary as well.

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CHARACTERISTICS OF NORTHERN RED OAK SEEDLINGS GROWN BY FAMILY IN A TENNESSEE NURSERY

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Abstract—Thirty-eight northern red oak (*Quercus rubra* L.) families were grown for 1 year in a commercial tree nursery in eastern Tennessee, and analyzed for first-order lateral root, height, diameter, and flush growth. Growth was negatively impacted by irregular fertilization and irrigation practices as well as heavy rains during the growing season. Average height and diameter growth were smaller than previously published standards for minimum oak planting stock size of 50 cm in height and 8 mm in root collar diameter (RCD). Provenance effects on nursery seedling growth were nonsignificant for progreny that was one generation removed from the original seed source, except for number of flushes during the growing season. Progeny from related mother trees, i.e., half-siblings, generally did not exhibit similar growth performance. Among-family differences in seedling growth suggests that nurseries should identify superior seed sources by evaluating the performance of progeny in the nursery. Root collar diameter was the best indicator of both height and number of first-order lateral roots (FOLR). Certain families had relatively high distributions of seedlings with above average RCD growth and low within-family variation in RCD. These results indicate that family mean should not be the sole criterion for selection of superior families. Additional nursery evaluations will identify mother trees consistently producing progeny with low numbers of FOLR, small heights, and small RCDs. These trees should be deleted from future seed collections or rogued, if in a seed orchard.

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FIELD PERFORMANCE OF IN VITRO PROPAGATED WHITE ASH MICROPLANTS

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Abstract—White ash (*Fraxinus americana* L.) can routinely be propagated by in vitro axillary shoot proliferation and in vitro rooting of microshoots; however, no reports exist on performance and clonal variation following field planting of microplants. To obtain preliminary estimates of among and within clonal variations, we established a small planting with twelve white ash clones in 1992. Ten non-stratified seed from fifteen individual tree collections were cut and germinated in vitro on a medium consisting of Murashige and Skoog (MS) salts and organics, thidiazuron, 6-benzyladenine, indole-3-butyric acid (IBA), 2 to 3 percent sucrose, and 0.7 percent agar. In vitro germinants exhibiting high axillary shoot proliferation rates were repeatedly subcultured to produce microshoots for in vitro rooting experiments.

Microshoots were pulsed for 1 week on quarter-strength MS medium supplemented with various combinations of 1 to 5 uM of IBA and naphthaleneacetic acid. Auxin-pulsed microshoots were then transferred to 0.25X MS medium without plant growth regulators where most microshoots produced up to seven adventitious roots within 1 to 2 weeks. When 6 to 8 weeks old, microplants were transplanted to soil in rootrainers and acclimatized under mist to greenhouse conditions. In the greenhouse, most microplants showed one flush of new shoot growth and extensive root development from the adventitious roots produced during the in vitro rooting phase. Microplants were overwintered in a cooler, then moved to a shade house for a month, and planted as in-leaf containerized stock in June 1992 at the SIU Tree Improvement Center in Jackson County, Illinois.

Overall the microplants averaged 2.7 adventitious roots per microshoot following in vitro rooting. Microplants from Family 14 averaged more than 3.2 adventitious roots compared to only 1.4 adventitious roots for Family 13 with the fewest roots. Average height for the original in-leaf planting stock ranged from 5 to 50 cm following field planting. Most microplants produced little additional height growth in the field the first growing season. Early in the third growing season, many microplants produced abnormal leaves and lateral shoots presumably in response to herbicide drift that resulted in multiple short shoots from the axillary nodes and narrow leaflet blades on short leaves. Net height growth averaged less than 20 cm for the third growing season. Severity of symptoms declined in the fourth growing season and most trees had recovered by the fifth growing season. Net height growth is averaging more than 70 cm per year and individual tree heights range from 0.3 to 5.0 m after five years. Multiple regression analysis of fifth year height indicated that the number of adventitious roots and severity of deer browse damage were not related to fifth year tree height; however, putative herbicide damage negatively affected growth.

Most clones have retained similar height rankings from the second growing season through the sixth growing season. Exceptions are Clone 14-10, one of the shortest clones at planting and now one of the tallest after six years, and Clone 15-2, the tallest clone at planting and now in the shortest one-third of the clones after six years. Clone 99 was one of the shortest clones at planting and is now above average in height. Interestingly, Clone 99 originated from organogenic callus, produced thin microshoots in vitro, and had a low in vitro rooting percentage. Initially, variation for height among clones was more than six times that of within clonal variation. After the six years, variation among clones was only twice that of the within clone variation.

Survival of the microplants of all clones after six years was between 70 and 100 percent except for Clone 6-3 where all microplants had died. Microplants of Clone 6-3 produced the fewest adventitious roots during the in vitro rooting period and had the lowest survival during greenhouse acclimatization. In 1997 the basal stem diameter averaged 6.3 cm for all the microplants. Microplants from clones of Family 2, 6, and 14 averaged 7.8 cm in diameter and were 3.8 cm larger than the microplants from Family 5 and 13, the families with the slowest growing microplants. Substantial variation existed among the clones within some families. For example, the basal diameter of Clone 6-1, one of the fastest growing clones, averaged 9.9 cm compared to only 5.7 cm for Clone 6-6 both from Family 6. Likewise, the basal diameter for Clone 4-6, another one of the fastest growing clones, averaged 7.8 cm compared to 4.3 and 4.7 cm for Clones 4-4 and 4-7 from the same single tree collection.

In conclusion, in vitro propagated microplants of white ash planted as in-leaf containerized stock can be successfully established in field plantings. Within clone variation for growth was significantly reduced when compared to among clonal variation. Additional field plantings using microplants from white ash exhibiting a wider range of in vitro axillary shoot proliferation rates and more clones per family are needed to test these preliminary results.

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