Presettlement floodplain forests along the Missouri and Mississippi Rivers were dominated by numerous tree species, including several hard mast species such as pecan (Carya illinoensis K. Koch), swamp white oak (Quercus bicolor Willd.), pin oak (Q. palustris Muenchh.), shellbark hickory (C. laciniosa Loud.), and black walnut (Juglans nigra L.) (Kozlowski 1984, Yin and Nelson 1995, Scott and others 1996). Since the 1800s, 70 to 90 percent of floodplain forest habitat has been lost in the continental United States due to natural changes in the river’s course, land clearing for agriculture, and the construction of dams and levees to protect valuable cropland and aid navigation (Knutson and Klaas 1995). These modifications have caused the reduction and localized disappearance of hard mast bottomland tree species (Yin and Nelson 1995).

Floodplain forests stabilize river banks, protect levees, and reduce flood impacts (Low 1994, Dwyer and others 1997, Geyer and others 2000). Floodplain forests also sequester carbon and reduce nutrient run-off by absorbing excess fertilizer applied to adjoining cropland (Sparks 1995). Furthermore, floodplain forests provide a diversity of habitat for fish and wildlife (Sparks 1995, Dwyer and others 1997, Yin and others 1997). Late-succession tree species, such as oak, provide mast for large and small mammals (Van Dersal 1938) as well as numerous species of birds (Hirsch and Segelquist 1978, Twedt and others 1999).

Although the Great Flood of 1993 degraded over 800,000 acres of cropland in the lower Missouri River floodplain by deposition of sand and scouring of soils, there now exists excellent opportunities for reestablishing hard mast species in floodplain forests (Low 1994). Hard mast-producing tree species such as swamp white oak, bur oak, pin oak, and pecan are important for numerous species of aquatic and terrestrial wildlife including deer, turkey, and waterfowl. These same species have difficulty

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becoming established due to a lack of locally available hard mast seed, intense competition from pioneer riparian tree species [e.g., silver maple (Acer saccharinum L.) and cottonwood (Platanus occidentalis L.)] and aggressive regrowth of annual weed species, inherent slow juvenile growth of hard mast species, continued flooding and deer herbivory (Buckley and others 1998, Dalrymple personal communication). An alternative to traditional bottomland reforestation with direct seeding or bare-root seedlings is the use of large containerized nursery seedlings produced under the root production method (RPM™) (Lovelace 1998).

ROOT PRODUCTION METHOD™

In the Upper Mississippi and Lower Missouri River floodplains, RPM™ seedlings have considerable advantages over traditional bare-rooted nursery stock including improved growth and survival, ability to be fall planted, terminal tips that reach above seasonal flood levels and browse height (≥ 5 feet), large basal stem diameter (≥ 0.6 inch), precocious flowering and mast production in the third or fourth year. Dey and others (2001) found that 4 percent of 30-month-old swamp white oak grown as RPM™ produced sound acorns the first year after planting in former cropfields along the Missouri River.

Selection of acorns from a superior mother tree can aid the production of desirable planting stock (Buchschacher and others 1991; Kormanik and others 1990, 1998, 2003). Most research on the effect of acorn size within species has shown that as acorn mass increases, seedling root collar diameter, height, leaf area, biomass, survival, and rate of emergence increases (Bonfil 1998, Ke and Werger 1999), and could be due to a greater amount of food reserves in the seed and cotyledons of heavier acorns (Tripathi and Khan 1990).

A few studies, however, have found no relationship between acorn size and seedling growth and morphology (Long and Jones 1996). If larger acorns emerge earlier and develop into larger seedlings, they may have a greater probability of reaching reproductive size in a competitive environment than smaller acorns (Seiwa 2000), but under competition free environments such as in a nursery, acorn size may be of little advantage.

Nursery planting stock is typically graded to determine acceptable size and evaluate competitiveness once outplanted. These criteria include height and root collar diameter (RCD) (Belanger and McAlpine 1975, Johnson 1984), number of first-order lateral roots (FOLR) (Kormanik and others 1998, Schultz and Thompson 1990) and root:shoot ratio (Romero and others 1986). More recently, root biomass has been considered one of the most important factors in the survival and establishment of nursery seedlings (Swamnath and Ravindran 1990) and therefore more attention has been given to producing seedlings with large, intact root systems.

A tree’s root system has the role of anchorage (Richards 1983, Schultz and Thompson 1992), storage of nutrients (Richards 1983; Bowen 1984, 1985; Schultz and Thompson 1992), water and salt absorption (May and others 1965, Aung 1974, Nielsen 1974, Richards 1983, Shultz and Thompson 1992), synthesis of nitrogenous compounds (Perry 1982), and production of hormones (Aung 1974, Richards 1983, Schultz and Thompson 1992). The root system can be modified by a number of cultural techniques. For example, basal root pruning in the nursery seedbed can increase the number of lateral roots and root tips (Schultz 1989; Schultz and Thompson 1990, 1992) that are important for seedling establishment (Struve 1990) and recommended for producing high quality planting stock. Air pruning of roots is another technique that is used in the RPM™ process to produce a large, dense, fibrous root system (Lovelace 1998).

Our overall purpose was to establish base line data on early morphological characteristics of bottomland oak species that may lead to precocious flowering in seedlings produced by the RPM™ process. Specific objectives of this research were

1) to determine the effect of acorn size and mass, and early seedling growth on morphological characteristics of 1-year-old seedlings and

2) to determine if a population of oak hybrids pre-selected for early and abundant acorn production would perform better than a randomly selected population of acorns when propagated under the RPM™ system.

MATERIALS AND METHODS

Acorns were collected in the fall of 1999 from two separate sources. One population consisted of randomly collected Q. bicolor acorns from Saline County, Illinois. The other acorns originated from Lincoln County, Missouri, from a population of half-sibling Q. bicolor x macrocarpa (Michx.), i.e., Q. x schuettei (Trel.). In the normal RPM™ process, acorns are graded by mass and diameter (Lovelace 1998). An aspirator is used to separate and discard the lightest
acorns below a pre-determined specific gravity, which are not considered sound. Remaining acorns are separated into two size classes, heavy and light, again based on pre-determined specific gravities.

Heavy and light acorns are then rolled across a plate with two different sized holes and separated into small and large size classes. Only the heaviest and largest acorns are stratified. In February, acorns are germinated in heated greenhouses in mesh-bottomed trays that allow for air pruning of the roots. After completion of the first shoot flush, seedlings are graded by precocious shoot growth and only the fastest growing seedlings (approximately the tallest 50 percent of germinates) continue in the RPM™ process.

After a series of transplants into increasingly larger bottomless containers placed on raised benches, seedlings are finally potted in shallow 3- or 5-gallon pots with a growth medium of rice hulls, pine bark, and sand that has 35 percent air space. Seedlings are then placed outside for the remainder of the growing season. Typical RPM™ seedlings attain heights of 4 feet or taller after 210 days of growth and they develop dense, fibrous root systems high on the root collar.

For this research, acorns were propagated into seedlings following the RPM™ process with the following important exceptions: as acorns and seedlings were graded, all seed and plant materials were separated and retained in their respective size classes instead of being discarded (i.e., both large and small seed, heavy and light seed, and precocious and non-precocious seedlings were retained). This resulted in eight treatments that represented all combinations of acorn mass (heavy or light), acorn size (large or small), and shoot growth (precocious or not).

Due to poor germination we had fewer seedlings than expected for some treatments and were forced to use two different experimental designs for each population. A 3 x 3 balanced lattice square design was used to examine all eight treatments (table 1). The Q. x schuettei population had four replications, each observation being the mean of two seedlings within a block. The Q. bicolor population also had four replications, but had fewer seedlings due to poor germination, and therefore, each observation was from one seedling. A 4 x 4 balanced Latin square design was used with only the four precocious treatments with one replication per population (table 2). Each block within each design contained three seedlings, from which a statistical mean was derived to obtain one observation.

At the end of the growing season, seedlings were destructively sampled and measurements were taken on seedling characteristics. Variables measured included root collar diameter (measured 2.5 cm above the root collar), height, root volume, root and shoot dry mass, and number of flushes. Root volume was measured by the displacement method described by Böhm (1979). Seedling mass was recorded after seedlings were oven dried and weighed on a top loading balance. SAS Version 8.2 (SAS Institute, Cary, NC) was used to conduct the analysis of variance, to test hypotheses relating acorn mass, acorn size, and initial shoot growth to first-year RPM™ seedling morphology, and to determine least significant differences (LSD) between treatment means.

RESULTS

Precocious shoot growth was not significantly related to first-year root or shoot size or morphology in either Quercus populations based on the Latin square design. Analysis of least significant differences showed that, seedlings from the light-small acorn treatment were consistently larger than any other acorn mass-size treatment combination (fig. 1).

Heavy Quercus x schuettei seed produced first-year seedlings with significantly larger root mass and volume, and shoot length and mass that did seedlings originating from lighter seed (table 3). Other researchers have found that seed mass is positively correlated or significantly related to early seedling growth (i.e., Eytingen 1917, Korstian 1927, McComb 1934, Rice and others 1993, Bonfil 1998).

A noted exception to the above occurred in the seed size treatment in the Quercus bicolor population, where seedlings from smaller acorns had...
Figure 1.—The effect of acorn mass and size and early growth on first-year morphology of RPM™ container-grown seedlings from two Quercus populations; Treatments: HS = heavy-small, HL = heavy-large, LL = light-large, LS = light-small. Means +/- standard error. Open bars = Quercus bicolor; shaded = Q. x schuettei.
significantly greater root mass and volume, and shoot mass and size than seedlings from larger acorns (table 4). The small acorns from the *Q. bicolor* population were found to have significantly larger values for seedling height and root volume than large acorns. There was no other significantly different main or two-way interaction effects (i.e., heavy-large vs. heavy-small) found.

Significant differences were found among all eight treatments (i.e., all combinations of acorn mass and size, and precocious shoot growth) that were analyzed with the lattice square design from both *Quercus* populations, but these differences were not consistent for one particular treatment with one exception (fig. 2). For example, the heavy-small-precocious treatment from the *Q. bicolor* population was consistently, but not always, significantly larger than the other treatments from that population.

*Q. bicolor* seedlings from public collection were consistently, but not always, significantly larger than the hybrid *Q. x schuettei* seedlings (figs. 1 and 2). Overall, however, there were no significant differences in seedling morphology between *Q. bicolor* and *Q. x schuettei* seedlings after one growing season. *Q. bicolor* seedlings did have a greater amount of variation as indicated by the large standard errors than did the half-sibling *Q. x schuettei* seedlings.

**DISCUSSION**

In previous studies, Rodger (1920) found no effect of acorn mass and size on seedling characteristics, but his sample was too small to make definitive conclusions. Long and Jones (1996) also found no effect of acorn mass and size on seedling characteristics, but suggests that poor control over acorn moisture content (in their study and others) and genotypic variation on seed size could have influenced their results. Studies that involved the removal of a portion of the acorn prior to sowing, or removal of the cotyledons shortly after germination, found that seedling growth was more affected by poor soil nutrition than by loss of acorn mass (Sonesson 1994, Anderson and Frost 1996). Furthermore, it has been speculated that acorn size may be more important in attracting wildlife for seed dispersal than for first-year-growth (Stiles 1980, Anderson and Frost 1996).

Conflicting results may also be due to differences in propagation methods. Traditionally seedlings have been grown in seedbeds as bare-rooted nursery stock under less than ideal conditions for rapid growth. Containerized RPM™ seedlings grown in the greenhouse under optimal growing conditions are under no stress and are absent of competition relative to seedlings grown in the wild or in nursery beds. Larger seeds may have advantages over smaller seeds if both are grown with competition (Wulff 1986, Bonfil 1998), but under low stress and competition-free environments, little appears to be gained from larger seeds.

In addition to the absence of stress or competition, RPM™ seedlings have an extremely dense, fibrous root system with numerous lateral roots providing a greater amount of surface area for the absorption and utilization of oxygen, water, and nutrients than bare-rooted seedlings (Lovelace 1998). Optimal growing conditions provided by the RPM™ system facilitate the production of a dense, fibrous root system and remove the effect of acorn mass, size, and precocious shoot growth on oak seedlings after one growing season.

**Table 3.—The effect of acorn mass on first-year morphology of RPM™ containerized *Quercus x schuettei* seedlings. Values are means ± 1 standard deviation.**

<table>
<thead>
<tr>
<th>Variables</th>
<th>Heavy</th>
<th>Light</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>RCD1 (mm)</td>
<td>12.4 ± 1.5</td>
<td>10.3 ± 0.4</td>
<td>No</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>100.7 ± 8.9</td>
<td>81.0 ± 2.2</td>
<td>Yes</td>
</tr>
<tr>
<td>Root volume (ml)</td>
<td>122.9 ± 27.7</td>
<td>88.3 ± 9.4</td>
<td>Yes</td>
</tr>
<tr>
<td>Shoot dry weight (g)</td>
<td>38.8 ± 6.3</td>
<td>23.6 ± 3.8</td>
<td>Yes</td>
</tr>
<tr>
<td>Root dry weight (g)</td>
<td>44.7 ± 11.6</td>
<td>30.4 ± 4.4</td>
<td>Yes</td>
</tr>
<tr>
<td>No. of flushes</td>
<td>4.6 ± 0.5</td>
<td>4.2 ± 0.5</td>
<td>No</td>
</tr>
</tbody>
</table>

1 Root collar diameter (measured 2.5 cm above the root collar)  
2 Means within a row differ significantly at the 5% level.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Large</th>
<th>Small</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>RCD1 (mm)</td>
<td>12.5 ± 1.2</td>
<td>12.6 ± 0.9</td>
<td>No</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>103.8 ± 12.1</td>
<td>112.8 ± 13.2</td>
<td>Yes</td>
</tr>
<tr>
<td>Root volume (ml)</td>
<td>113.3 ± 27.1</td>
<td>128.3 ± 21.2</td>
<td>Yes</td>
</tr>
<tr>
<td>Shoot dry weight (g)</td>
<td>39.1 ± 10.0</td>
<td>42.9 ± 10.7</td>
<td>No</td>
</tr>
<tr>
<td>Root dry weight (g)</td>
<td>46.7 ± 11.2</td>
<td>52.6 ± 9.2</td>
<td>No</td>
</tr>
<tr>
<td>No. of flushes</td>
<td>4.9 ± 0.4</td>
<td>5.0 ± 0.3</td>
<td>No</td>
</tr>
</tbody>
</table>

1 Root collar diameter (measured 2.5 cm above the root collar)  
2 Means within a row differ significantly at the 5% level.
Figure 2.—The effect of acorn mass and size and early growth on first-year morphology of RPM™ container-grown seedlings from two Quercus populations; Treatments: LSN = light-small-non-precocious; LSP = light-small-precocious; LLN = light-large-non-precocious; LLP = light-large-precocious; HSN = heavy-small-non-precocious; HSP = heavy-small-precocious; HLN = heavy-large-non-precocious; HLP = heavy-large-precocious. Means +/- standard error. Open bars = Quercus bicolor; shaded = Q. x. schuettei.
Population Differences
We evaluated the influence of genetics and the RPM™ process on seedling morphology after one growing season by comparing RPM™ seedlings from two different seed sources. Acorns of the hybrid *Q. x schuettei* were expected to perform better than the *Q. bicolor* acorns from public collections obtained from the state nursery. However, we found no significant differences in seedling morphology between populations and a few significant differences among treatments. *Quercus bicolor* seedlings were consistently larger than *Q. x schuettei* seedlings, but differences were not statistically significant.

There is insufficient evidence to conclude that observed differences in seedling morphology between *Q. x schuettei* and *Q. bicolor* acorns after one growing season are indicative of future outplanting success and precocity. Seed collected from superior seed trees have been shown to have increased viability and germinate earlier (Buchschacher and others 1991), growth responses that could prove advantageous in a natural setting (Seiwa 2000). Timing of germination, as well as values of all measured variables, varied more in *Q. bicolor* than in half-sibling *Q. x schuettei* acorns, possibly due to the greater degree of genetic diversity found in the randomly collected *Q. bicolor* acorns (Mercier and others 1996).

**IMPLICATIONS**

Propagation of nursery seedlings requires a large supply of acorns and is often adversely impacted during years of poor seed production. If seedlings were grown under optimal conditions such as in the RPM™ process, it would prove advantageous to retain all viable seed for the production of high quality nursery seedlings. Nurseries using a process similar to RPM™ could produce a greater number of seedlings by eliminating the process of discarding lighter, smaller, and slow-germinating acorns.

A result of retain all viable seed would be that plantings contain a greater amount of genetic diversity, which is advantageous in highly competitive or disturbance prone environments such as floodplains. Variation in germination rates could eliminate complete seedling mortality from late spring floods, where slow-germinating seed would sprout after floodwaters had receded, thus providing some regeneration (Streng and others 1989).

A longer-term objective of this research is to determine if selection for seed size or precocity leads to early and consistent mast production after outplanting. Seedlings not destructively sampled during this study are being outplanted to monitor precocity in mast production.

**ACKNOWLEDGMENTS**

Without the assistance from all the individuals at Forrest Keeling Nursery, none of this would have been possible, and for that we are grateful. The technical assistance of the Department of Forestry and the Center for Agroforestry at the University of Missouri and the Missouri Department of Conservation State Nursery is also gratefully acknowledged. This work was funded under cooperative agreement CR 826704-01-0 with the US EPA. The results presented are the sole responsibility of the P.I.’s and may not represent the policies or positions of the EPA.

**LITERATURE CITED**


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