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# **Progeny Testing an Invaluable Tool**

"Progeny Testing is an Invaluable Tool for Inferring Resistance and American Traits Among Our Most Advanced Hybrid American Chestnuts"

By Tom Saielli, Mid-Atlantic Regional Science Coordinator

American chestnut, once a tall, magnificent tree with broad crowns, has been reduced to scattered stump sprouts growing at the bases of long-dead trees, the result of the accidental introduction of the fungal pathogen, Cryphonectria parasitica. Although there are many approaches to combatting C. parasitica and returning American chestnut to its former prominence, TACF focuses on backcross breeding. During backcross generations, phenotypic selections are somewhat straightforward, and although some uncertainties may exist regarding the selection process, there is some confidence in this methodology. However, backcross trees are eventually intercrossed to produce BxF2 seeds, which are planted in seed orchards at 150 seedlings per family, per plot...and making selections at this level is significantly more challenging. How do we select the one best tree from among 150 seedlings?

At our research farms in Meadowview, VA, over 60,000 trees have been planted in two large seed orchards (Duncan and Wagner) and after years of culling fiftythousand susceptible trees, thousands of trees still remain. Our goal is to continue the process of culling the susceptible trees so that only the most resistant trees remain – assuring that the BxF3 progeny produced have the best chance for long-term survival.

We can distinguish the most susceptible trees through artificial inoculation, but that method is not precise enough to select the most resistant trees. The challenge: How to make the final selections and end up with only the most resistant trees in the orchard? One method is to screen progeny.

Progeny Testing refers to a test of selective traits of an individual, done by observing the progeny of that individual. This type of test is significant because it helps to



Students at Virginia Tech extract DNA from chestnut leaf samples

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# A Cure for Nature Deficit Disorder

# By Cathy Mayes, Virginia Chapter President

You probably remember *Last Child in the Woods* by Richard Louv, which was such a rage a decade ago extolling how important it is for people to get outdoors, away from cars, and crowds, and subdivisions. About this time of year, many of us are especially missing the outdoors. Short days, biting winds, and dormant plants conspire to make us crabby.

Fear not: The American Chestnut Foundation has a therapy program for those of you suffering from Nature Deficit Disorder this winter. We have a long list of outdoor projects to begin as the weather warms. We invite you to join us whenever you can.

Here's a short listing of the projects on tap, sorted by geography. No dates yet: we're dependent on weather and logistics, but we've started a list of people to contact when the dates are set. If you'd like to be contacted, get in touch with our office.

# **Central Virginia**

- Fill-in planting at Middle Mountain orchard in Albemarle County
- Culling trees at Stony Brook Orchard in Madison County
- Inoculating trees at Innisfree in Albemarle County
- Inoculating trees at The Ranch in Culpeper County
- Orchard maintenance at Claytor Nature Center in Bedford County

# Northern Virginia

- Fill-in planting and tree measurement at Rockley Foundation Orchard in Fauquier County
- Fill-in planting at Smithsonian Conservation Biology Institute (SCBI) in Front Royal
- Seed orchard establishment at Banshee Reeks Nature Preserve, Loudoun County
- Seed orchard establishment at Sky Meadows State Park, Fauquier County
- Culling trees at Old Whitewood Orchard in Fauquier County
- Inoculating trees at Mt. Zion Orchard in Loudoun County
- Culling trees at Dam Orchard in Fauquier County

## Southwest Virginia

- Fill-in planting at Catawba Orchard in Roanoke County
- Measuring growth and survival of trees at test planting sites

Contact information: email <u>vachest-</u> <u>nut@verizon.net</u> or phone (540) 364-1922.



Working at the SCBI Orchard

### Раде З

# Report from Meadowview

By Fred Heberd

As I write this in early March, the weather has turned cold again, but one can tell spring is coming. The daffodils are up and out due to the warm weather in February, and the sun is brighter and warmer, coming up earlier and setting later. So it's time to plant chestnuts!

The Virginia Chapter will be planting its first crop of B3-F2s this year, which is an exciting transition to see. These nuts were harvested from Virginia orchards where the trees had been tested for blight resistance by inoculation with the blight fungus. Only the most blight-resistant trees were kept. Among those, the most American-looking trees were selected, all the rest cut down and killed.

The B3-F2s are being planted into seed orchards at Banshee Reeks Nature Reserve in Loudon County and Sky Meadows State Park in Fauquier County. We hope eventually to plant about 20,000 nuts in our seed orchards and are starting this year with 1,440 in three American backgrounds. Planting should occur within a few weeks given adequate weather. Cathy Mayes has a list of activities in her column that includes plantings at these two sites.

We also will be planting 665 'Nanking' B3s at three orchards, the product of controlled pollinations made last June. These plantings will complete the three orchards, which we have been struggling with due to various factors. To help identify those factors, we will be taking soil samples at the orchards for mineral analysis, measuring tree height and survival, subsoiling at two of them to break up a clay hardpan, and planting potted seedlings at the third so raccoons and voles do not revisit that orchard seeking a tasty and nutritious (!) meal of chestnuts. The potted seedlings will be started soon in the greenhouse.

Prior to June, Cathy Mayes's list has numerous opportunities for culling rejects from orchards inoculated last year, weeding, inventorying, measuring and mowing. In June, we will inoculate three orchards, make final selections and culls in three and do more controlled pollinations.

For controlled pollinations, we have already completed those for our 'Graves' source of blight resistance and will probably this year finish inoculating breeding orchards containing 'Graves' backcrosses. We have completed half of our desired 20 lines for the 'Nanking' source of resistance. A strenuous push this year should come close to finishing breeding of those. Our first 'Nanking' breeding orchard will be inoculated this year, but the others are fairly far behind.

The Virginia Chapter Breeding program is progressing swimmingly, in my opinion. I congratulate you.



Chestnut Research Orchards, Meadowview Research Farms, VA

# 2018 Spring Calendar

**April 6-7** Mid Atlantic Garden Faire, SW Virginia Higher Ed Center, Abingdon. Members of the Southwest Virginia Restoration Branch will staff an information table to answer questions, hand out literature, and recruit volunteers.

**April 21** Abingdon Arbor Day/Earth Day celebration. Southwest Virginia Restoration Branch will staff an information table to answer questions, hand out literature, and recruit volunteers.

**April 21** Orchard work day at Rockley Foundation Orchard in The Plains (Fauquier County): In-fill planting, measuring, mulching.

**Apr. 27** Arbor Day celebration at the State Arboretum of Virginia/Blandy Experimental Farm. Cathy Mayes will be on a panel discussing the remarkable trees of Blandy, which of course include several species of chestnut and one of our breeding orchards. The program will begin at noon. It is open to the public, free to FOSA members, but registration is required. See blandy.virginia.edu.

**May 19-20** Trail Days, Damascus; Talk: Chestnut Story and Finding Surviving American Chestnuts



### Virginia Chapter Display Boards Status

Many of you have probably seen one of the Virginia Chapter's display boards. Perhaps this was the first time you became aware of The American Chestnut Foundation. The Virginia Chapter has two boards. One is currently kept in our Marshall, VA office and one is kept in Charlottesville, VA. Unfortunately, our two boards are out of date. For this reason your Board of Directors has authorized four more display boards with updated pictures and captions. Two will be kept in our Marshall office, two in Charlottesville, one near Blacksburg and one in southwest Virginia. When not needed for public events, they can be loaned out to schools and other organizations. For that reason, these new boards need to be self explanatory.

Ned Yost and I are currently selecting pictures and captions for the new boards. Hopefully, they will prompt interest and questions about our organization and our program. Our efforts have been slowed by a lack of high quality pictures. We have many email quality pictures but they do not look good when enlarged to 8X10 pictures (see the last page of this issue for a call for photos).

The boards will be divided into three topics: history, our breeding program and future benefits of restoration.

If enough high quality pictures can be found, we expect to have these boards ready in the April/May time frame.

If you would like to help with this project, please contact me at <u>wlawsii@aol.com</u>.

Warren Laws VP Education

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# Chainsaw Safety Training with Matt Brinckman

By Tom Saielli, Mid-Atlantic Regional Science Coordinator

Over the years, The American Chestnut Foundation has grown from a small and somewhat relaxed organization, into a relatively large and more professional one. While we appreciate having a bigger role in the world of conservation organizations, we must also embrace the responsibilities that come with it: responsibilities such as safe working conditions, adequate training, and taking necessary precautions to ensure the health and safety of our members and volunteers.

In an effort to stay ahead of the game, the Virginia Chapter has recently adopted a Safe Practices Manual that serves as an aid for leaders to prepare and manage work events, minimizing the occurrence of



accidents and chapter liability. Bucket trucks, ladders, and chainsaws, which are often used by staff and volunteers, require special attention and training before operation. Enter Matt Brinckman.

Most members and volunteers will remember that Matt was the Mid-Atlantic Regional Science Coordinator from 2012 to 2016, and many may be aware that Matt has a background in forestry. He's also skilled in chainsaw training! So, when the Virginia Chapter decided to train its volunteers on basic chainsaw safety, we were thrilled that Matt volunteered to lead the training.

On January 20, Matt and I, along with eight Virginia Chapter members, met at the Stony Brook orchard in Virginia for the safety course. We began at the Stony Brook barn; it was a sunny and warm day for mid-January. Matt began with a review of chainsaw maintenance and the required personal protective gear, accessory tools and equipment, and first aid kits which should always be accessible when operating a chainsaw. Matt then demonstrated the safest and most efficient ways to cut down and buck small trees. Following the presentation and demonstration, the group made its way to the Stony Brook orchard to practice one of our newly acquired skills—culling.

At the end of the day, about 140 trees had been culled, volunteers were satisfied, and Matt (after some bribing) gave everyone a passing grade! These hard-working Virginia Chapter members are now ready to get out and cull more orchards. And with these new skillsets, we can ensure that our chainsaw-wielding members can cull orchards in the safest and most efficient manner possible.

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BxF2 families are planted 150 per family, per plot, in seed orchards throughout the state chapters and Meadowview Research Farm

determine the breeding value of a parent tree when evaluation cannot be accurately determined from phenotypic evaluation alone. This is basically a tool used for selecting various desired traits in a parent (e.g., a tree in a seed orchard) by observing traits in the progeny, such as blight resistance, Phytophthora resistance, timber form, Americanness, etc.

Progeny are established in fields, planted in incomplete blocks and tested for resistance after three to four years. Sometimes progeny are established in forest settings, which allow us to study progeny over several decades in a natural chestnut environment. Over the last ten years TACF has established field and forest progeny tests throughout the chestnut range. Working with staff, chapter members and various cooperators, more than 600 Meadowview seed orchard trees have been established in approximately fifty progeny tests.

Progeny testing can be a highly accurate way to evaluate the blight resistance of the BxF2 trees in our seed orchards, but it is also expensive, labor intensive, and much too slow (currently 5,000 trees remain at our Meadowview seed orchards; we aim to select 500 trees—we've progeny tested about 600 trees over the last 10 years).

In an effort to speed things up and

increase throughput, TACF has been experimenting with small stem assays (SSA), a technique developed by Bill Powell and his team at SUNY-ESF. The protocol has been shown to distinguish highly resistant transgenic seedlings from susceptible, nontransgenic seedlings, but TACF seeks to determine whether the SSA is sufficiently accurate to detect the difference among somewhat resistant and non-resistant hybrid trees.

Why progeny test with small stem assays? We can obtain results with one year in greenhouse vs. three years in field, we can increase sample sizes within families beyond what is feasible in the field and in the longrun, it is easier and less expensive to progeny test using SSA's.

Additionally, TACF is collaborating with Jason Holiday at Virginia Tech to sequence DNA from most of the remaining trees at the Meadowview seed orchards. By comparing genetic sequencing data to the results of the progeny tests data (a subset of



Hill Craddock and Jared Westbrook assess a tree for blight resistance at the Wagner Farm

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## From Matt Brinckman:

"Thanks for having me out for a fun day of chainsaws and chestnuts. Below are links to the VA SHARP Logger Program online trainings I helped put together at Virginia Tech. If you are going to watch both, I suggest watching them in the order below. Let me know if you have any questions or need any assistance in the future."

Basic Chainsaw Safety and Directional Felling: <u>http://sharplogger.vt.edu/online/</u><u>Basic%20Chainsaw%20Safety%20-%</u>202017/index.html

Limbing and Topping Safety: <u>http://</u> <u>sharplogger.vt.edu/online/Limbing%</u> <u>20and%20Topping%20-%20closed%</u> <u>20captioning/index.html</u>



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the Meadowview seed orchard trees have been progeny tested to date) we will be able to develop genomic prediction models for evaluating disease resistance and other traits. This method may prove faster and more reliable and can be performed even when parent trees are not flowering yet (an advantage over progeny testing). The downside is the cost. At approximately \$50 per sample, genotyping must be limited to the small fraction of trees that remain after years of phenotyping and progeny testing. However, once the remaining seed orchard population size is within reason, even chapters will be able to take advantage of genomic selections to aid in making the final selections in their seed orchards.



Tom Saielli and Georgia Chapter volunteers putting in a progeny test in the Chattahoochee National Forest

# Do You Need to Update Your PDF Reader?

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#### **Photos Wanted**

We are always on the lookout for good quality photos for *The Bur*, display boards and the web site. If you have the opportunity to make photos, we'd love to see them. Close-up photos of American chestnut leaves, burs and seeds are always useful. Also needed are photos of volunteers working in orchards and at chestnut-related events.

Help us tell the exciting story of our work to bring the mighty chestnut back to Virginia's forests.

The best photos are high resolution photos that can be blown up to a large size when needed for displays. Jpeg photos are fine. Please send your photos to Norm Reid at <u>jnreid@hughes.net</u>. Be sure to include who, what, when and where information.



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