


# 2016 Annual Report



THE  
AMERICAN  
CHESTNUT  
FOUNDATION®







OUR VISION is a robust eastern forest restored  
to its splendor.

OUR MISSION is to return the iconic American chestnut  
to its native range.

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These are exciting times for The American Chestnut Foundation (TACF). After many years of breeding and assiduous field work by our chapters, recent developments in our scientific research will allow us to better evaluate the genetics of our trees in order to improve disease resistance and diversity. With support from the New York Chapter, researchers at SUNY-ESF have developed a virtually 100 percent American chestnut tree, which promises to provide resistance to chestnut blight.



With science rapidly evolving, it was evident to our staff and Board that TACF needed a new Strategic Plan to address new challenges both now and over the next decade. Not only did we need to plan immediately for what we know now, but we also needed to contemplate ways to prepare ourselves to tackle new opportunities. The new science of tomorrow will likely enable TACF to more effectively move toward our goal of restoration of the American chestnut on a large scale.

Our Strategic Plan as well as much of the work accomplished by our Board and committees this year begins the integration of three major parallel tracks: breeding, biocontrol, and biotechnology (see p. 3). Each of these approaches will complement each other and strengthen our work. As we all know, wrestling with decisions about future directions is difficult and time-consuming, but it is also illuminating. We are a passionate and determined group. Our vision is broad, and spans many decades, just like our tree. Once the American chestnut is restored, TACF's ultimate goal is the return of healthy eastern forests, and we believe this journey will be a template for the restoration of other threatened species.

In this major effort, TACF is honored to work with an incredible team of supporters in achieving our goal. Volunteers, members, chapters, partner organizations, research affiliates, and staff have all been an integral part of the planning and implementation process. On behalf of TACF's Board of Directors, we are grateful for your dedication and support.

Sincerely,

A handwritten signature in black ink that reads "Michael Doochin".

Michael D. Doochin  
Chairman, Board of Directors



## 2016 Science Briefing



TACF Director of Science Jared Westbrook evaluates resistance for a progeny test at the Georgia Forestry Commission in Dawsonville, GA. Photo by Jeff Donahue



The American Chestnut Foundation (TACF) has worked diligently throughout FY 2016 to develop a vision for American chestnut restoration. This critical vision builds upon the work of the past, embraces new technology, and fosters collaboration among all those dedicated to restoring this iconic tree.

Scientists affiliated with TACF recently authored an open-source scientific article called “**Rescue of American chestnut with extraspecific genes following its destruction by a naturalized pathogen**” that provides background and progress on backcross breeding as well as transgenic approaches to blight-resistant American chestnut. This is one example of TACF’s commitment to continue building on the work of our predecessors.

In addition, a comprehensive draft of TACF’s **10-year breeding and science plan** was approved by its Board of Directors. Utilizing new technologies, this plan seeks to speed up selection within the backcross-breeding program by incorporating early screening and genomics to select trees for disease resistance.

TACF’s Board also led an exceptional effort in fostering collaboration with the approval of the **3BUR** document (Breeding, Biotechnology, and Biocontrol United for Restoration). 3BUR states that TACF will seek to merge the backcross and transgenic programs while also pursuing these approaches separately, pending regulatory approval to release a blight-resistant transgenic American chestnut.

All of these efforts resulted in the approval of TACF’s **2017-2027 Strategic Plan**, which details steps the Foundation will take within the next ten years to advance from research and development to species restoration. Genomic research and integration of both the transgenic and backcross programs are key components of each of these plans.

## DE-MYSTIFYING DISEASE RESISTANCE AND SPEEDING UP SELECTION WITH GENOMICS

TACF is investing in genomics to open up the black box of disease resistance in TACF’s breeding populations. The goal of ongoing genomics research is to understand what chromosomes and genes contribute to disease resistance. TACF is performing research to answer questions such as: How many genes contribute to blight resistance? Do American chestnut backcross trees with different Chinese chestnut ancestors harbor unique resistance genes? Did different American chestnut backcross trees inherit the same resistance genes from their Chinese chestnut ancestor?

The answers to these questions have significant bearing on TACF’s breeding program moving forward. If more than two or three genes control resistance to chestnut blight, and backcross trees inherited different subsets of resistance genes from Chinese chestnut ancestors, genomics will help TACF determine which subset of resistance genes specific backcross trees inherited. TACF will utilize this information to guide future breeding between trees with different resistance genes to enhance resistance in future generations.



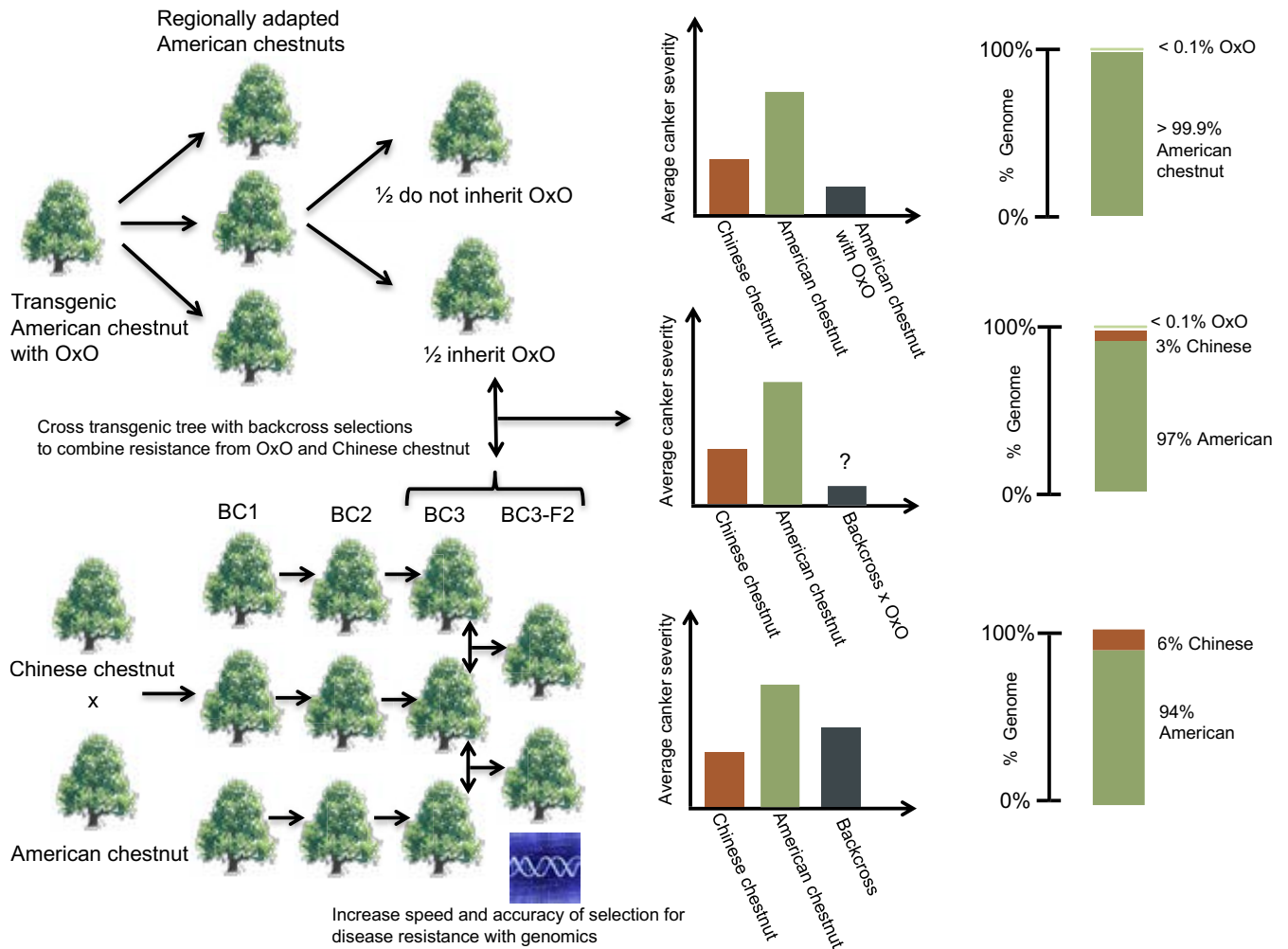
Gene mapping at Virginia Tech.

With collaborators at Virginia Tech and Clemson University, TACF is mapping genes for resistance to chestnut blight and Phytophthora root rot, the two major diseases that have devastated American chestnut across its native range. Mapping resistance genes is accomplished by scanning the genomes of backcross trees for correlations between DNA variants and variation in disease resistance in TACF’s backcross populations.

Funding from the Tucker Foundation has allowed DNA sequencing to be conducted on backcross trees selected for blight resistance as well as those trees’ Chinese chestnut ancestors. The work intends to show whether backcross trees inherited the same or different regions of DNA from Chinese chestnut ancestors.

Mapping resistance genes requires a high-quality chestnut reference genome, which places all 38,000 genes in the chestnut genome into the context of the chestnut’s 12 chromosomes and 800 million DNA base pairs. With funding from the Colcom Foundation, TACF is partnering with the HudsonAlpha Institute of

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Biotechnology to sequence and assemble a reference genome for American chestnut.

Genomics will also speed up selection of the most disease resistant trees. Since 2002, more than 60,000 advanced-generation backcross trees have been planted in seed orchards at TACF's Research Farms in Meadowview, Virginia. These trees have been artificially inoculated with the chestnut blight fungus, and more than 90% of the most susceptible trees have been culled. To date, there are about 5,400 trees remaining in Meadowview seed orchards. TACF's final goal is to cull all but 500 - 600 of the most resistant trees. As culling continues, it becomes increasingly difficult to visually assess which trees are most blight-resistant.

TACF is partnering with Virginia Tech to complete selection of the most resistant trees in Meadowview seed orchards using a method called genomic selection. With a grant from the USDA National Institute of Food and Agriculture, a method to predict disease resistance from DNA sequence is being developed by sequencing

DNA from mother trees whose progeny have been screened for resistance to chestnut blight and or Phytophthora root rot. The genetic resistance of these mother trees is estimated from the average degree of disease-severity among their progeny. DNA sequencing on these mother trees will be used to estimate genome-wide relationships between DNA sequence variants and variation in the average disease resistance of the progeny of these mother trees. The relationship between DNA variants and disease resistance will in turn be used to predict the underlying genetic levels of resistance of mother trees whose progeny have not been screened for resistance. TACF received additional funding from the Allegheny Foundation and an anonymous donor to sequence DNA and to predict the genetic resistance of more than 3,500 of the 5,400 remaining mother trees at Meadowview. This DNA sequencing will help TACF complete selection of the most resistant trees in Meadowview seed orchards within three to five years.



POTENTIAL INTEGRATION OF THE TRANSGENIC AND BACKCROSS BREEDING PROGRAMS

For nearly three decades, collaborators at State University of New York College of Environmental Science and Forestry (SUNY-ESF) have pursued a transgenic approach to introduce blight resistance into American chestnut. The transgenic approach uses a bacterium to transfer candidate genes that potentially enhance disease resistance from other species into American chestnut. Collaborators at SUNY-ESF discovered that a gene from wheat, oxalate oxidase (OxO), enhances blight resistance in American chestnut to levels that are potentially greater than Chinese chestnut. When a transgenic American chestnut containing OxO is bred with a susceptible American chestnut, ½ of their progeny inherit the blight resistance-enhancing OxO gene. The blight resistance conferred by OxO is dominant, meaning a seedling progeny needs to inherit only one copy of the gene from one parent to have the full blight-resistance conferred by the gene. This is potentially a major breakthrough for American chestnut restoration. The transgenic tree can

be crossed with susceptible wild American chestnuts in order to conserve much of the genetic diversity and potential regional adaptation that remain in the species after its eradication by chestnut blight.

Breeding transgenic trees with backcross trees has the potential to produce progeny with a more robust resistance to chestnut blight than either backcross or transgenic strategies pursued independently. There is a potential for even greater levels of blight resistance to develop if backcross trees harboring genes for blight resistance from Chinese chestnut genes are crossed with transgenic American chestnut trees with the OxO gene. Combining the resistance mechanisms of OxO and Chinese chestnut through breeding may provide the progeny of these crosses with a multifaceted resistance that is more durable to evolutionary changes in the chestnut blight fungus, thereby reducing the chance that the fungus could overcome the resistance mechanisms of American chestnut trees.



Yoks Bathula harvests transgenic trees in New York using a supplemental wire mesh bag, required by SUNY's permit. Photo by Andy Newhouse



Volunteers test trees for resistance by inoculating them with blight fungus at Meadowview Research Farms.



Prepping a strain of blight fungus (EP155) to inoculate trees at Meadowview Research Farms.



Front and side view images above show transgenic and non-transgenic seedlings side-by-side. Photos taken and composites arranged by Andy Newhouse.



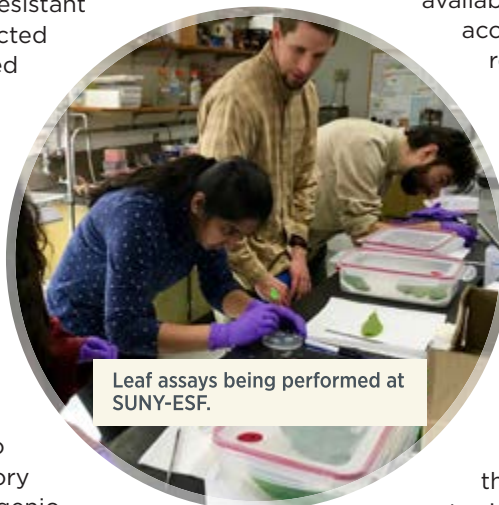
In addition to potentially providing more durable resistance to chestnut blight, breeding backcross trees with transgenic trees is likely to be the most efficient and effective approach for incorporating resistance to *Phytophthora* root rot into American chestnut trees. Seedling progeny from such a cross which have inherited the OxO gene can be rapidly identified through an assay of oxalate oxidase activity. Transgenic blight-resistant backcross progeny would then be inoculated with *Phytophthora cinnamomi*, to identify individual trees that are most resistant to *Phytophthora* root rot. Selected individuals would then be interbred to further enhance resistance to *Phytophthora* root rot, while retaining the OxO transgene. A transgenic x backcross approach will require two additional generations of breeding and selection for inheritance of blight resistance from OxO and resistance to *Phytophthora* root rot from TACF backcross breeding program.

Researchers at SUNY-ESF intend to submit their application for regulatory approval to release the transgenic American chestnut with the OxO gene to the USDA and EPA in 2017. A final decision for regulatory approval could potentially be made as early as 2021. As part of their application to release transgenic American chestnut that is blight resistant, they have conducted research to demonstrate that insertion of the OxO gene does not affect the growth and metabolism of

American chestnut trees and does not have any adverse effects on non-target organisms such as mycorrhizal fungi or insect pollinators.

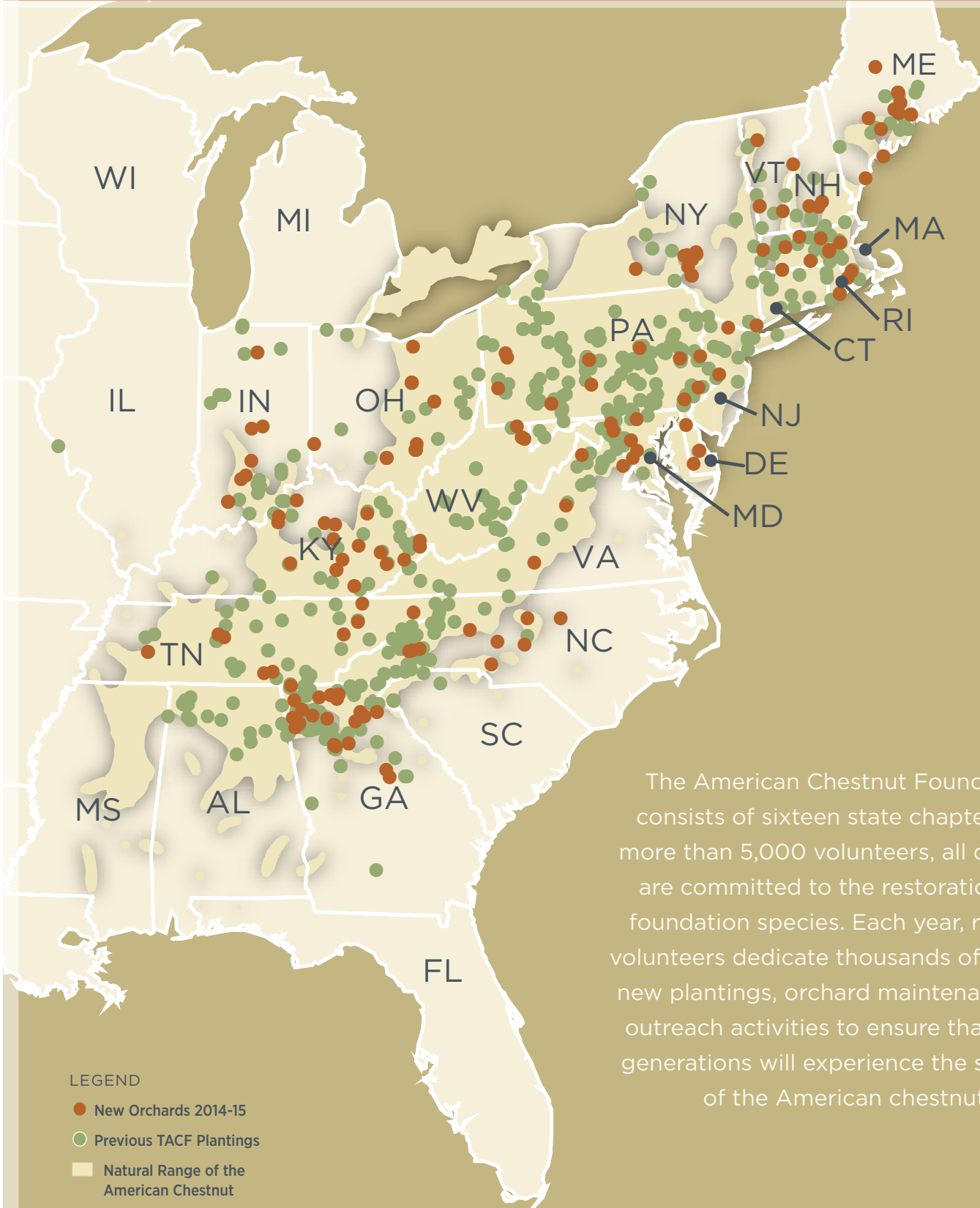
#### IN SUMMARY

The Foundation appreciates the differing opinions of its volunteers and members regarding the use of transgenic technology and backcross breeding. TACF will pursue these approaches in parallel to ensure multiple pathways to mission success using best available scientific methods. TACF will accelerate selection of the most disease-resistant trees within the backcross program using genomics so that individuals will have an option to plant non-transgenic American chestnut backcross trees with improved blight-resistance. For individuals who support the transgenic program, TACF will identify and plant wild American chestnut trees in germplasm conservation orchards. Provided that regulatory approval for the transgenic trees is obtained, these trees can then be crossed with the transgenic American chestnut to incorporate much of the remaining genetic diversity within the native species into the breeding program. TACF also intends to breed backcross trees with transgenic trees to combine the blight-resistance conferred by OxO and genes from Chinese chestnut. We will also cross backcross trees with transgenic trees to hopefully combine resistance to chestnut blight and *Phytophthora* root rot.





# 2016 Regional Accomplishments



The American Chestnut Foundation consists of sixteen state chapters and more than 5,000 volunteers, all of whom are committed to the restoration of a foundation species. Each year, regional volunteers dedicate thousands of hours to new plantings, orchard maintenance, and outreach activities to ensure that future generations will experience the splendor of the American chestnut.

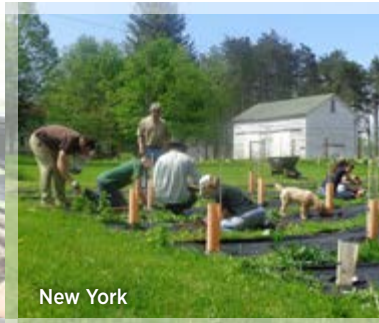




Maine



Vermont



New York



Ohio



Massachusetts



Indiana



Pennsylvania

### NEW ENGLAND REGION

State Chapters: Connecticut, Massachusetts/Rhode Island, Maine, Vermont/New Hampshire

#### Regional Stats:

- Approximately 35,000 chestnuts harvested
- Approximately 10,000 chestnuts planted
- 33 new plantings
- 37 outreach activities, serving approximately 100,000 individuals
- 118 new members enrolled

#### Notable Accomplishments:

- The Massachusetts Horticultural Society Gold Medal for Excellence in Hybridization was awarded to TACF.
- The Connecticut chapter planted a 1.5-acre American chestnut sanctuary in collaboration with the Greenwich Land Trust. More than 350 potentially blight-resistant chestnut seedlings were planted as a part of the 14 acre forest preserve.
- The Maine chapter organized the first airplane survey flights to locate surviving chestnuts by using true color and inferred GPS.
- The Vermont/New Hampshire chapter established two American chestnut reintroduction trials using potentially blight-resistant chestnut – one at UVM's Jericho Research Forest and another at the New Hampshire Division of Forests and Lands' Vincent State Forest.

### NORTH CENTRAL REGION

State Chapters: Indiana, New York, Ohio, Pennsylvania/New Jersey

#### Regional Stats:

- Approximately 41,250 chestnuts harvested
- Approximately 36,875 chestnuts planted
- 9 new plantings
- 45 outreach activities, serving approximately 17,500 individuals
- 189 new members enrolled

#### Notable Accomplishments:

- The Indiana chapter created and implemented the new Southern Hills Restoration Branch.
- The Pennsylvania/New Jersey chapter established a research planting that holds more than 27,500 chestnut trees in New Tripoli, Pennsylvania.
- A large-scale chestnut education plot at the 4-H Center that serves more than 6,000 people annually in Camp Palmer, Ohio was established.
- More than 2,000 “mother” tree chestnuts were distributed state-wide to New York chapter members.





West Virginia



Virginia



Carolinas



Alabama



Maryland



Tennessee

### MID-ATLANTIC REGION

State Chapters: Maryland, Virginia, West Virginia

#### Regional Stats:

- Approximately 15,000 chestnuts harvested
- Approximately 1,200 chestnuts planted
- 16 new plantings
- 57 outreach activities, serving approximately 3,500 individuals
- 209 new members enrolled

#### Notable Accomplishments:

- The Virginia chapter established an online database to track all ceremonial plantings for all potentially blight-resistant chestnuts, with more than 300 recorded.
- The Maryland chapter signed an MOU with the Central Maryland Research and Education Center of the University of Maryland to establish a second seed orchard for the state.
- The West Virginia chapter planted a 100 chestnut seedling demonstration orchard at the West Virginia University Research Forest.

### SOUTHERN REGION

State Chapters: Alabama, Carolinas, Georgia, Kentucky, Tennessee

#### Regional Stats:

- Approximately 14,000 chestnuts harvested
- Approximately 4,000 chestnuts plants
- 43 new plantings
- 50 outreach activities, serving approximately 8,600 individuals
- 157 new members enrolled

#### Notable Accomplishments:

- More than 300 chestnuts were harvested and control pollinated to further the Carolinas chapter research on Phytophthora root rot resistance.
- The Georgia chapter established two new, high-profile plantings at the Carter Center and Governor's mansion.
- The Kentucky chapter developed a collaborative program with Eastern Kentucky University, which will include a seed orchard, several education programs, and demonstration plantings at two natural areas.
- The Alabama chapter helped implement a new restoration branch.
- The Tennessee chapter tracked and recorded more than 3,000 surviving American chestnut trees.



# Thank You

TO ALL OUR VOLUNTEERS AND SUPPORTERS

The American Chestnut Foundation deeply appreciates the hard work of its chapter volunteers and committed supporters. Our mission is long-range, and your contributions allow this conservation success story to continue. We are truly grateful for your help in restoring this iconic species.



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## A Legacy Tree

### THE PROMISE OF A HUNDRED FUTURE FORESTS

In 2017, The American Chestnut Foundation will relocate its Legacy Tree orchard to the Glenn C. Price Research Farm in Meadowview, VA. This new orchard reflects advances in the breeding program to produce blight resistant American chestnuts. The legacy tree orchard will be composed of progeny of controlled crosses between BC<sup>3</sup>-F<sup>2</sup> parents that have been identified as having among the highest blight resistance in the population. The progeny of these blight resistant parents will be planted away from other susceptible trees. In 5 - 10 years, when the trees in this new Legacy tree orchard flower, they will intercross with each other to produce seed with improved blight resistance as compared with the previous generation.

Legacy Trees are approximately 15/16 American chestnut and 1/16 Chinese chestnut. Each tree is sponsored by an individual(s) - either in their own name, the name of a company or organization, or in honor of a friend or relative. The sponsorship is acknowledged by an individual plaque placed by the named tree.

The Foundation's Legacy Tree sponsors have made a significant contribution to help fund the scientific research of this foundation species. The seeds produced in the orchard contain the greatest level of American chestnut characteristics and the most blight-resistance to date.

Legacy Trees are truly a living legacy of TACF's scientists, staff, state chapters, and volunteers. This magnificent orchard is only the beginning of the Foundation's restoration efforts. Scientific rigor, diligent support, and ongoing patience will bring our breeding strategy to fruition over the next 100 years.





OUR LEGACY TREE SPONSORS

21st Century Parks Inc. in honor of George Gibbs, Louisville, KY

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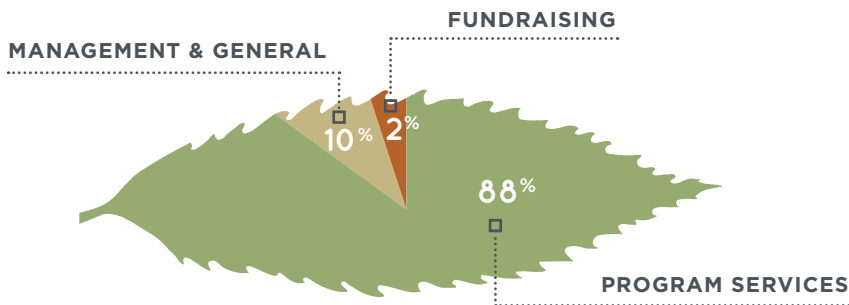
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Stephen and Suzan Zoukis in honor of Cashiers Village

**Legacy Sponsor names appear on individual plaques located near each tree**

# 2016 Financial Overview

PUBLIC SUPPORT AND OTHER REVENUES		% of Income
Contributions	\$1,933,312	71%
Membership	365,647	13%
Federal Grants	98,886	4%
Investment Income	321,443	12%
Merchandise Sales	6,275	0%
Realized losses on investments	(9,016)	0%
Other Income	16,314	1%
Loss on sale of assets	(4,856)	0%
<b>TOTAL REVENUE</b>	<b>\$2,728,005</b>	
EXPENSES		% of expense
<b>Program Services</b>		
External Research	\$462,743	16%
National Office Operations	481,040	16%
Marketing & Communications	145,618	5%
Regional Science	662,074	22%
Education & Outreach	5,670	0%
Chapter & Membership	218,612	7%
Research Farm Operations	564,233	19%
Forestry & Mine Land	44,309	2%
<b>TOTAL PROGRAM SERVICES</b>	<b>\$2,584,299</b>	<b>88%</b>
<b>Supporting Services</b>		
Management & General	\$299,685	10%
Fundraising	\$68,801	2%
<b>TOTAL SUPPORTING SERVICES</b>	<b>\$368,486</b>	<b>12%</b>
<b>TOTAL EXPENSES</b>	<b>\$2,952,785</b>	



TACF IS EXTREMELY PROUD  
OF ITS 2016  
**4-Star Rating**  
FROM CHARITY NAVIGATOR.



# The American Chestnut Foundation



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