ScienceWatch –Genetic Engineering Can Restore the Oak-Chestnut Forest*

"We call this a century project. To get it to look even somewhat like it did before the blight is going to take centuries, it's for the next generation—planting a tree you'll never enjoy the shade of." – C. Maynard


Transgenic seedlings resist blight.

No one alive today knows what our forest once looked like—the oak-chestnut forest that once ranged across the eastern US. That's because a blight-causing fungus (*Cryphonectria parasitica*), introduced in 1904, killed the 3.5 billion American chestnut (*Castanea dentate*) trees living from Maine to Louisiana. This keystone species once accounted for 25% of our hardwood forest. The nuts fed many forest species, including vast numbers of passenger pigeons. The massive120-foot-tall trees provided nesting sites for birds and rot-resistant wood for railroad ties, telephone poles, barns and churches. Forty years after we accidentally brought the fungus here the American chestnut was gone.

The airborne parasite enters a wound and forms a canker that secretes oxalic acid. The acid kills the surrounding cambium, and the fungus feeds off the dying tissue. The growing canker eventually girdles the tree, killing it. The root system remains unharmed so new sprouts emerge but they too soon succumb. Today the American chestnut is an occasional shrub in the forests it once dominated. The loss was stunning.

Billions in today's dollars were also lost. The nuts provided a profitable subsistence living for settlers, while others worked in the mills of a thriving lumber industry. The dead trees left an open landscape that became doubly scarred by anthracite coal mining, which people in Appalachia were forced to turn to for a living. As coal mining declined, government regulations required mining companies to "restore" the land. But that resulted in open pasture, not forests. Recent efforts to restore the forest have shown, as might be expected, that the thin, acidic, well-drained soils best support chestnut trees. But planting trees that will never mature is a short-term solution.

In 1980 The American Chestnut Foundation (TACF) sought to generate resistant trees that could restore the forest. They hybridized the American chestnut with its Asian relative (*C. mollissima*), which is resistant to the fungus, and began backcrossing the hybrids with their American parent to generate "Americanized" resistant trees. Four generations of backcrosses produced hybrids that contain only 1/16 of the *C. mollissima* genome, but multiple genes appear to control resistance so only 20% of the hybrids are resistant. Moreover, the hybrids can't compete well in the forest.

In 1990 forest biotechnologists William Powell and Charles Maynard, SUNY College of Environmental Science and Forestry, used a newly developed method to selectively transfer the gene(s) for resistance from Asian to American trees. They discovered that the Asian chestnut's resistance stems from its ability to destroy the fungus-synthesized oxalic acid. Without its key weapon the fungus can't kill healthy tissue so it's restricted to the original infection site and the tree continues to grow.

By 2013 they created a transgenic seedling, Darling 58, genetically identical to the original American chestnut but with one extra gene that makes it blight resistant. The gene (Oxo) they inserted into American chestnut embryos comes from bread wheat. It makes oxalic acid oxidase, an enzyme that disarms the fungus by destroying oxalic acid. Oxo is a common antifungal defense gene found in cereals and many other food crops such as strawberries, beets and peanuts.

In 2020 Powell's team began tackling the regulatory hurdles required to release a genetically modified organism (GMO) into the wild—a tough nut to crack. First they showed their transgenic trees were safe. They fed bees pollen containing oxalic acid oxidase. They studied the effects on tadpoles of leaves left in water. They looked for effects on beneficial soil fungi. No adverse effects were found and chemical analyses on chestnuts from transgenic trees showed no differences from chestnuts produced by unmodified trees.

Next they applied to the USDA, requesting deregulation of Darling 58. By then TCAF admitted their hybridization efforts weren't working and they embraced the application. After its review the agency opened a public comment period. Over 60 percent supported the application. The team still must file additional petitions with the FDA on the food safety of transgenic nuts and with the EPA concerning the environmental impact of the transgenic trees. If approved Darling 58 would be the first GMO released with the intention of spreading in the wild. It will be a long process.

But now additional support comes from an unlikely ally. In the March/April 2021 issue of the Sierra Club magazine journalist Kate Morgan writes, "For its part, the Sierra Club has counseled caution, but it sees Darling 58 as a low environmental risk, since it would be closely monitored. … The Club noted that while it still 'carries some uncertainties,' genetic engineering could with proper precautions produce an organism that 'provides an environmental benefit.'"

It remains to be seen if the oak-chestnut forest will once again flourish and few of us alive today would see it. But let's hope.

Saul Scheinbach

*For more information see "Resurrection of the American Chestnut" (www.hras.org/sw/swjan2019.htm), and "Can Genetic Engineering Bring Back the American Chestnut?" (www.nytimes.com/2020/04/30/magazine/american-chestnut.html).