# **Bay Journal**

# **Back-cross American chestnut project raising hopes for tree's restoration**

Volunteers breeding, planting thousands of disease-resistant seedlings

Leslie Middleton on October 26, 2015



*Matt Brinckman, with The American Chestnut Foundation, pollinates the flowers of a pure American chestnut that has survived the blight to create a family line adapted to local conditions. (Leslie Middleton)* On a late June morning, Matt Brinckman reached into the branches of a 50-foot American chestnut tree adorned with 100 or so small white paper bags — each of which fully covered the flower at the end of the

"They are receptive," he called down to Jerre Creighton, research forester with the Virginia Department of Forestry, who was running the bucket truck that lifted Brinckman up into the tree.

Brinckman, a regional science coordinator with The American Chestnut Foundation, removed one of the bags and dipped the flower into a container the size of a film canister to cover the female parts with pollen from a tree specially bred by the foundation.

He clipped the paper bag over the pollinated flower so that when he returns in the fall, he'll be able to identify the seeds (nuts) that have resulted as a cross between this American chestnut tree — one of the few in the area that has survived to produce nuts — and a tree of known progeny raised at the foundation's Meadowview Nursery.

Warren Laws, a member of the foundation's Virginia chapter, observed from the ground. During June and July, Laws coordinated volunteers here on the eastern flank of Virginia's Blue Ridge Mountains to work in chestnut "breeding orchards." Occasionally, Brinckman, who works out of an office at the Department of Forestry, helps out. Some days, Creighton brings the forestry department's bucket truck for getting up into the trees. Other days, the local electric company donates one.

Over the course of a few summer months, the simple act of manual pollination will be replicated by the foundation's volunteers and partners on hundreds of trees, most of them in breeding orchards under the care of state chapters from Alabama to Maine, the historic range of the American chestnut.



The mission of The American Chestnut Foundation is to "restore the American chestnut tree to our Eastern woodlands to benefit our environment, our wildlife and our society." The key to this restoration is to develop a tree resistant to the chestnut blight that virtually destroyed all of the American chestnuts in the

first half of the 20th century yet also retains the form, growth characteristics and forest adaptation characteristics of the native pre-blight tree.

This visionary-scale restoration is being undertaken by a foundation with a full-time staff of only 18. "Our organization is highly leveraged," said Lisa Thomson, president and CEO of the foundation, which is headquartered in Asheville, NC.

"We couldn't do what we do without our volunteers — over 6,000 of them — or without our many partners," Thomson said. Partners include scientific labs, academic institutions, industry, foresters, other nongovernmental organizations, and state and federal agencies. Each contributes expertise, resources and support. Partners have advanced the science of plant breeding and forest restoration; provided land for breeding and seeds orchards; and helped the foundation grapple with issues ranging from genetic modification to hybridization methodology.

After a little more than 30 years of work, the foundation is poised to start introducing a disease-resistant chestnut into the wild.

## The beginning of the blight

In 1904, a pathogenic fungus, Cryphonectria parasitica was discovered on an American chestnut tree in the New York New York Zoological Garden. It had been inadvertently imported on a chestnut tree from Asia. The Asian tree had co-evolved with the fungus — soon to be known as the chestnut blight — and had natural immunity to it.

Not so for its cousin, the American chestnut, prized by people and wildlife who relied on the abundance of seed these towering trees reliably produced. Called "a perfect tree" by many — it grew straight, tall and fast, and provided rot-resistant lumber for houses, fences and furniture. It was also the dominant and foundational tree of the Appalachian forest.

Lacking immunity to the blight, the American chestnut (Castanea dentata) is easily infected by the organism that is spread by air and grows under the bark, where it destroys the tree's vascular system and kills it by girdling. Though the tree fights back by growing telltale cankers around infection, all but a rare few die. The blight spread through the range of the American chestnut in the early 1900s, killing an estimated 40 billion trees.

By 1950, almost all of the American chestnuts were gone. The forests, and the people who relied on them, adjusted to a changed environment.

Today, wild American chestnuts can be found in the forest understory along much of the tree's historic range, most resprouted from the trunks of dead or felled trees. This is possible because the blight does not affect the tree's roots. But the once dominant tree is unable to compete in the totally transformed forest that still harbors the blight. And only a few will survive long enough to flower and produce a few nuts before succumbing.

Protection, and eventually restoration, started almost immediately, as chronicled in Susan Freinkel's "American Chestnut: The Life, Death, and Rebirth of a Perfect Tree." Because Chinese and Japanese chestnuts tree varieties were immune to the blight, researchers looked for ways to transfer their immunity to American chestnuts.

The U.S. Forest Service attempted to graft scion wood from surviving American chestnuts onto Chinese chestnut rootstock. In the 1950s, scientists tried to induce a mutation through irradiation that would confer disease resistance to the tree. Others experimented with "hypo-virulence," injecting a virus known to combat the chestnut blight.

But in the late 1970s, "back-cross breeding" between the American chestnut and blight-resistant Asian chestnuts started to show promise. Three scientists who were pursuing this method — Philip Rutter, David French and the late Charles Burnham — started The American Chestnut Foundation in 1983, and by 1989, had established the Meadowview research farm.

Back-cross breeding is a hybridization technique used to increase the chances that desirable traits from one compatible species will be conferred to another.

Successive crosses of offspring from each selected pollination back to a pure American chestnut produce trees that have a higher chance of possessing at least two of the three Asian genes thought to confer blight resistance. The first cross creates an offspring that is genetically one-half American chestnut. The second cross is between the 50-50 hybrid back to another pure American chestnut to increase the "American" genetics. This results in a tree that is 75 percent American. Two more back-crosses result in a 87.5 percent then 93.75 percent American chestnut.

But the only way — so far —to test immunity is to grow the trees for several years and then purposefully inoculate them with the chestnut blight. At each stage, the few trees that exhibit blight resistance are allowed to grow and contribute to the growing number of family lines of trees that are part Chinese chestnut, adapted to the region, and possess the "American" tree's characteristics: rapid, straight growth; open crown; leaf; bark structure; and appearance.

While much of the research and mass seed production takes place at Meadowview, the important work of breeding for genetic diversity and local adaptation takes place at more than 300 breeding orchards under the care of the foundation's state chapters.

To keep these orchards growing takes committed volunteers who manage pollination, weed, inoculate, and ultimately cull trees that can't resist the blight.

Chapter organization varies. Virginia's five branches correspond to different state regions, with more forming each year. The Maryland chapter is organized around its various projects — multiple breeding orchards, demonstration plantings and special projects, including one at the Patuxent Institution in Jessup, where inmates have been raising chestnut seedlings for plantings around the state.

"Because we are volunteer-led, each chapter is slightly different," said Sara Fitzsimmons, who is Brinckman's counterpart in the foundation's north central region. "As we grow the number of trees to plant for restoration, we have to grow our volunteer base, as well as staff, chapters and partnerships." But maintaining the grassroots nature of the organization is important, she said, even as tens of thousands of trees are being planted a year. "We're hosting a program this year showing how people can extract DNA from chestnuts. People will be amazed that you can do this in your kitchen."

### **Reason for optimism**

Cathy Mayes, a past president of the Virginia Chapter who serves on the foundation's national board, said ,"What I've found so striking is that here is a project that will not produce overnight results, where volunteers may never live to see the results of their efforts." But they are passionate, she said, about doing something positive for the future. And they are optimistic.

There is reason for optimism. In 2006, the foundation's breeding program achieved what they call the Restoration Chestnut 1.0, and they have now produced more than 120,000 seeds that are being used in trials geared toward understanding the best ways to reintroduce the tree to Eastern forests. The 500 trees planted in three National Forests in the Southeast appear to be thriving, though full testing could take five years or more.

This is the next frontier for the foundation. "No one really knows how the chestnut will fit into the ecology of the forest," Creighton said. Forestry management as a science was just getting going as the chestnut was being wiped out. There is little scientific documentation of the forests from the era when the American chestnut was the dominant species.

Creighton, whose research responsibilities include the long-leaf pine and other commercially valuable trees in Virginia, also tends some American chestnut trees at Lesesne State Forest in Nelson County, where 400 acres were donated specifically for chestnut research in the 1940s. There is a dense stand of American chestnuts — some almost 50 years old — the survivors from almost 11,000 seedlings that came from early cross-breeding experiments in the 1960s in Connecticut, before the rigorous documentation necessary for back-crossing was developed.

Unlike The American Chestnut Foundation's program where family lines are meticulously documented, the trees are being used to build up more lines of genetic diversity, albeit without the same level of record-keeping. "It's a different approach, but still valuable," Creighton said, especially as federal and state funding for chestnut research has waxed and waned over the years, making it difficult to sustain research.

### Mapping the genome

While slow and steady back-cross breeding is the backbone of the foundation's work, advances in molecular genetics have offered techniques to advance the development of a blight-resistant tree. Scientists believe that two or three nondominant genes in the Chinese chestnut confer blight resistance. Meanwhile, the federally sponsored workgroup, Forest Health Initiative, is mapping the Chinese chestnut's genome.

"If we can identify the gene or genes, we may be able someday to sample the seedlings in the breeding program to see if they have the right genetic makeup," Brinckman said. Rather than waiting three to five years, inoculating the trees and observing which ones survive the blight, the foundation's back-cross breeding cycle would be dramatically accelerated.

Since its organization in 1990, the New York chapter has supported the development of a blight-resistant tree using transgenic (gene transfer) biotechnology. In 2013, researchers at State University of New York, College of Environmental Science and Forestry reported that they were able to transform the American chestnut tree using a gene from wheat that confers blight resistance that is equal to or better than that of the Chinese chestnut.

A genetically modified tree is years away from proving itself as a restoration option and must run the gamut of federal review — and public acceptance — before it can be used in restoration efforts. Though not everyone involved in chestnut restoration applauds this kind of genetic modification, the foundation has publicly endorsed the research as complementary to its back-cross breeding program.

"We are moving into large-scale seed production and thinking about how to reintroduce the chestnut tree," said Thomson, the foundation's president since early 2015. There are active workgroups in the foundation exploring how and where to target large-scale restoration efforts, but significant questions remain about just how to integrate the tree into the woods.

Tom Dierauf, a retired forest researcher in Virginia who was involved in planting the 10,000 hybrid seedlings at Lesesne State forest, thinks the restoration will not be easy.

"Chestnut trees evolved with fire, and probably became dependent on fire for regeneration," Dierauf said. "Fires probably provided open understories with sunlight that seedlings needed to grow satisfactorily."

Chestnuts may be like oaks, Dierauf said, which today are gradually being replaced by maples, beeches, and other more shade-tolerant species, in part because of the lack of fires in our forests.

Chestnut restoration may require mechanical or chemical control of woody competition, and the planting of seedlings may always be required.

But these unknowns are not slowing the foundation's quest to return the American chestnut to the Appalachian landscape, nor quelling the optimism of its members."What I — and everyone involved with the foundation — like about the chestnut restoration," Thomson said, "is that rather than being on the defensive, we as environmentalists are on the offensive."

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