SPRING 2019 ~ ISSUE 2 VOL. 33

Chestnut

THE JOURNAL OF THE AMERICAN CHESTNUT FOUNDATION

A BENEFIT TO MEMBERS

THE AMERICAN CHESTNUT FOUNDATION®

Transgenic Pollen ~ Meadowview Farm Seed Distribution ~ American Chestnut in Visual Art ~ Seed Stratification

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Lisa Thomson

President and CEO

DEAR CHESTNUT ENTHUSIASTS,

In our effort to share "chestnut stories" across our constituency, it always surprises me how many unique experiences are still to be discovered. These stories, often passed down through generations, have ensured that the hope for restoration of the American chestnut endures.

Hope is alive because of the steadfast support of donors and members like you. TACF's spring appeal, often focused on Meadowview farm operations and capital equipment, has a different theme this year. We are highlighting the breadth and reach of our four regional science coordinators (RSCs) and how they support not only the chapters, but also the overall mission of TACF. In the event you have not had the pleasure of getting to know them individually, allow me to share a bit about each of them. Sara Fern Fitzsimmons, director of restoration and RSC for the North Central region, is a 15-year veteran of TACF, after she began her chestnut tenure as a Duke University Stanback intern, and has been based at Penn State since 2003. Kendra Collins has been our New England RSC since 2008. Just before working with TACF, she finished up her graduate work at the University of Vermont (UVM), researching the cold tolerance of American chestnut. Tom Saielli followed up on Kendra's cold tolerance work at UVM, and subsequently became the Southern Region RSC in 2012. Four years later, Tom relocated to Charlottesville after the departure of Matt Brinckman, opening the door for our newest RSC, Ben Jarrett, who was an intern with the Virginia Chapter in 2016, then worked in the private sector for two years. The call of the American chestnut pulled Ben back into service as the Southern Region RSC last July, joining the team in Asheville. As you can see, internships are an excellent investment with which to grow future chestnut leaders!

These dedicated and hardworking scientists do not only conduct research and experiments, but also work with partners, give countless presentations and attend outreach events, and work side-by-side in the field with the majority of our labor force: our volunteers. Their work is mission-critical and takes tremendous amounts of energy, enthusiasm and careful time management.

Meanwhile, please accept my thanks for your readership. In my travels, I hear such positive things about our award-winning *Chestnut*, which I am delighted to report is read, and enjoyed, in its entirety by so many of our members. If you have thoughts or comments, or would like to share your own chestnut story, please let us know. We would love to hear from you!

With gratitude,

Lisa Thomson

Lisa Thomson, President and CEO
The American Chestnut Foundation

Follow me on Twitter (@MadameChestnut).

Wild American Chestnut

A wild American chestnut in western North Carolina towers over the surrounding foliage and shows off its blooming catkins.

Photo by Paul Noah, 2018 Duke University Stanback intern.

THE AMERICAN CHESTNUT FOUNDATION®

WHAT WE DO

The mission of The American Chestnut Foundation is to return the iconic American chestnut to its native range.

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Dr. Brian McCarthy

Chairman, Board of Directors

As we take a moment and reflect upon the previous year and the new year ahead, I first and foremost want to thank you for being a part of one of the most ambitious and extraordinary acts of humankind – the proposed restoration of the American chestnut tree. Thank you for all that you do, because without your dedication and support, we would not be as close as we are to re-establishing such a special and beneficial tree to our eastern forests.

During the 2018 annual fall meeting in Huntsville, Alabama, I was honored to be elected as the new board chair of The American Chestnut Foundation (TACF). I'm honored to not only lead a wonderful organization, but honored to continue on the solid foundation established by previous chairs Michael Doochin, Kim Steiner, Glen Rea, Dick Will, and many other great colleagues and volunteer leaders of TACF, many of whom have become lifelong friends.

Despite my love of chestnut and this organization, I do have a day job. I serve as the Associate Dean of Faculty, Research, and Graduate Studies for the College of Arts & Sciences and Professor of Forest Ecology in the Department of Environmental and Plant Biology at Ohio University (say that 10 times fast). I became involved with TACF and its mission largely by happenstance some 20 years ago. As a new Assistant Professor – interested in all things related to hardwood trees and their ecology – I was approached by a then graduate student (now board member) Carolyn Keiffer and asked what I knew about American chestnut. At the time, I knew only the basic story of its demise but her curiosity piqued my interest. Several years later I attended my first TACF annual meeting (likely at the Atwood Lake Resort and Conference Center in Dellroy, Ohio) and was hooked. From there I applied for, and received, one of TACF's External Research Grants to do ecological monitoring of the West Salem chestnut stand (with Carolyn), traveled to Meadowview to learn more about the breeding program and the rest, as they say, is history.

The story of how I became interested and involved with TACF is neither the exception nor the rule and that is what has kept me involved for so many years. Everyone who wants to be a part of the reintroduction of American chestnut is welcomed with open arms into the TACF family. Scientists, volunteers, farmers, business owners, entrepreneurs, doctors, lawyers, students, writers, painters, construction workers... people from all walks of life can work collectively and enthusiastically toward a common goal...to save the American chestnut tree!

The Future of American Chestnut

DEPENDS ON FUTURE GENERATIONS

By Nancy Pryor, Carolinas Chapter

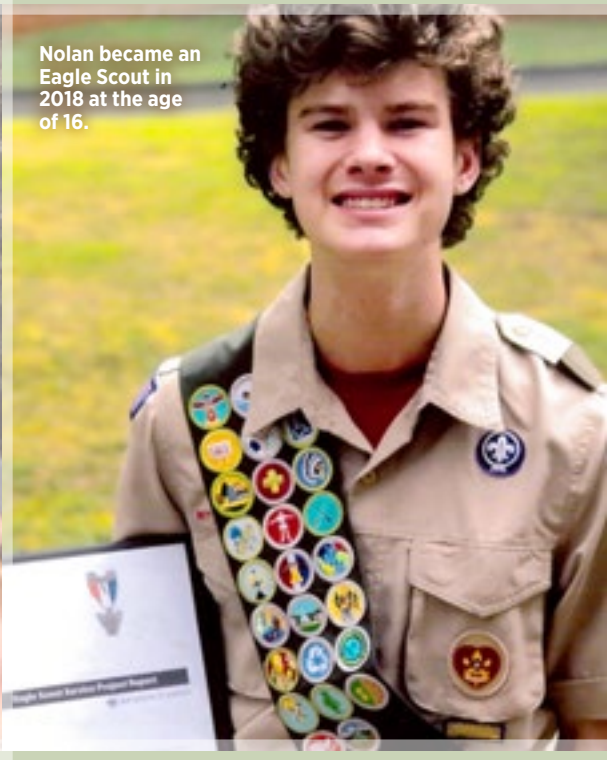
Imagine our forests restored with the tall and mighty American chestnut tree. What a woodland wonder that would be!

Thanks to The American Chestnut Foundation's (TACF) generous donors, determined scientists, and dedicated members and volunteers, the tree's restoration is within our reach. TACF's scientific research and diversified approaches are producing remarkable results.

We celebrate their success, but at the same time, we are cognizant of future challenges. How will this productive path to progress be continued and maintained? Who will take up the mantle?

Future generations will be invaluable in tackling this pressing issue. As efforts toward sustaining and improving our precious forests are crucial for mission success, we must work to ensure that there will be no shortage of young men and women willing to commit to this cause.

(l-r) Nolan McGinley, Scott Pryor (Poppy), Kathleen McGinley, Tina & Gerry McGinley, and Maggie McGinley pose for a family photo in Chattahoochee Forest, late January 2019.



Nolan became an Eagle Scout in 2018 at the age of 16.

As a small child, Nolan plants chestnut seed at the Pryor Farm orchard in 2005 with the help of K.O. Summerville, Carolinas Chapter. Nolan's dad Gerry observes the planting.

Dr. Paul Sisco pollinates a tree at the Pryor Farm orchard in 2018.

There are already examples of this environmental awakening. My family began participating in American chestnut restoration when Dr. Paul Sisco, former regional science coordinator at TACF, selected a site on our property, located at the foot of Sugar Loaf Mountain in Henderson County, NC. As this property had been handed down for generations, our immediate and extended family was eager to help. Appreciation for preservation and conservation has been ingrained in our upbringing.

At the first orchard planting on the farm, one of our grandsons, Nolan McGinley, quickly became an extraordinarily conscientious worker. At the time, he was just four and a half years old, but his young age did not deter him from the task at hand. He worked for hours alongside a very patient K.O. Summerville of the Carolinas Chapter. K.O. helped Nolan perform his job accurately and precisely. He instructed that the correct position of the chestnuts would affect the chances of them sprouting optimally. It was very tedious and time-consuming work for the young boy and his mentor. Nolan's parents, Tina and Gerry McGinley, along with his Poppy (Scott Pryor), stayed nearby to encourage and support him. The whole family was amazed and proud of Nolan's efforts.

As the years passed, those same chestnuts produced many healthy trees. Dr. Sisco used the method of selecting the most resilient trees by inoculating each with chestnut blight. The most vulnerable were cut and destroyed, but the most resistant were left to grow. My husband, Scott Pryor, along with other TACF members, made sure the trees were given special care. Dr. Sisco continues to work and supervise volunteers at the Pryor Farm orchard.

Nolan's experience at the planting became a catalyst for his steady participation in numerous conservation endeavors. His first attempt at sharing what he learned from his involvement with TACF came when he received an assignment in kindergarten. He chose to prepare and present a step-by-step informational chart that aligned the process of how a chestnut grows. He earned a school-wide award for his efforts. This gave him additional incentive to pursue environmental issues.

More recently, Nolan assisted members of his Boy Scout Troop with their projects, along with executing his own. Working together, they are making a difference.

In the spring of 2018, he completed the Eagle Scout requirements. He has acquired an acute awareness of the impact our individual attitudes and actions have on Mother Nature.

Family encouragement can be a powerful key to unlock the interest necessary for our youth to implement the ambitious goals set forth by TACF.

Collectively, the mission before us is to cultivate behavior that will result in meaningful and active participation. The generations beyond us will be a valuable force in the transformation and health of our forests.

Transgenic Pollen:

CREATING THE NEXT GENERATION OF BLIGHT-TOLERANT TREES

By Vernon Coffey, Research Support Specialist, State University of New York College of Environmental Science and Forestry

In June of 2018, the New York Chapter of The American Chestnut Foundation (NY-TACF) and the State University of New York College of Environmental Science and Forestry (SUNY-ESF) hosted a chestnut pollination workshop. The team of researchers who developed the 'Darling 58' transgenic blight-tolerant American chestnut tree worked with workshop participants to pollinate orchard trees with transgenic pollen. About half of the seeds are expected to inherit the blight tolerance transgene. Workshop participants plan to use the skills they gained to pollinate their own chestnut trees with transgenic pollen as soon as it becomes available.

Transgenic American chestnuts flowering in a high-light growth chamber. Chestnuts can produce pollen in less than a year using this technique, and pollen can be frozen for later use. Photo by Vernon Coffey.

Darling 58

The 'Darling 58' American chestnut, developed at SUNY-ESF in Syracuse, NY, has a gene borrowed from wheat, Oxalate Oxidase (OxO), which allows the tree to defend itself against damage from the blight fungus. The fungus is still able to live on the bark, but causes only superficial cankers on stems. (To learn more about the Darling 58, visit this page on SUNY-ESF's website: https://www.esf.edu/chestnut/)

The Darling 58 trees, along with their offspring, can currently be planted only in a handful of permitted research plots. Federal regulatory petitions are being prepared to allow the trees to be released to the public, and if all goes well, approval may be granted within the next few years. Pending approval, SUNY-ESF researchers want to get these trees in the hands of TACF members and state chapters as soon as possible so that they can be used in educational and horticultural plantings, and eventually used to restore American chestnut to its native habitat. Because only a limited number of seeds will be available in the first few years, researchers also plan to provide frozen pollen and scions for grafting. Recipients can then pollinate blight-susceptible mother trees and produce their own blight-tolerant seedlings.

Controlled Pollination

Researchers at SUNY-ESF offered the pollination workshop so that chestnut enthusiasts could learn the skills needed to perform their own controlled pollinations. The basic principles are the same ones that have been used to breed chestnuts for centuries. One difference is that the pollen was produced on transgenic trees growing indoors in high-light growth chambers. The intense light speeds up the production of male (pollen producing) flowers (see Baier K, Maynard C, Powell W. 2012. "Early flowering in chestnut species induced under high dose light in growth chambers." *The Journal of The American Chestnut Foundation*. XXVI no. 3:8–10). No Darling 58 trees are yet old enough to make female (seed bearing) flowers, so all the crosses were made using transgenic pollen on blight-susceptible mother trees.

To make a controlled cross, the female flowers are covered with a bag to exclude unwanted wind-borne pollen, and removed only briefly to place a small amount of pollen directly onto the female flower. The bag is left in place throughout the growing season to protect and keep track of the pollinated flowers, and a wire mesh bag is added later in the summer to deter squirrels from pilfering the seeds. The flowers develop into the familiar spiny burs containing (typically) three seeds per bur.

Over 1,000 nuts were harvested from transgenic pollinations in 2018, including those pollinated during the workshop, more than in all previous years combined. Approximately half of successfully pollinated nuts will inherit the OxO gene and have the blight tolerance of the transgenic parent.

Most of the pollinated mother trees were pure American chestnuts, but there were a few other species as well. A successful cross was made with an Allegheny chinquapin. This hybrid could be incorporated into a backcross breeding program with pure Allegheny chinquapins, retaining the OxO gene for blight tolerance while selecting for the chinquapin phenotype. Another cross was made with a European chestnut. The offspring

Workshop participant Molly Soucy places a pollination bag with workshop leaders Hannah Pilkey (left) and Vernon Coffey (far right). Photo by Linda McGuigan.

Workshop participant Dale Travis uses a glass microscope slide to apply pollen to a female chestnut flower. Photo by Nathan Liang.

A transgenic scion grafted onto a blight-susceptible tree in an orchard begins to sprout. Grafted scions can produce pollen in the first or second year after grafting, which can then be used to pollinate the female flowers on the same tree (or any other chestnut tree). Photo by Vernon Coffey.

About two weeks before the workshop, SUNY-ESF Grad Student Tyler Desmarais climbs a ladder to place pollination bags. Immature female flowers, like the ones shown in the foreground, are bagged to exclude wind borne pollen until they are ready to be hand-pollinated. Photo by Vernon Coffey.

could be used in further breeding to develop a blight-tolerant hybrid tree for agricultural nut production.

Some of the harvested seeds were tested to compare the levels of nutrients such as fats, carbohydrates, protein, vitamins, and tannins between transgenic and non-transgenic nuts (no significant differences were detected). Other seeds were planted indoors and placed directly into high-light growth chambers to accelerate flowering, producing pollen as quickly as possible for the next generation of outcrossing. The rest will be planted in permitted orchards where they will be used to produce more blight-tolerant seeds once they reach maturity, or distributed as seedlings to TACF chapters and members, once approved by federal regulators.

POLLINATION AND GROWING RESOURCES

Chestnut Pollinator's Guide

By Philip Rutter, founding president of TACF
Published in 1990 by Badgersett Research Corporation, available online:
badgersett.com/sites/default/files/info/publications/Bulletin1v1_0.pdf

Chestnut Growers – Breeding and Growing

An online guide from Penn State, which includes sections on pollinations, grafting, biocontrol (mudpacking), and other topics:
ecosystems.psu.edu/research/chestnut/breeding

Breeding for Diversity

The Darling 58 American chestnut and its offspring need to be outcrossed with hundreds of trees throughout the chestnut's range in order to produce a diverse restoration population (see "A Plan to Diversify a Transgenic Blight Tolerant American Chestnut Population" by Jared Westbrook, *Chestnut, The Journal of The American Chestnut Foundation*, Fall 2018). Genetic diversity allows a species to be resilient and adaptable, able to survive future challenges. Although billions of American chestnut trees were killed by chestnut blight, millions have survived, preserving much of the genetic diversity that existed before the blight's introduction.

Many state chapters have set up germplasm conservation orchards (GCOs) containing pure American chestnut seedlings (see "Conservation of Wild American Chestnut Germplasm Through Germplasm Conservation Orchards" by Sara Fitzsimmons, *Chestnut, The Journal of The American Chestnut Foundation*, Spring 2017). If a chapter wants to participate, these could be used for transgenic pollinations to enhance range-wide genetic diversity. Additionally, many citizen scientists have planted pure American chestnuts on private land. When the Darling 58 chestnut is approved by federal regulators, these citizens will be ready to receive pollen and perform controlled pollinations on their own mother trees. You can participate in this effort by working with TACF to find and propagate wild trees in your area.

Nicolas Orellana measures a hybrid chestnut while Vincent DeMartis records at Greenwich Land Trust's American Chestnut Sanctuary. Photo by Steve Conaway.

American Chestnut Inspires

AND ENGAGES YOUNG CONSERVATIONISTS

By Steve Conaway, Conservation & Outreach Director, Greenwich Land Trust

On a hot July day in 2015, the sun beat down on a group of high schoolers while they meticulously lifted each plastic tree tube and pulled the weeds around the base of young chestnut saplings. "I wish these trees would hurry up and grow," muttered one teen, "then at least we could get some shade." But the lesson that day was there is no instant gratification in growing chestnuts and their hard work would help these trees prosper well into the future. As the years passed, the promise of the chestnut planting began to take shape and the story of restoring a species at the edge extinction resonated more and more with its young caretakers.

Chris Aldrich, Director of Stewardship, leads a group from the local Boys and Girls Club in trenching a drip irrigation system for Greenwich Land Trust's American Chestnut Sanctuary. Photo by Steve Conaway.

Youth Corps teen James Brunetti observes fungal spores. Educational opportunities often relate directly to the conservation projects the teens are working on. Photo by Steve Conaway.

Ben Hein (left) volunteering at the 2014 orchard planting. Feeling personally invested in the Land Trust and chestnut restoration Ben returned as a college intern in 2017. Photo by Misha Conaway.

Greenwich Land Trust's Youth Corps was founded in 2014 with the goal of enhancing stewardship of Greenwich Land Trust nature preserves while offering paid summer work experience to local teenagers, and exposing the next generation of conservationists to environmental careers. That same year, Greenwich Land Trust was working with The American Chestnut Foundation (TACF) to establish the first progeny test orchard in Fairfield County, Connecticut. With over 75 nature preserves dotting the map of Greenwich, the Land Trust tasked the newly formed Youth Corps with finding the perfect site for growing chestnut trees. That first group of seven teenage employees were taught principles of soil science, forestry, and horticulture. They set out to different nature preserves with tape measures and soil probes under the guidance of their summer supervisors. After a season of soil testing and mapping, all the necessary factors aligned on one gently sloping field with space for hundreds of hybrid chestnuts. In November of 2014, bareroot seedlings from TACF's backcross breeding program were planted at the site. Some of the teen employees joined dozens of other community volunteers for the planting event.

Along with the progeny testing, a weed management study was included that utilized randomized treatments of different weed controls. Each summer, a new group of Youth Corps employees measured tree growth and recorded weed conditions, learning the importance of precise data collection and how their observations could contribute to restoration efforts throughout the eastern United States. Over the years, the Youth Corps cared for the trees, hand watering and fertilizing the newly planted saplings, spreading mulch, building rodent protection, and eventually installing 6,000 feet of buried drip irrigation. The importance of their work was always reinforced by the story of the American chestnut's tragic decline and the promise of its hopeful return.

The Youth Corps was built in partnership with the Town of Greenwich's Social Services department, which had run its own youth conservation program for many years. The town's Greenwich Youth Conservation Program offered paid community service opportunities to a younger cohort of teens each summer. Graduates from this program were invited to apply to the Land Trust's Youth Corps to expand on their conservation education. As Youth Corps employees they were given more responsibility and ownership of their work. They relished the opportunity to share their new knowledge about the natural world with their younger peers in the town's program and taught them about the projects they were working on. One

Youth Corps employee especially liked sharing the impactful work with chestnut restoration, commenting "We gave an awesome presentation about the chestnut orchard to the younger group. It showed our knowledge, leadership skills, and was a lot of fun!"

Over time the Youth Corps grew, doubling in size to 14 teenagers employed for the summer. Their work diversified to include trail building, invasive plant control, and tending a vegetable garden that yielded hundreds of pounds of produce for the local food bank. But chestnuts remained a common thread, with the trees literally growing alongside the program. In order to extend further opportunities to Youth Corps graduates, the Land Trust created an internship for college-aged conservationists. Ben Hein, a participant in the inaugural session of the Greenwich Land Trust Youth Corps, returned as an intern in 2017. As a 2014 Youth Corps employee he had pulled soil samples that led to the site selection and volunteered in the fall to plant the hybrid chestnut seedlings. Ben's interest in environmental conservation continued in college, pursuing a degree at Cornell University in Environmental and Sustainability Sciences. As a Land Trust intern he continued to work with the progeny test orchard, supervising data collection and performing analyses. Seeing the progress of the chestnut planting over the years was especially important to him, "I helped plant the trees. I think it would be pretty cool to bring my family there in 10 or even 20 years and show them what I did as teenager and how these trees have grown."

The hard-working teenagers in the Youth Corps have helped properly care for the hybrid chestnut trees entrusted to Greenwich Land Trust by TACF and in return the trees have deeply enriched the program and its participants. The American chestnut serves as a tangible example to inspire these young conservationists on what is at stake and how they can contribute to the conservation and success of our vital resources.

Youth Corps employee Kathy Alvarez leads a group of her younger peers from the town's Youth Conservation Program through the Greenwich Land Trust vegetable garden. She discussed garden plant pathogens, segueing into a demonstration on chestnut blight. Photo by Steve Conaway.

Youth Corps employee Nicolas Orellana (center) quizzes his younger peers after a tour of land trust conservation projects. Photo by Steve Conaway.

Greenwich Land Trust Youth Corps employee Kathy Alvarez leads a group of her younger peers from the Greenwich Youth Conservation Program through the Land Trust greenhouse, explaining different plant pests they encounter. Photo by Steve Conaway.

How One Tree and One Family

MAKE A HUGE DIFFERENCE FOR THE AMERICAN CHESTNUT

By Kathy Patrick, GA Chapter President

Dr. Austin Flint and his lovely wife, Bea, own an 800-acre ranch in Waleska, Georgia, situated about 50 miles north of Atlanta among beautiful rolling hills. About one acre of the ranch is home to the Georgia Chapter of The American Chestnut Foundation's (GA-TACF) seed orchard, the first one planted in the state. The Flint orchard, initially planted in 2017 using B_3F_2 and B_4F_2 seedlings from Georgia, Tennessee, and Kentucky, has roughly 1,600 trees planted on a sunny slope protected by a deer fence that members constructed.

This seed orchard is designed to produce blight-tolerant seeds for restoration in wild locations in Georgia and nearby states. The ranch was the lunch and field tour location for GA-TACF's 2018 Annual Chapter Meeting.

Dr. and Mrs. Flint have a ranch caretaker, Carlos Chex, who lives onsite to help with property maintenance and new projects. The Flints sponsor a student intern, Kylie Moura, who attends Reinhardt University in Waleska. Kylie spends a portion of each week during the non-growing season making sure the fencing and tree cages are in place, while studying plant science in general, as well as chestnut-specific topics. During the growing season she will make sure mowing and weeding is taken care of, handle inoculations and chemical treatments, and help with plantings as needed. Kylie's internship is for one year, which extends past her graduation date, thus highlighting what an enthusiastic intern she is!

The Flints bought the ranch in 2001 as both a second home and a new

Aerial view of the Flint seed orchard. Photo by Dr. Russell Flint.

"The work being done by The American Chestnut Foundation to restore the American chestnut is so wonderful; I see it as an honor to my father who loved the American chestnut, as well a benefit to all mankind." – Dr. Austin Flint, February, 2019

avocation in anticipation of Dr. Flint easing into retirement from his career as a radiologist. They put the land into a permanent conservation easement, and one of their sons also owns the farm adjacent to the Flint property. The Flint ranch house is a delightful mix of Bavarian style crossed with Southern shaded porches that wrap around the home. Dr. Flint says being at the ranch is truly his "happy place" as he loves venturing out on his ATV and driving his dump truck or other pieces of farm equipment he and Carlos have collected over the years to support the improvements they have made to the property. The ranch has numerous pine stands, grazing pastures, beautiful streams (including one with a swinging bridge), a "bear-cam" to remotely watch bears' habits, wildflower plots, American Indian petroglyphs, and a great fishing hole Dr. Flint stocks with trout. But best of all it has a wild American chestnut tree!

Dr. Flint discovered the American chestnut tree not long after they bought the ranch. In 2014, a resident of the area contacted Dr. Martin Cipollini, Berry College professor and GA-TACF science coordinator, about the tree. This is how the relationship between Dr. Flint and the GA-TACF Chapter began. Once Dr. Cipollini saw the tree and the property, he suggested to Dr. Flint that the ranch would be perfect for a chestnut seed orchard. Dr. Flint was thrilled at this suggestion and eager to begin working with American chestnut trees as well as the chapter. The rest, as they say, is history!

Dr. Flint's father grew up in Mt. Airy, GA, a tiny town in the northeast part of the state. According to Dr. Flint, his dad, one of eleven children, saw plenty of American chestnut trees there as a boy in the early 1900's. His father dearly loved the trees and reportedly, in the fall, would "wade through" all the chestnuts on the ground. So, when Dr. Flint found the wild American chestnut on his property, he knew he wanted to honor his father's memory – and love of the tree – by helping to restore the species to the eastern forests. Incidentally, his wild American chestnut tree is still alive and has been incorporated into the chapter breeding program using its pollen and seeds.

Dr. and Mrs. Flint support restoration of the American chestnut through the generous donation of their land as a seed orchard site, funding for a Reinhardt University student intern, and numerous invitations to their ranch for orchard tours. In doing so, they are truly honoring his father's memory, while also playing a huge role themselves toward, as Dr. Flint says, the "benefit of all mankind."

GA Chapter volunteers planting at the Flint seed orchard. (l-r): Mark Stoakes, chapter vice-president; Steve Wallin, chapter member; Kathy Patrick, chapter president; Dr. Martin Cipollini, chapter treasurer; Jack Rogers, chapter secretary; Jonathan Price, chapter member; and John French, past chapter president. Photo by Dr. Martin Cipollini.

GA-TACF Chapter President Kathy Patrick hard at work at the Flint seed orchard! Photo by Dr. Martin Cipollini.

2019 National Volunteer Week

TACF is built from the shoulders of its volunteers. Each year, these volunteers from across our 16 state chapters spend countless hours helping TACF move closer towards its goal of American chestnut restoration. In honor of National Volunteer Week, which took place April 7-13, this page is dedicated to our many committed and passionate volunteers. Thank you for all you do!

"If America's beloved [American chestnut] tree can thrive again in the Appalachian canopy, it is because of those who neither accepted its demise, nor abandoned its story to legend."
~ Matt Collins, journalist

"I am only one, but I am one. I cannot do everything, but I can do something. And I will not let what I cannot do interfere with what I can do."
~ Edward Everett Hale, author

"What we have done for ourselves alone dies with us; what we have done for others and the world remains and is immortal."
~ Albert Pike, writer

"Volunteers do not necessarily have the time; they just have the heart."
~ Elizabeth Andrew, volunteer

"As you grow older, you will discover that you have two hands – one for helping yourself, the other for helping others."
~ Audrey Hepburn, actress








MEADOWVIEW FARM
Seed Distribution

By Judy Antaramian, TACF Membership Coordinator

Every year, early March is a busy time for a particular group of staff members, and a date long-awaited for by our seed level members. On March 5, myself, David Kaufman-Moore, donor relations manager, and Shana Zimnoch, gifts and records specialist, travelled to Meadowview Research Farms in Meadowview, VA, joining with staff there to meet the challenge of packing and shipping more than 4,000 seeds in one business day.

The trip was a lovely two-hour drive through the NC and TN mountains. When we arrived, farm staff had the seeds (harvested in the fall of 2018) packed in baggies, and boxes assembled. We created a makeshift assembly line, dug in, and didn't stop until the more than 550 boxes were packed and ready to be shipped!

These potentially blight-tolerant seeds are delivered to seed level members of The American Chestnut Foundation (TACF), some of whom have been growing seeds from Meadowview Research Farms for many years, while others will be trying their hand at this experience for the first time.

A number of members have informed us that they have received their seeds, planted them in pots, and are eagerly awaiting the day to transfer them into the ground. Participating at this level of membership isn't just about planting and growing trees, it supports TACF's commitment to restore the American chestnut tree.

With the 2019 distribution behind us, we are already working on the list for March 2020. If you're interested in becoming a seed level member, please visit the Seed Level Membership page on our website (acf.org/store/seed-level-membership/) or call our national office at (828) 281-0047.

We are grateful to all of our members for being part of this important and hopeful mission, ensuring that future generations will one day experience forests filled with healthy American chestnut trees.

Carolinas Chapter member Paul Sisco (center) stopped by the farm for a visit and graciously volunteered to help TACF staff pack the boxes. Also pictured (l-r): Judy Antaramian, Shana Zimnoch, and David Kaufman-Moore. Photo by Laura Barth.

Interested in becoming a seed level member?

Visit our Seed Level Membership page at acf.org/store/seed-level-membership/ or call our national office at (828) 281-0047.

Carolyn is dwarfed by the large American chestnut tree behind her, circa 2002. Photo by Dr. Brian McCarthy.

Carolyn Keiffer

TAKING ON THE TOUGH ASSIGNMENTS

By Scott Carlberg, Carolinas Chapter

A random 18-inch American chestnut seedling reached out for sunlight in Ohio in 1988, but it captured a heart instead; Dr. Carolyn Keiffer's heart. She didn't expect to find a botanical soulmate that day.

It was a love-at-first-sight kind of thing, though Carolyn didn't know it at the time. "I was in grad school at Ohio University, and on a field expedition," said Carolyn. The class was in Wayne National Forest, a quarter million acres of Appalachian foothills in southeastern Ohio. The hunt was for wildflowers in the undergrowth.

"Suddenly our leader, a quiet plant taxonomist, dropped to his knees," said Carolyn. "This quiet guy became animated as he looked at this seedling with a half dozen leaves. It was an American chestnut." The guide explained the fight that little tree made to get to sprout at all, much less grow into a mature tree.

That seedling rooted in Carolyn's memory, too. Two years later she had to pick a topic for a plant pathology class. "I chose chestnut blight. I didn't know what I was getting into." That is because Carolyn wrote to various chestnut experts for material. They did not just respond. They answered with enthusiasm, dedication, and something else: "I never met a grumpy chestnut person. They are all positive people."

One of them was Fred Hebard, known for TACF's breeding and resistance program. He urged Carolyn to apply for a TACF research grant to begin a long-term forest ecology study in West Salem, Wisconsin, in 2002. Carolyn's sense of awe grew in La Crosse County, the site of 60 acres of private forestland and the largest remaining stand of American chestnuts. "Pivotal to my understanding of what an American chestnut forest must have looked like," she says. The project continues today.

Carolyn is now a professor of botany in the Department of Biological Sciences at Miami University, Middletown, Ohio. She is a founding member of the OH-TACF Chapter, current chapter president, TACF board member, and volunteer with TACF for more than 20 years.

Trees are Carolyn's tools. Restoration ecology her career, salvaging degraded, damaged, or destroyed ecosystems. Mother Nature's go-to professionals, these people heighten plant diversity and re-establish a biological balance between nature and culture.

Here's an example.

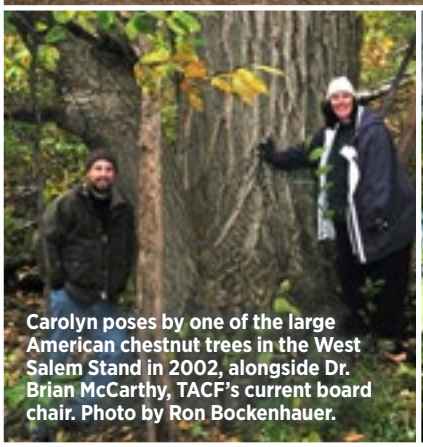
The Fernald Preserve is a former uranium production facility in a rural, residential area northwest of Cincinnati. Production stopped in 1989. "An onsite disposal facility remains at the site containing approximately three million cubic yards of contaminated soil and debris," says the Ohio Environmental Protection Agency (EPA).

"It was my very first solo research project as a new faculty member," said Carolyn. "I wondered if this was a chance to try chestnuts." She did, and the results were good she said, although, "White tail deer like to eat chestnuts." All for a good purpose either way.

Restoration returned natural plant and animal communities to the site. The Fernald Preserve is now a green space park with wetlands, ponds, prairies and upland forest areas.

Carolyn's botanical knowledge rescues strip mining areas, too. Over some eight decades, since the use of huge mechanical shovels, bulldozers and draglines, parts of Ohio had strip mining, or, surface mining, operations. Rock and soil above the coal seam is removed and the coal is taken away. Reclamation shows varied degrees of success.

Carolyn (front, holding hard hat) and a group of students at Miami University pose for a photo at Wayne National Forest near Nelsonville, OH in October 2014. Photo by Gary Chancey.



Carolyn poses by one of the large American chestnut trees in the West Salem Stand in 2002, alongside Dr. Brian McCarthy, TACF's current board chair. Photo by Ron Bockenhauer.

Carolyn, standing beside Dr. Janise Bauman, observes large leaves of an American chestnut tree in the West Salem Stand in 2007. Photo by Dr. Keith Gilland.

"In Ohio there are thousands of acres of old strip mine lands," said Carolyn. "Many never reclaimed or reclamation didn't work. I felt if we got the right plants in the right places, something good would happen."

Heavily compacted soil covered the sites, mostly from heavy mining equipment. Only weedy herbaceous plants grew. "Our earliest sites were just coal and shale. Nothing you or I would call soil, with a low pH, in the 4s. Maybe some lichens and a few stunted yellowish pines," said Carolyn.

Early tests tried small stands of trees, sometimes even bird stands built of two-by-fours, just for places to land and nest. "If you can get a tree to live, others will come in. Mother Nature will help them turn back into a forest." She got a few chestnuts to grow. "They weren't happy, though."

Heavy equipment plowed the rock-hard surface, ripping cross hatch patterns, giving plants places to set roots. Pure American trees and about 1,200 early chestnut hybrids were planted. Also in this project: Dr. Brian McCarthy, now TACF board chair, and Jenise Bauman, a doctoral student of Carolyn's at the time. "I visited this past fall, which is 12 years from the start. Some chestnuts are blighted, but some are 25 feet tall. New woody seedlings are coming in."

As a professor of botany, Carolyn cultivates other kinds of seedlings. The minds of students. "I hear some people say that college students are spoiled and jaded. I find that students love to help, like to get things done."

Jenise is a case in point. She is now an associate professor at Western Washington University in, maybe not surprisingly, restoration ecology. "Carolyn expected hard work and professionalism from her students. As my PhD advisor, she balanced student support and guidance while encouraging my own independent growth as a scientist."

As a professor Carolyn has sent more than two dozen scientists out to jobs as diverse as college professors, US EPA, natural food flavorings, The Nature Conservancy, soil and water conservation service and the Ohio Department of Natural Resources.

Not just scientists, though. "I teach an environmental course for non-science majors. They come into the class uninformed about the environment. What bothers me is when they do not care they are uninformed and say, 'I don't want to know because it is too depressing.' I try to help them understand and care."

Since Carolyn is a botanist, one question stands out: "Carolyn, why do you love the American chestnut instead of another tree species?"

Considering that Carolyn cannot grow the American chestnut where she lives because of the high pH and limestone, her answer is especially interesting. "Chestnuts are amazing trees. They have food productivity that is important for wildlife. The grain is pretty, and the wood has been useful. The tree grows fast compared to other nut-producing trees. Historically, people and livestock have depended on the American chestnut for food."

The American chestnut has supported our nation as it expanded, and, said Carolyn, "When the trees were gone, we really lost something. While its history is a story itself, its tenaciousness is something else. We can go out in the forest and possibly find a chestnut, even from a root system 100 years old. The trees have been doing their best for 100 years and we have the science and the will to save it. And we should."

200 MILLION ACRES OF AMERICAN CHESTNUT NATIVE RANGE

Supporting OUR RSCs

FOUR TACF REGIONAL SCIENCE COORDINATORS




Kendra Collins, Sara Fitzsimmons, Ben Jarrett, and Tom Saielli cover nearly 200 million acres of the American chestnut's native range – that's a lot of land! These RSCs assist the 16 chapter breeding programs, facilitate external partnerships and collaborations, and engage with current and potential members in becoming an integral part of our chestnut restoration efforts. They are our frontline staff science ambassadors.

Through the years, along with the support of funders like The Manton Foundation, David Greenewalt Charitable Trust, Richard King Mellon Foundation, chapters, and other gracious patrons, TACF ensures the work and scope of the RSCs has been able to grow. With your support, we can enhance the innovative work of our current RSCs, not only by providing them with additional supplies, travel funds, and professional development opportunities, but by allowing them to hire and work with talented young interns. These interns lighten the burden during the busy growing season and foster the next generation of scientists who will move us even closer toward American chestnut restoration.

"The four individuals who make up TACF's Regional Science Coordinators (RSC) should probably also be referred to as Responsible Scientific Counselors, because without their support, coaching, and patience, we would all be lost."

– Caitlin McMahon, TACF Member

PART 3
of a
3-part series

The American Chestnut Tree: Not Gone, Not to Be Forgotten

People are introduced to the American chestnut in surprising ways. In this third article, Doug Gillis explores how people learn about the American chestnut through visual art that includes sketches, paintings, and even quilt work. Such arts and crafts can capture one's imagination and reinforce the desire and need to see the tree restored to its eastern forests and woodlands.

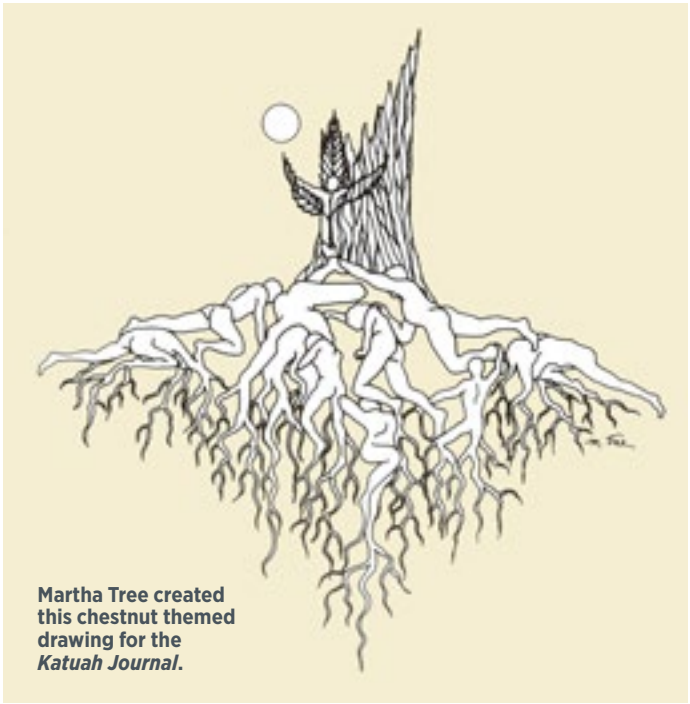
The American Chestnut Foundation and its chapters, through promotion and outreach, expose people to the history and culture of the tree, to the science and technology that will rescue it, and to the pleasures gained by being involved in its restoration. At the end of this article, we invite you to share your knowledge of visual art that uses the American chestnut as a theme.

Learning About American Chestnut

THROUGH VISUAL ART

By Doug Gillis, Carolinas Chapter

An artist or craftsperson producing art related to the American chestnut transforms a conceptual idea into an object to be appreciated for its beauty and emotional power. The object might be a piece of ceramic with a leaf imprint, a drawing or painting telling a story, a bas-relief carved on a chestnut board, or a pattern used in producing a quilt. An observer often adds additional meaning to the artwork based on his or her interpretation.



Martha Tree created this chestnut themed drawing for the *Katuah Journal*.

Martha Tree created the chestnut themed drawing that appeared on page 15 of the *Katuah Journal*, Issue 21, Fall 1988. The drawing conveys great emotion without the use of words. The tree struggles to survive. Humankind intervenes. Roots morph into human forms that push life force into a sprout. The sprout develops leaves being supported by another, smaller figure. That figure, in my interpretation, is a dryad, the spirit that lives in the tree as long as the tree lives. Restoration of the tree is the hope.

Visual artwork can tell a story with minimal use of words. An example is the American Chestnut Botanical Print by Bruce Lyndon Cunningham. The print can be viewed at: https://www.acf.org/store/american-chestnut-botanical-print/. The art depicts the form of the tree; one half fully leafed out and the other half without leaves to illustrate its limb structure. Leaf shapes are shown in spring, summer, and fall colors. Twig and bud characteristics are illustrated. Roots emerge from the seed, leaves sprout from the nut, and a seedling grows to continue the cycle of life. The drawing includes images of female flowers and male catkins, developing chestnut burs, and a ripened bur that opens to spill nuts that sprout later.

Coreysha Stone used her artistic talent to produce numerous acrylic paintings depicting the American chestnut tree through the seasons. The paintings were displayed at the art show, "Bringing Back the Giants," to benefit the ME-TACF Chapter. The paintings, one of which is displayed in this article,

American Chestnut Botanical Print by Bruce Lyndon Cunningham.

An acrylic painting of chestnut burs by Coreysha Stone.

"Chestnut Mountain" skinny quilt by Pat Straka.

include a tree in winter, leaves in spring and fall colors, burs not yet opened, and a cluster of nuts. Coreysha's strong connection between her art and the natural world is evident in her chestnut paintings.

TACF converted a quilt square pattern, "Chestnut Restoration," into a 4" by 5" decal given to new members in 2015. The quilt square pattern is a custom design done by Martin Webster for the Quilt Trails Series of Western North Carolina. This unique design captures the distinctive shape and dentate margin of American chestnut leaves. The alternating colors of the leaves represent the changing seasons. The pentagon brown shapes at the top of the pattern are chestnuts falling and the same brown shapes at the bottom are chestnuts ready to sprout. The brown shaded stripes in the lower half of the pattern are of the earth and its mountains and the blue shaded stripes, the sky. Symbolically, the upward pointing green leaf (on the left) represents the American chestnut as the dominant tree of eastern North America at the beginning of the twentieth century. The downward pointing yellow leaf represents the tree's tragic decline due to chestnut blight. The upward pointing green leaf (on the right) represents the hope of restoration – bringing blight-tolerant chestnut trees back to the native range.

Custom quilt square design by Martin Webster for the Quilt Trails Series of Western North Carolina.

Skinny quilts created by Pat Straka of Westminster, SC boast of rich colors and stunning backgrounds. The first quilt she donated to TACF, appropriately named "Chestnut Leaves," was raffled off at the 2016 Annual Fall Meeting in Louisville, KY. It came with a wall hanger made of American chestnut that was handcrafted by Carolinas Chapter member, K.O. Summerville. Her second quilt, "Chestnut Mountain," was raffled off before the holidays in 2017.

Help TACF catalog examples of visual art that uses the American chestnut as a theme. Share pictures and your interpretation of the art. Contact TACF's *Chestnut* editor Jules Smith at jules.smith@acf.org.

THE IMPORTANCE OF CREATING A DIVERSE POPULATION OF

American Chestnut Trees

By Ben Jarrett, Southern Regional Science Coordinator

The American Chestnut Foundation's (TACF) 3BUR (Breeding, Biotechnology, and Biocontrol United for Restoration) plan paves the way for future research and restoration efforts to create a blight-tolerant and genetically diverse American chestnut population. Both the biotech and backcross breeding approaches rely on breeding in genes for blight tolerance (either the wheat oxalate oxidase gene or Chinese chestnut genes) into a genetically diverse and regionally adapted population of wild American chestnut trees. The restoration population must be diverse enough to adapt to the changing climate and to maintain itself over generations of trees without inbreeding. The 16 state chapters of TACF were mostly founded to create their own backcross breeding programs with local American chestnuts. The backcross breeding programs across chapters, as well as Meadowview Research Farms, represent hundreds of local American chestnuts from across the native range. We are looking to our members and collaborators to help TACF obtain more diversity in our program for use in backcross breeding and breeding with the transgenic chestnut tree developed at State University of New York College of Environmental Science and Forestry (SUNY-ESF).

A few wild American chestnut trees with green burs can be seen in the lower third of this photo taken in Western North Carolina off the Blue Ridge Parkway. Photo by Ben Jarrett.

As our climate warms, the southern edge of the historical range of the American chestnut has begun to shift northward. There are anecdotal reports from Alabama and Georgia that American chestnuts are dying and not re-sprouting like they have done since the blight struck decades ago. The Southeastern United States was a refuge for American chestnut population during the last ice age. Consequently, southern populations have a higher than average levels of genetic diversity. Capturing and conserving this diversity is important for restoring American chestnut in a way that the tree will thrive in a changing climate.

Capturing the genetics of a particular tree or group of trees is called germplasm conservation. The most common way of conserving American chestnut germplasm is collecting nuts from a tree and then planting the trees in a Germplasm Conservation Orchard (GCO). Native trees used in backcross breeding programs are also conserved as their genes live on (albeit partially) in their progeny. New, flowering trees are getting harder to find on the edges of the range, in areas where chestnut was not as common historically and where many trees were lost to *Phytophthora* root rot. Many trees in the southern edge of the range never produce viable nuts. Due to the rarity of nut-producing trees in these regions, TACF and research partners have begun looking at alternative ways of conserving native germplasm across the range, particularly in underrepresented areas. Research projects include the development of habitat suitability models to predict chestnut "hotspots," grafting American chestnuts and subjecting them to high light treatments to accelerate flowering, and other vegetative propagation methods. The effort to conserve germplasm isn't just on an academic level, TACF's citizen scientists play an important role in the process – primarily by helping to locate more trees.

In July 2017, the TreeSnap app for smart phones was launched to help tree researchers connect with citizen scientists. The app allows anyone to report the location and condition of American chestnuts. TreeSnap enables users to take photos of the trees and answer questions about blight severity, tree size, and whether or not the trees are flowering. Cell phones automatically capture the GPS locations of the trees while users are entering this information. When permission has been obtained by the landowner, TACF requests a leaf and twig sample to verify that the tree is an American chestnut. Since the

The scions begin to leaf out within a few weeks of grafting. This American chestnut germplasm was collected in the Alabama wilderness. Photo by Dr. Hill Craddock.

A member of the WV-TACF Chapter documents the location of a wild American chestnut tree using the TreeSnap app.

app's launch in 2018, more than 1,200 American chestnut submissions have been recorded. These submissions are being used by TACF staff and collaborators to collect nuts, pollen, and scion for grafting. Collaborators are also training predictive models to identify climate and soil conditions where American chestnuts are likely to be found.

TreeSnap was also used last summer as a data collection tool by TACF scientists to document the collection of American chestnut leaves for DNA analysis. In fall 2018, a full genome of American chestnut was finished. This breakthrough opens the gate for many new research projects. One of the first has been named The American Chestnut Landscape Genomics Project. In collaboration with Virginia Polytechnic Institute and State University, TACF began collections from all areas of the range to determine how genetic diversity of American chestnut populations are related to geographic locations and climate variation across the species range. In our sampling for DNA analysis, we sought to fully represent the ranges of elevation, precipitation, and mean annual temperature where American chestnut occurs. For example, in North Carolina, collections were made from trees at 800 feet elevation and at 5,500 feet elevation (and at many intervals in between). Once this project is complete, a robust data set will be used to target locations for germplasm conservation. We also plan to combine this information with climate change models. Together, we could assist the migration of genetic material to where it is most likely to succeed. Since the climate is warming, trees adapted to southern climates may need to be moved further north or to higher elevations to be best suited for long-term survival of populations.

Over the last twenty years, the scientific community has produced a lot of breakthroughs in genetics that have allowed TACF to begin answering questions on genetic diversity that have never been answerable until now. With this major effort towards germplasm conservation, our research can continue moving forward to create a population of blight-tolerant trees with enough genetic diversity to withstand the test of time.

Seed stratification

IS NOT REQUIRED FOR GERMINATION, BUT IS IMPORTANT FOR SEED SURVIVAL

By Bruce Levine, Maryland Chapter and Laura Barth, Meadowview Horticulture and Pathology Specialist

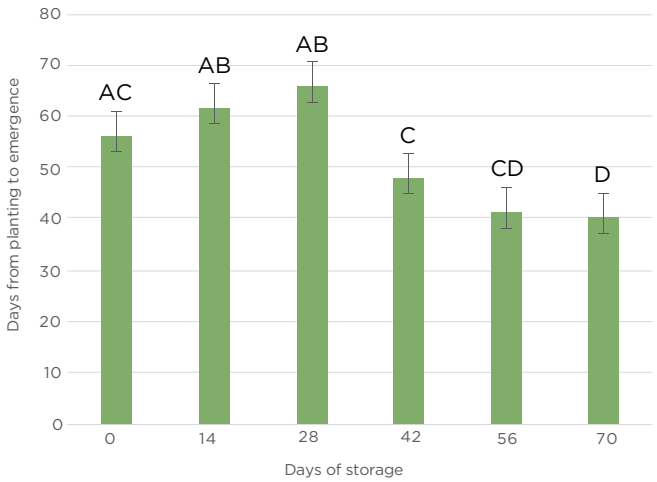
Conventional wisdom says that chestnut seeds must be stratified (stored at a cold temperature) in order to germinate and produce seedlings. But unlike acorns and crop seeds, little has been published regarding chestnut. For those of us who raise chestnut seedlings in greenhouses the information is important. The shorter the stratification period, the earlier the seeds can be planted, allowing greenhouse trees to grow larger in one season. To clarify this question, The American Chestnut Foundation (TACF) has been studying the stratification requirements for chestnuts since 2017 at the University of Maryland and at TACF's Meadowview Research Farms in Meadowview, VA.

Over two years, seed lots of equal size from various American families from different regions of the American chestnut's natural range, and one family of Chinese nuts, have been subjected to varying lengths of cold and warm storage, and then planted in a greenhouse. The time from planting until the chestnut plumule (the leafy sprout) emerged from the soil (planting to emergence, or P to E) was recorded for each seed. Then the various treatment blocks were compared. The process was challenging due to nut rot, and space limits that prevented us from growing enough seeds to compare every combination of species, family line, temperature and time in one attempt. We also lost one line of American seeds when we overheated them when treating for weevils. More work is required, but we did produce some interesting initial findings, which we summarize below.

1. Cold treatment is not necessary for germination or seedling establishment.

The time required from planting to emergence was not significantly different for seeds stored at cold temperatures (0-5°C) than for those stored at room temperature (22-24°C). Looking at Chinese seeds (most Americans died during weevil treatment), we found that seeds in warm storage were slower to emerge on average **(Fig. 1)**, but the difference was not statistically significant (a *t*-test with unequal variance produced P values greater than 0.1 for all storage periods). Even seeds planted immediately after harvest (zero days of storage) germinated and emerged with an average of 55 days. This contrasts with studies of acorns that show that all oak species have an absolute requirement for a stratification.[1,2] The conventional wisdom concerning chestnut may simply be an assumption based on scientific observation of oaks. The data we collected suggest that no period of cold treatment is required for chestnuts to germinate and grow into seedlings, although we need to confirm this with multiple Chinese and American lines.

Figure 2

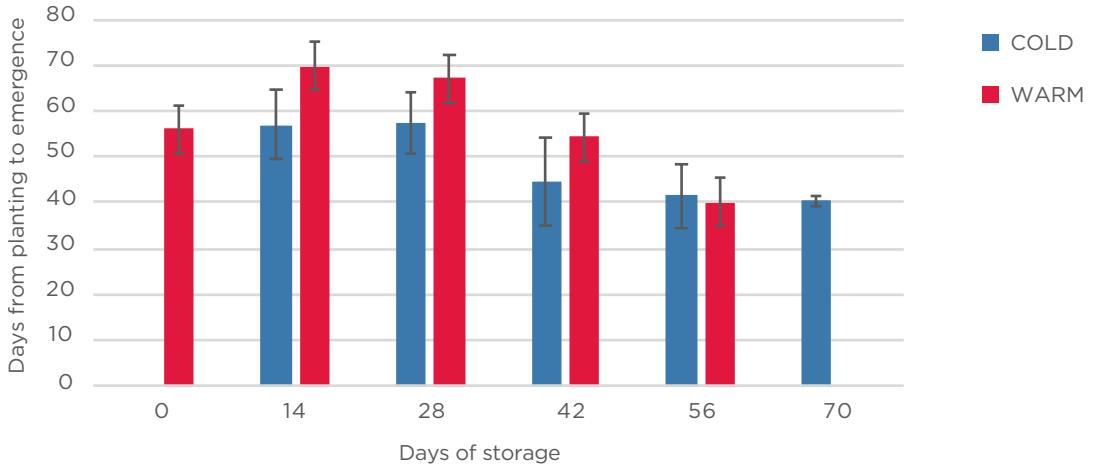


The influence of storage time on P to E time in one family of Chinese seeds, without accounting for storage temperature. Bars with the same letter are not statistically different from each other (*t*-test with unequal variance, α=0.05). The short 0-day bar may reflect the fact that only seedlings that established most quickly could resist the growth of nut rot microbes.

2. Chestnut seeds require a period of maturation averaging about 70 days before emergence, but most of this is not temperature-dependent.

Our data indicate that the maturation period has two phases: 40-60 days of dormancy in storage or after planting, and 10-30 days between planting and emergence for seedling establishment. The length of the dormancy period does not appear to be temperature-dependent **(Figs. 1 and 2)**, though the subsequent seedling establishment period

Figure 1



Influence of storage time and temperature on time from planting to emergence.

Figure 3a

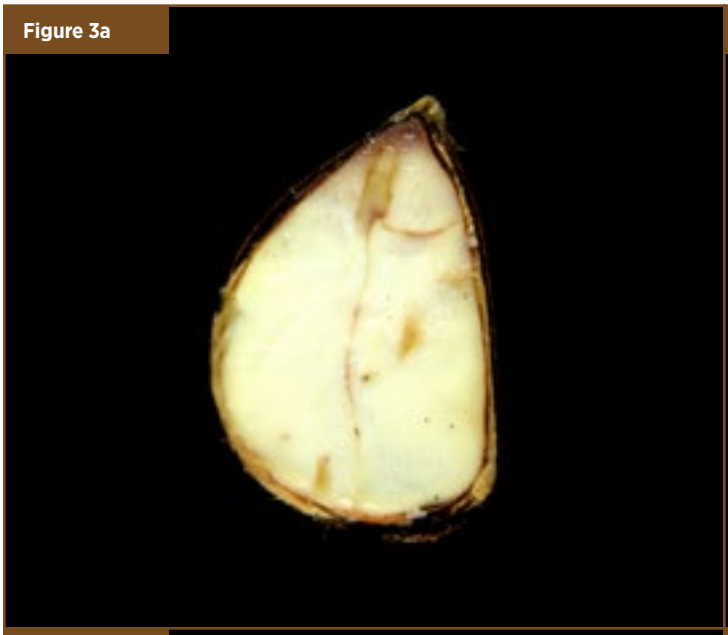


Figure 3b

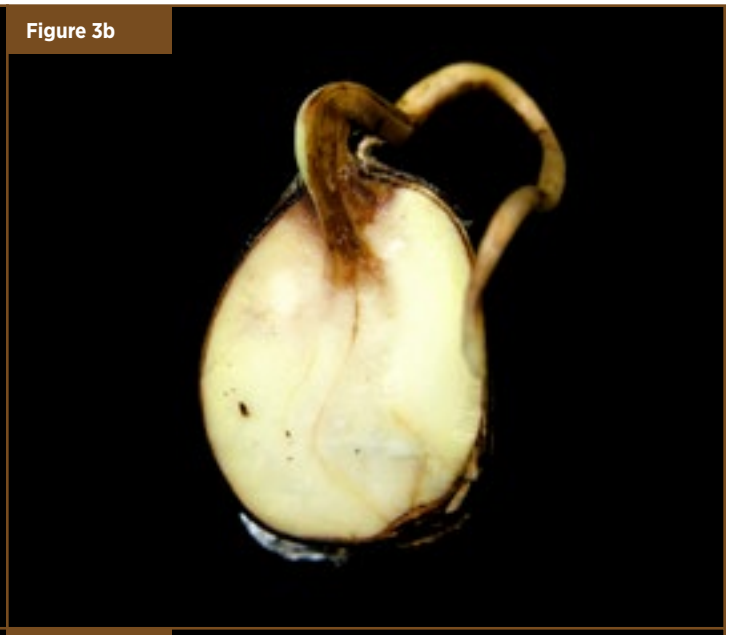


Figure 3c

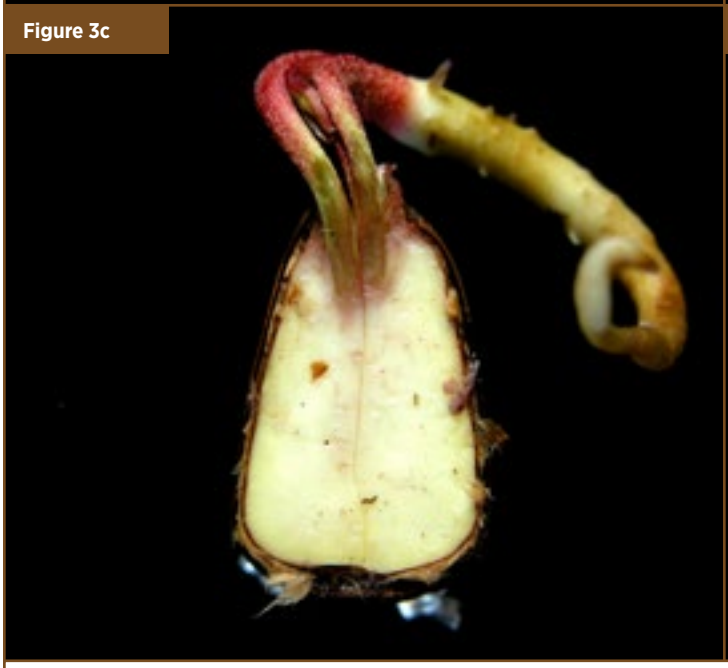


Figure 3d

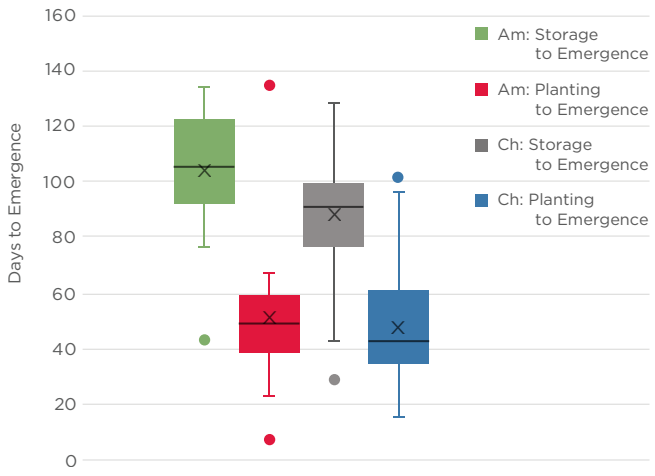
The stages from germination to seedling establishment: a) a chestnut seed at the end of dormancy forming a radical, b) a germinated seed with an elongated radical, c) cotyledons elongate and emerge from the shell, d) the plumule begins to extend from the crotch between the two cotyledonal petioles. Photos by Bruce Levine.

may be. Dormancy periods in seeds allow a rebalancing of phytohormones that regulate the process of maturation, including the mobilization of nutrients for use by the embryo, and the softening of the pericarp to allow a radical to emerge from the nut.[3] Seedling establishment is the period from germination to emergence of the seedling from the soil. Our 2017 study indicated that prolonged cold storage simply suspends the maturation process – giving seeds more than 60 days of cold storage resulted only in a commensurate additional delay in plumule emergence.

3. The presence of radical at time of planting does not imply rapid seedling establishment.

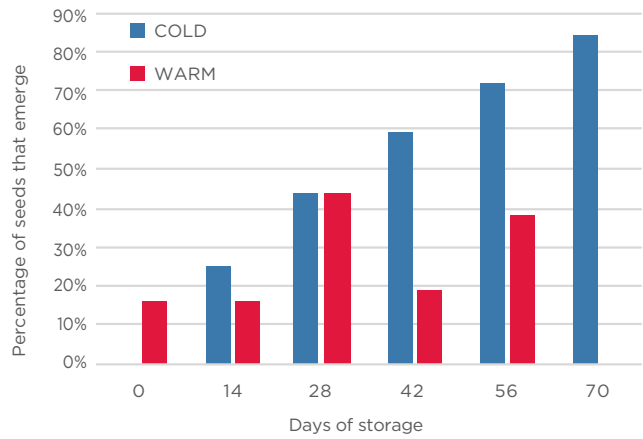
There was no significant difference (*t*-test with unequal variance, P = 0.57) in time to emergence for seeds that did or did not have radicals at the time of planting. However, seeds that had already produced a plumule in storage emerged significantly more rapidly than seeds that had not yet produced a plumule (P < 0.005). Growth of a radical is a prerequisite to plumule production, but our data suggest that the transformation of a germinated

Figure 4



A comparison of storage to emergence times and planting to emergence times for Chinese and American chestnut seeds.

Figure 5



Percentage of seeds from one Chinese family that survived to emergence under different storage periods and temperatures.

seed into a seedling is regulated by a separate process from germination itself. Temperature likely plays a part in triggering this, as we saw more plumules in seeds kept in warm storage. During the seedling establishment period, the radical turns into a proper root, then the cotyledons begin to elongate, leaving their nutrient-rich bulk inside the remnants of the nut, while remaining connected to the root via cotyledonal petioles. The first apical meristem develops in the crotch where the cotyledonal petioles attach to the root. It is this meristem that forms the plumule that ultimately pushes through the soil. **(Fig. 3)** Data collected in 2017 suggested a seedling establishment period of 10-20 days, while data collected in 2018 on seeds from the same mother tree suggested about 40 days. The difference was most likely due to temperature – the 2017 nuts were grown in a slightly warmer greenhouse. Light may also be a factor, but not an essential one – even seeds stored in complete darkness will eventually produce plumules.

4. The maturation period for Chinese seeds appears to be shorter than for Americans

We compared 139 Chinese seeds and 21 American seeds (survivors of the weevil treatment), grown under identical conditions in 2018 **(Fig. 4)**, and saw no significant difference in time from P to E (*t*-test with unequal variance, P=0.363), but a very significant difference in time from S to E (P=0.001). The mean P to E period for Americans was only two days longer than for Chinese, while the mean S to E period for Americans was 16 days longer than for Chinese. This suggests that the dormancy period for Chinese seeds is shorter, though it merits further study with more family lines.

The natural range of American chestnut extends into colder regions than the native range of Chinese chestnut, and we must take care when making hybrid lines not to breed cold tolerance out while breeding blight resistance in. A previous study of chestnut seed cold-hardiness found both that Chinese seeds are less cold tolerant than American seeds, and that cold tolerance of the seeds appears to correlate with cold-hardiness of the resulting seedling.[4] Dormancy periods may be an additional climatic adaptation trait we have to monitor.

5. Cold storage greatly improves seed survival.

While seeds may mature during dormancy in warm storage, they are much more susceptible to mold and nut rot. For both Chinese and American seeds, longer periods of cold storage also correlated with greater survival, though for reasons that are not clear from our study **(Fig. 5)**. Cold storage can also prolong the state of dormancy, and help give growers more control over the planting schedule.

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Availability of blight-tolerant American chestnut trees

By Jared Westbrook, Director of Science, The American Chestnut Foundation

A LIMITED QUANTITY OF SEED WITH IMPROVED BLIGHT TOLERANCE IS AVAILABLE NOW

One of the most frequently asked questions TACF staff get when people call the national office is, "When will blight-tolerant American chestnut trees be available?" Other frequently asked questions are "How blight-tolerant are the backcross trees?" and "How 'American' are the backcross trees?" With results from recent genomics research, we are now able to answer these questions more clearly.

In our two seed orchards at Meadowview Research Farms, we generate between 25,000 and 100,000 seed per year through open-pollination among the trees in these orchards. Many of the seeds from these orchards come from parents that we have not yet thoroughly vetted for blight tolerance. A limited quantity of seed from select parent trees with elevated blight tolerance is available now as part of the member seed distribution program. The seed that we are distributing to members this year comes from mother trees that meet the following selection criteria:

❶ The mother trees' main stems have survived blight infection for at least eight years and have stem traits indicative of blight tolerance.

❷ Blight cankers on inoculated progeny of the selected mother trees are less severe than the current population average.

Of nearly 60,000 trees planted in Meadowview seed orchards since 2002, less than 1,000 trees have flowered and produced progeny for evaluation of blight tolerance. This year for the first time, we have used genomics to predict average progeny canker severity and the long-term blight tolerance of parent trees to make rapid progress in selection in seed orchards. As we continue to cull susceptible trees, we expect that average blight tolerance of the seed coming from these orchards will increase. We expect to complete selection of the 1% most blight-tolerant trees in one of the Meadowview seed orchards by the end of 2020. We anticipate completing selection in the second orchard in 2021 or 2022.

Thirteen of 16 of TACF's chapter breeding programs are also in the process of planting and selection in backcross seed orchards. These seed orchards are composed of backcross progeny from wild American chestnuts in each chapter's region. Thus, they represent genetic diversity and adaptive capacity remaining in extant *Castanea dentata* populations. Many chapters have just started planting their seed orchards or have just performed their first inoculations with the blight fungus. Seed production from parents carefully selected for blight tolerance is five years to a decade away for many of these orchards. However, a few chapters including the Carolinas and PA/NJ have made progress in selection and are evaluating progeny from selected parents for blight tolerance in field and greenhouse trials.

How blight-tolerant are the backcross selections?

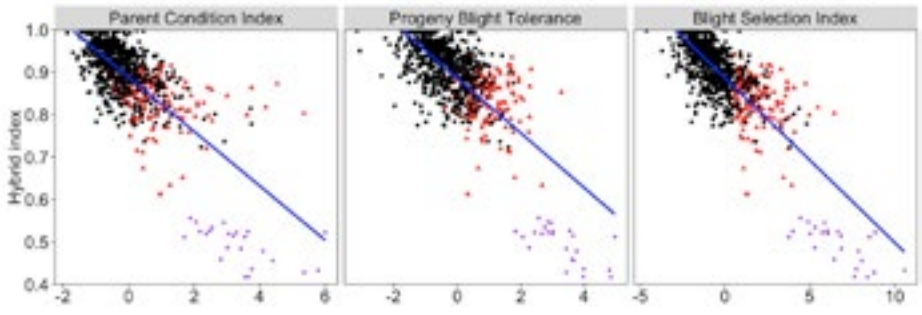
When we inoculate and evaluate our most advanced generation (BC_3F_3) American chestnut backcross trees' blight tolerance, we include blight-tolerant Chinese chestnuts, susceptible American chestnuts, and 50/50 hybrids of Chinese and American chestnut in our trials for comparison. On average, we find that BC_3F_3 progeny and the selected BC_3F_2 mother trees have blight tolerance that is between that of American chestnut and 50/50 hybrids of Chinese and American chestnut. **Figure 1** shows

Figure 1

Comparisons of the stems of pure American chestnut (A), a BC_3-F_2 selection (B), and an F_1 (C)

the stems of pure American chestnuts as compared with selected BC_3F_2 mothers and 50/50 Chinese/American hybrids. The pure American chestnut stems die and resprout repeatedly after blight infection. The "best of the best" BC_3F_2 selections often have extensive cankers on the main stem, but the cankers are superficial and the stems survive. The 50/50 Chinese/American hybrids often have cankers that are smaller and more superficial than on the BC_3F_2 selections. After we complete selection of the most blight-tolerant trees in seed orchards, we plan to plant progeny from these orchards in forest settings to determine if this level of blight tolerance is sufficient for restoration.

Figure 2



Proportion of BC_3F_2 genomes inherited from *C. dentata* (hybrid index) versus blight tolerance. Blight tolerance was assessed via the Parent Condition Index (a sum of five late-developing blight trait on BC_3F_2 stems), Progeny Blight Tolerance (average progeny canker severity, reversed in scale), and Blight Selection Index (Parent Condition Index + Progeny Blight Tolerance). Red triangles are BC_3F_2 selections, purple diamonds are the Chinese chestnut x American chestnut hybrids and black dots are inferior trees to cull. Blue lines are the regressions between hybrid index and blight tolerance traits. From Westbrook et al. (2019).

How 'American' are the backcross selections?

With genomic testing, we have recently estimated the portion BC_3F_2 trees' genomes inherited from Chinese chestnut v. American chestnut. On average, third backcross trees are expected to inherit 15/16ths (93.75%) of their genome from American chestnut and 1/16th (6.25%) from Chinese chestnut. However, with selection for blight tolerance in each generation, significant portions of Chinese genome may remain especially on chromosomes with genes for blight tolerance. We found that BC_3F_2 seed orchard parents selected for blight tolerance inherited 84% of their genome from American chestnut on average, with a maximum of 99% and a minimum of 61% genome inheritance from American chestnut parents. Furthermore, we found that the percentage of backcross trees' genomes inherited from American chestnut was negatively correlated with blight tolerance **(Figure 2)**.

These results suggest that blight tolerance is controlled by more genes than previously assumed and that backcrossing to American chestnut has diluted out some blight tolerance from Chinese chestnut.

What is next for developing blight-tolerant American chestnut populations?

In addition to completing selection in the current backcross seed orchards,

we are pursuing multiple alternative strategies to develop blight-tolerant populations. First, we are advancing additional backcross lines from additional Chinese chestnut parents through fewer backcross generations to American chestnut. These new backcross lines will be less 'American' on average, but will have greater blight tolerance. We aim to find the optimal balance between blight tolerance and American chestnut traits. Second, we plan to outcross transgenic blight-tolerant American chestnuts trees containing a wheat oxalate oxidase gene to a diverse collection of wild American chestnut trees. Federal review of transgenic American chestnut is ongoing and a decision about whether or not these trees may be distributed to the public is expected in the next few years. Trees from early outcross generations would be available almost immediately after federal approval; however, we estimate that three additional outcross generations will be required to dilute out the transgenic founder genome and incorporate sufficient genetic diversity for regional adaptation. Third, we are pursuing cutting-edge genomics research to identify genetic variants that underlie blight tolerance in Chinese chestnut. This work may enable us to edit the genome of American chestnut to enhance blight tolerance. With all of these approaches applied in parallel and possibly in combination, we remain optimistic about the prospect for large-scale restoration with blight-tolerant American chestnuts.

REFERENCES

Westbrook J.W., Zhang Q., Mandal M. K., Jenkins E. V., Barth L. E., Jenkins J. W., Grimwood J., Schmutz J., Holliday, J. A. (2019). Genomic selection for blight tolerance in American chestnut (*Castanea dentata*) backcross populations. Evolutionary Applications. In review.

2019 Chestnut Photo Contest

Have you taken a photograph that should be highlighted on the cover of *Chestnut* magazine? We want to see it! Send your best chestnut-themed photo(s) to TACF by September 2, 2019 to enter the contest. The winner will receive a complimentary one-year TACF membership and his/her photo will be featured on the cover of a future issue of *Chestnut*.

HOW TO ENTER & CONTEST TERMS:

All entries must be submitted digitally via e-mail or a link to a cloud drive by September 2, 2019;

Entries must include name of photographer and contact information;

Entries must include a full caption including names of subject(s), location, and title;

Entries must be at least 2500 x 3430 pixels (7.6 MBs or larger) and submitted in a jpeg or tiff file format;

All entries must relate in some way to the American chestnut;

Participants are limited to five entries per person;

Entries must be previously unpublished and cannot be entered into another contest.

EMAIL ADDRESS FOR SUBMISSIONS: jules.smith@acf.org

Visit acf.org/our-community/news/enter-to-win-tacfs-2019-chestnut-photo-contest/ for rules and details.

Photo by Andrew Newhouse.

Sandra Anagnostakis:

A TRAILBLAZER AND "THE KEEPER OF THE STUFF"

By Emily Nowels

One day in the mid-1960s at the Connecticut Agricultural Experimentation Station (CAES), geneticist Dr. Richard Jaynes approached Dr. Sandra Anagnostakis carrying a chestnut branch with a blight canker and asked, "Why don't you work on this fungus?"

From that point Dr. Anagnostakis, a fungal geneticist, became a trailblazer in her work with the American chestnut, building a lasting legacy.

"She is probably one of the two most published authors of the chestnut in the United States," said Dr. Fred Hebard, Chief Scientist Emeritus at The American Chestnut Foundation. "Her work was instrumental in the current work on hypovirulence. She was also the one who explored the genetics of vegetative incompatibility, and [...] she's continued and expanded the breeding program."

"You know, I'm basically an optimist." she said. "I wouldn't be working on chestnut if I wasn't an optimist."

Sandra treating chestnut blight cankers to overcome vegetative incompatibility. Photo by Gary Braasch.

Dr. Anagnostakis began her work growing cultures in the lab at CAES, and eventually heard about the blight-tolerant trees growing in Europe. She wrote to a scientist there, who sent her a few cultures, which she began studying. She and her colleagues at CAES soon discovered that the fungus had a virus that inhibited its growth–a major finding that inspired hope for saving the tree. Unfortunately, as they continued to study how the virus was transmitted from one strain to another, they also discovered vegetative incompatibility, which would prove to be a major roadblock in spreading the virus.

Eventually, Dr. Anagnostakis and her colleagues decided that they needed to put their findings to work in the field. After a few greenhouse tests, they received approval from the USDA to plant outside. They planted roughly 75 trees at Lockwood Farm in Hamden, Connecticut, and the trees quickly became infected with chestnut blight. For the next four years, they treated every canker they could reach, and the cankers stopped expanding.

However, in the end, they had to stop treating individual cankers and see if the trees were able to survive on their own. According to Dr. Anagnostakis, at this point about half of the trees survived, and other half died back to the ground. Some resprouted and continued to grow, while others succumbed entirely to the virus.

"So this little plot of trees is my example that not all American chestnuts are created equal. Clearly some of them have more resistance. Or maybe they callus faster. Or maybe they have something in their genetic makeup that allows them to respond faster," Dr. Anagnostakis said.

Many of these trees still survive today. Dr. Anagnostakis and her coworkers went on to plant trees in the forest to see how they survived in a natural environment. Unfortunately, the competition in the forest proved too stiff for the American chestnuts to survive.

From there, Dr. Anagnostakis researched the tannin levels in the trees and oxalic acid levels in the fungus. Although none of these studies or experiments provided a singular answer to saving the American chestnut, each provided insight into new strategies and areas of future research.

Dr. Anagnostakis frequently worked with other American chestnut pioneers along the way. She recalled one day at the station they got a call from a young man who was interested in chestnut breeding. He was calling for a ride to the station from downtown. He had received a ride from a friend with a motorcycle, so they would be able to recognize him by the helmet under his arm.

"So we went downtown and sure enough there was this tall, skinny guy with a motorcycle helmet under his arm, and it was Fred Hebard," Dr. Anagnostakis said chuckling.

Now, Dr. Hebard is well known at The American Chestnut Foundation for his long career as chief scientist at the research farms in Meadowview, VA. This meeting was the first of many between Dr. Anagnostakis and Dr. Hebard. Once Dr. Hebard began his work with the American chestnut, he would frequently travel to gather chestnut material for breeding. During these visits he would stay with Dr. Anagnostakis at her home.

"She lived in a huge old house, and it was broken up into apartments. All the paneling and all the trim was chestnut," Dr. Hebard recalled. "I slept on the landing between second and third floor. There was all this chestnut paneling all surrounding me. That was pretty cool."

In addition to her impressive scientific contributions to the field, Dr. Anagnostakis has jokingly (and yet aptly) given herself another title: "The Keeper of the Stuff."

And for good reason.

Along the way, Dr. Anagnostakis learned from Dr. Charles Burnham that the USDA had extensive records of all of the imported chestnut trees dating back to the early 1900s. She recognized the value in these documents and was overjoyed to take them when the previous owner was cleaning house.

Inside the catalog lives an extensive list of chestnut trees sorted, more or less, by date of import and location. This means someone from Illinois could call Dr. Anagnostakis at CAES, and ask about a chestnut tree in their backyard. With the person's

Sandra working in the lab during her earlier days at CAES.

address and providing that the tree was registered, she should be able to tell them what type of chestnut tree the person is looking at, the date it was planted, and even potentially who planted it. As Dr. Anagnostakis put it, "This is a treasure trove!"

In addition, Dr. Anagnostakis inherited all of the breeding records. "They are notebooks[...], all stained and tattered and much used. They go back to the first chestnut breeding that the USDA did, right up to 1950." she said. "So there is a little notebook for each year saying what they crossed, where the trees were, and some other things. Some of the writing leaves much to be desired, but mostly it's decipherable."

Dr. Anagnostakis has been scanning all of this information and uploading it online to her webpage through CEAS. But realizing that she will not be able to house this valuable information forever, she began looking for a permanent home for the catalog. So, she reached out to the National Archive, who agreed to accept all of the files in a special collection.

Though Dr. Anagnostakis officially retired several years ago, she is far from finished. In addition to working on the USDA catalog, she is officially an Emeritus Scientist at the CAES. She still has an office and access to the farm and plantation. "Usually what happens is the people retire, and then they keep their office and equipment and write books," Dr. Anagnostakis said. "But I didn't want to do that. I want to keep working on the trees."

While she no longer works in the lab, she continues to breed backcrosses, upkeep the official running list of cultivars, and will occasionally go out to check on a tree someone calls about. "I will keep doing crosses as long as I am able to drive to New Haven," Dr. Anagnostakis said. This understated and yet fierce dedication to her work feels like a snapshot of how she has approached much of her chestnut career.

So where does Dr. Anagnostakis see the future of the American chestnut?

"You know, I'm basically an optimist." she said. "I wouldn't be working on chestnut if I wasn't an optimist."

TACF PLANNED GIVING

PLAN TODAY, GIVE TOMORROW

A simple way of living your legacy toward the restoration efforts undertaken by TACF, is to leave a gift through your will or estate. Leaving a gift to TACF after your lifetime allows us to continue our critical research toward creating a future of forests filled with American chestnut.

Charitable bequests are typically allocated in one of two ways: either as a percentage or as a specific dollar value. For donors who support more than one charitable organization, choosing to allocate certain percentages to their favorite charities might be the best choice. Because the value of an estate will fluctuate, choosing to allocate through percentages ensures the resulting gifts to your favorite charities will remain equitable.

Even if you have already completed your estate plans, there is still time to include TACF. In some states a simple codicil can be used as a supplementary document to update your will without having to redo the entire document.

Should you or your attorney need assistance with the language to incorporate TACF into your will or a codicil to your will, please feel free to contact us. We are here to accommodate you in whatever means necessary to make the restoration of the American chestnut part of your legacy, knowing that bequests are among the most meaningful of charitable gifts.

David Kaufman-Moore,
TACF Donor Relations Manager

THE AMERICAN CHESTNUT FOUNDATION®

Remembering Wisconsin's Largest American Chestnut Tree

By Robert Holland, TACF Member

At some point in the 1880s a farming family from the state of Pennsylvania moved onto a farmstead along what is now La Crosse County Highway C in the township of Hamilton, just north of West Salem, Wisconsin.

According to tradition the farmer's mother, still a resident in PA, sent her son a quantity of American chestnuts gathered from trees growing on the ancestral farm in the midst of the original chestnut forest. These chestnuts were planted into WI soils where they germinated, grew, thrived, and reproduced. Planting sites chosen were in spaces along the line fences (fences that delineate property boundaries between adjacent property owners). The seed was planted along those fences so as to preserve all the cultivatable lands for agricultural use.

The American chestnut tree in this photograph from 1998 was the only known survivor of the original plantings. At that time, it stood in a mixed hardwood forest. However, one could see remnants of the old fence wire that had been utilized for constructing the very first line fence. Short pieces of the wire protruded from the trunk at fence height. Further, this very tree was rated as the largest American chestnut tree in the state of Wisconsin.

During this same period, the American chestnut forest, in a mixed stand of species also called the southern Wisconsin hardwood forest, was known as the largest American chestnut grove in the world. Other species in this forest included black cherry, red oak, white oak, shag bark hickory, red elm, and white elm, among others.

Interestingly, in 1902 that farmstead was sold to a man named Martin Hicks. The paperwork pertaining to the sale of the land noted that American chestnut trees were growing on the property.

Chestnut Chocolate Chip Cookies

(gluten-free)

Certified chef and food writer, Valentina Kenney Wein spends her weekends in her "kitchen retreat" creating delicious, hearty, comforting food to nourish her family and share with her readers. You can follow her blog at http://cookingontheweekends.com. Originally published in the May/June 2012 issue of *The Journal of The American Chestnut Foundation*.

Yield: 3 1/2 dozen

Ingredients

6 tablespoons unsalted butter, softened
1/4 cup granulated sugar
1/2 cup golden brown sugar
1 1/2 teaspoons vanilla extract
1/2 teaspoon sea salt
3/4 teaspoon ground cinnamon
1 egg
1 cup chestnut flour*
3/4 teaspoon baking soda
1 1/4 cups semi-sweet chocolate chips
About 3 whole roasted and peeled chestnuts

Method

Preheat the oven to 350°F and line baking sheets with parchment paper. Set aside.

In a medium-sized mixing bowl, cream the butter with the sugars, vanilla, salt and cinnamon. Mix on low for one minute.

Sift the chestnut flour into the batter with the baking soda. Mix just until the flour is fully incorporated.

Fold in the chocolate chips and use a 1-inch ice-cream scoop to shape your cookies, placing them on the parchment-lined baking sheets about two inches apart.

Slice the roasted chestnuts thinly and put a half of a slice on top of each ball of cookie dough. (Only add this step if you plan to eat the cookies the day you bake them – the texture of the nut isn't as good the next day.)

Bake in the preheated oven until the cookies are golden brown, about 9 minutes.

Let them sit for a minute on the baking sheet, and then carefully use a flat-bottomed, metal spatula to move them to a cooling rack. Serve at room temperature.

*Chestnut flour has a shelf life of about one month. Store it in an airtight container in a cool place. You can buy chestnut flour at specialty stores or online (www.nuts.com).

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2019 TACF Annual Meeting

OCTOBER 19, 2019
GETTYSBURG, PA

Gettysburg, PA is a lovely and welcoming country town. From the sacred hallowed grounds of the battlefield, to the unique shops and restaurants downtown, it is rich in history and hospitality.

This setting will be the backdrop for The American Chestnut Foundation's 2019 Annual Fall Meeting on Saturday, October 19, 2019 at the Wyndham Gettysburg.

Join us as we gather to discuss the history, significance, and future of the American chestnut tree! A variety of speakers will share personal chestnut stories, scientific advancements toward restoration, and experts from Gettysburg will bring the town's history to life.

SAVE THE DATE! Online registration and hotel reservations begin June 1, 2019.