

IDENTIFYING AND ENRICHING FOR AMERICAN BEECH TREES THAT ARE RESISTANT TO BEECH BARK DISEASE

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Beech bark disease begins when bark tissues attacked by the scale insect (*Cryptococcus fagisuga* Lind.) are rendered susceptible to infection by fungi of the genus *Nectria*, leading to the weakening and eventual death of the tree. Some American beech (*Fagus grandifolia*) trees remain disease free in stands long-affected by beech bark disease and challenge trials have shown that they are resistant to the scale insect (Houston, 1982). Increasing the number of resistant beech trees while reducing the proportion of susceptible trees is currently thought to be the best management approach to minimize the overall impact of beech bark disease (Mielke et al. 1986). However, even in heavily infested areas, trees that remain clear of scale may be “escapes” and not truly resistant. We have set out to design a research program with two related goals; 1) to determine if the artificial inoculation technique (Houston, 1982) is an efficient way for distinguishing between resistant and susceptible trees and 2) to develop methods to propagate resistant trees once identified, through both vegetative techniques and cross-pollinations. The following is a summary of our research progress to date.

Identifying Resistance

Previous work by David R. Houston reported the development of a technique to artificially infest beech with the beech scale (Houston 1982). This technique was successfully used to artificially infest seedlings as young as one year old and to confirm the resistance of older scale-free trees. However, appropriate tests to determine whether this technique could successfully distinguish between resistant and susceptible juvenile root sprouts and seedlings are needed.

Field trials

Insect eggs or adult, egg-laden insects were collected from foam traps that were put in place the previous year (2002) and used to set up field challenge experiments at both the Allegheny National Forest (ANF) in Pennsylvania and Ludington State Park (LSP) in Michigan. A cluster of 12 putatively resistant trees was included in the ANF study along with 2 susceptible controls. In LSP, a cluster of 20 putatively resistant trees was challenged along with a cluster of 8 susceptible trees. The DBH of the trees ranged from 2.4 to 10.3 inches. In both areas, the clusters of trees appeared to be root sprouts originating from the same parent source. Tissue samples taken from each individual tree will be used to extract DNA and, using molecular markers, determine which individuals are truly clonally identical. Fifty adult, egg-laden insects (average of 8-10 eggs per adult) or 500 individual eggs were counted out and placed onto moistened polyurethane foam pads that measured 4 inches x 4 inches. The pads were placed up against the bark and tied into place. The number of reproductive scale colonies established will be determined in the summer of 2004. By looking at clonal replicates of varying size and age, an indication of the amount of variability produced using the artificial challenge technique will be determined.

Seedlings

An artificial challenge experiment using both full- and half-sib seedlings has also been set up. The crosses and resulting seeds will be discussed in additional detail in the next section. The challenge experiment is being performed cooperatively with the Holden Arboretum in Kirtland, Ohio. A total of 438 six-month old seedlings were challenged. Polyurethane foam pads measuring 1 inches x 3 inches were moistened and either 30 egg-filled adults or 300 individual eggs were counted out and placed on the foam prior to adhering it to the stem of each seedling. Six weeks after the pads

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were put into place, a handful of seedlings from susceptible parents were checked and crawlers had already hatched from the eggs and attached to the bark. However, not until summer of 2004 will the seedlings be inspected for the presence of reproductive scale colonies.

Propagating Resistant Beech

To minimize the impacts of beech bark disease, it is necessary to have a source of resistant American beech to be used in pre-emptive plantings ahead of the disease front as well as for restoration of aftermath forests. Our efforts have focused both on vegetative means of propagation and sexual reproduction. Techniques involving vegetative propagation are critical for preserving superior, resistant trees. However, for long term success, it is necessary to maintain genetic variability as well as disease-resistance.

Softwood cuttings

Softwood cuttings were taken from candidate resistant trees at LSP in late May, 2003, at the beginning of leaf emergence. Cut stems were soaked overnight in either a solution of 1.2 mM IBA in water or 1.2 mM IAA in .02 N NaOH. After the overnight soak, they were placed in rooting media (3 parts sterile play sand, 2 parts perlite, and 1 part Rediearth) and kept under mist. Roots began to sprout after 21 days and after 30 days. However, in order to allow maximum root growth the cuttings were not transplanted until 60 days. Unfortunately, by this time root rot had set in and a number of cuttings were lost. Table 1 shows the number of rooted cuttings that achieved new shoot growth. By optimizing the rooting media and misting protocol for maximum drainage and by potting rooted cuttings earlier we are hopeful that the percent of successful rooted cuttings will be greatly improved.

Table 1.—Success rates of softwood cuttings of American beech.

Treatment	Rooted (%)	Shoot growth (%)
IBA	22	38
IAA	53	19

Open-Pollinated Seeds

Open-pollinated seed was obtained from two of the parents used in the cross-pollinations study, the resistant tree 1506 and the susceptible tree 1504. Open-pollinated seed was also collected from the susceptible tree 1511 and from a resistant tree located in Maine. The tree from Maine is of particular interest because it is located in a region that has been intensely managed for beech bark disease—susceptible trees have been removed. Therefore, there is a significant chance that the seeds from this tree are derived from resistant pollen donors. Between 24-35% of the seeds collected from trees at Ludington State Park (1506, 1504, and 1511) were full. This figure is only slightly higher than the reported 13 - 29% of sound nuts collected from 20 trees in East Lansing, MI (Gysel, 1971). The percentage of seeds collected from the Maine tree that were sound was much higher (75%) than those collected from Ludington State Park. This value was comparable to the values reported by Leak & Graber (1993) for seed collected from beech in the White Mountain National Forest. Over a 6-year period of time seeds from this area were consistently between 75 and 88% sound. The lower number of sound seed collected in Michigan, compared to those collected in Maine, may be attributable in part, to the fact that Ludington State Park is toward the northern most border of the natural range of American beech.

Cross-Pollinated Seed

The results of the controlled cross-pollinations are listed in Table 3. Overall, the germinative capacity (the percent of sound seed that germinated) was variable, ranging from 12 to 84 percent.

However, compared to open-pollinated seeds from this study, the average germinative capacity of cross-pollinated seeds was greater. The percent of barren seed was similar in both the cross-pollinations and the open-pollinations, with the exception of the 1504 ♀ x 1506 ♂ cross which produced a slightly lower, 13% sound seeds. This similarity between cross-pollinated and open-pollinated seed production provides an indication that the pollination bagging process did not negatively effect seed development. The production of a lower percentage of sound seed in the 1504 ♀ x 1506 ♂ cross could possibly be attributed to an incompatibility between the parents or that 1504 does not produce vigorous pistillate flowers. Open-pollinated 1504 flowers produced seed with a low germinative capacity and the two controlled crosses that used 1504 as the maternal parent produced seed with lower germinative capacities compared to seeds from crosses that used 1505 or 1506 as the maternal parent. Differences in compatibility between pairs of parents is not uncommon and identifying compatible mating pairs is an important part in developing seed orchards for tree improvement (Lambeth 1993).

Table 2.—Controlled cross-pollinated seed

Cross (♀ x ♂)	Seeds					%Full	Germinative Capacity	Total No. Plants
	Full	Germinated	Rotten	Empty	Total			
1506 (S) x 1504 (R)	11	84	0	146	241	39	81 %	77
1504 (R) x 1506 (S)	49	31	10	585	675	13	12 %	11
1504 (R) x 1501 (I)	35	0	0	98	133	26	37 %	13
1505 (R) x 1504 (R)	28	33	0	170	231	26	84 %	51

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