

RESTORING THE AMERICAN CHESTNUT



NESAF WINTER MEETING
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KENDRA GURNEY
NEW ENGLAND
REGIONAL SCIENCE COORDINATOR



AMERICAN CHESTNUT: THE PAST 100 YEARS



PRE-BLIGHT USES,
BLIGHT INTRODUCTION AND SPREAD,
EARLY SPECIES RESTORATION WORK



American Chestnut: The Tree



- Major component of eastern forests
- Fast growth, large size, extremely rot resistant



American Chestnut: The Tree



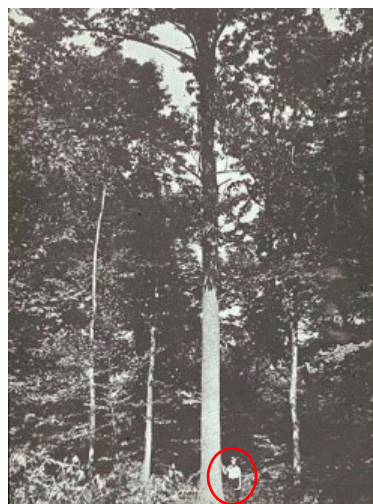
- High-value timber species
- Tannins used in tanning leather



American Chestnut: The Tree



- Nuts valuable to wildlife
- Nuts also valuable to people and livestock
- Culturally significant

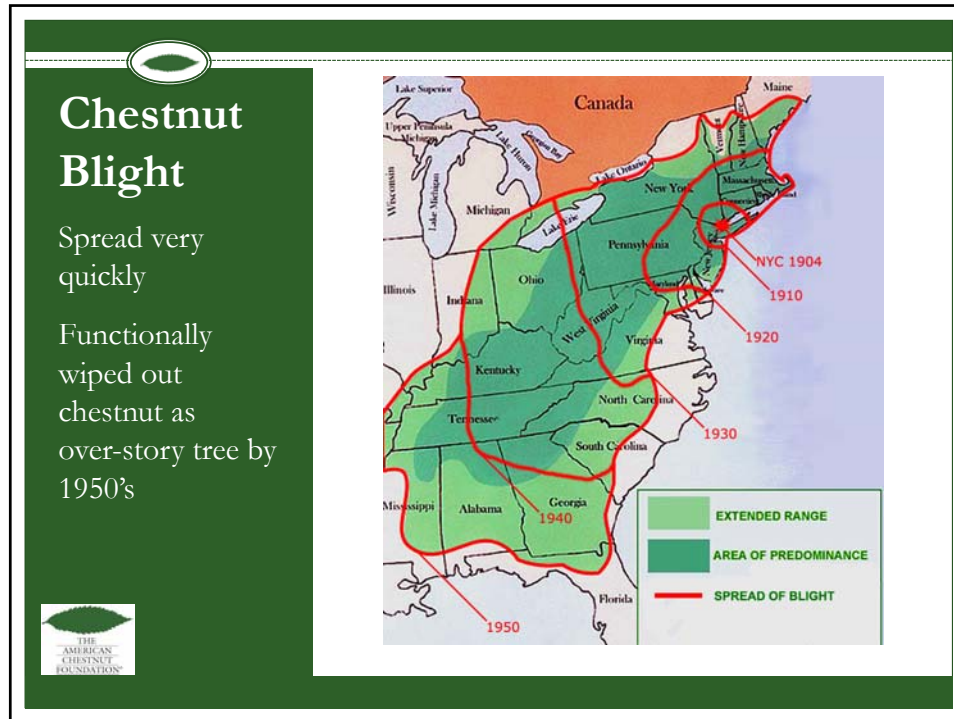


Chestnut Blight



- Blight first identified in New York City in 1904





Chestnut Blight

- Fungus (*Cryphonectria parasitica*) girdling canker
- Two types of spores – airborne ascospores (sexual spores) and sticky, spiral conidia (asexual spores)

THE AMERICAN CHESTNUT FOUNDATION

Chestnut Blight Cankers





Healing cankers	Sunken cankers
	

Early Restoration Attempts





- Cultural methods
 - Tree surgery, fungicide, forest gap barriers, eventually removal
- Identifying natural resistance among American chestnuts
 - 90% of existing trees determined to have escaped blight
 - Small number existing with a low level of natural blight resistance
- Replacement tree to fill niche
 - Expeditions to Asia, high hopes for Chinese chestnut
 - Ultimately failed, due to poor understanding of Chinese chestnut habitat requirements
 - Chinese chestnut since proven too small to compete in a mature forest setting



Chinese chestnut

Early Breeding



- USDA Breeding Program
 - American chestnut x Chinese chestnut, Japanese chestnut or other
 - Only 50% American chestnut, or less if backcrossed to another Asian chestnut parent
 - Poor American characteristics and blight resistance – project abandoned in 1960's
- CAES Breeding Program
 - Started by Arthur Graves in 1920's and continued today
 - Wide variety of hybrids and backcrosses
 - Looking for blight-resistance and timber form
 - Working with the fungus as well



TACF Backcross Breeding



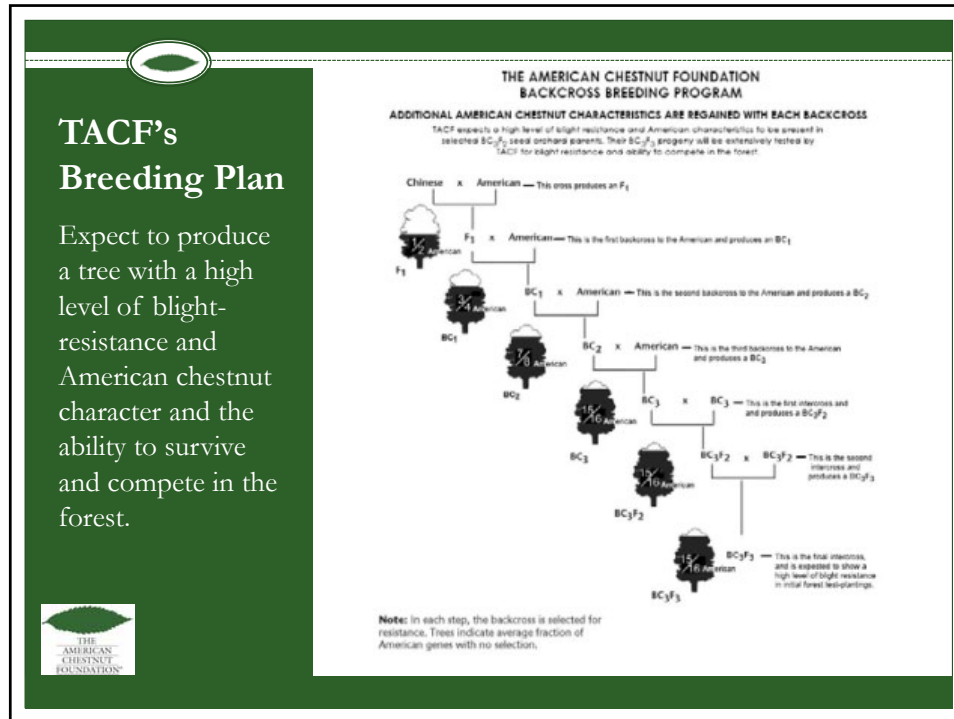
- Combination of **hybrid** and **backcross** breeding
- **Hybrid** cross captures blight-resistance
 - American x Chinese
- **Backcrosses** breed out Chinese chestnut character and include more American chestnut character
 - Repeat enough times to re-capture desired amount of American character (3 or 4 times)
- **Intercrosses** (hybrid of two backcrosses) increase blight-resistance further
 - Each intercross increases chance of breeding trees homozygous for resistance (2 times)



American chestnut



Chinese chestnut



Backcross Breeding Program

- **F1:** all are moderately resistant.
- **All backcrosses:** 1 out of 8 theoretically should be moderately resistant. The rest will vary in resistance from none to less than moderate.
- **First intercross:** 1 out of 64 theoretically should be highly resistant.
- **Second intercross:** all theoretically should be highly resistant.

Resistant canker

Susceptible canker

Hypovirulence



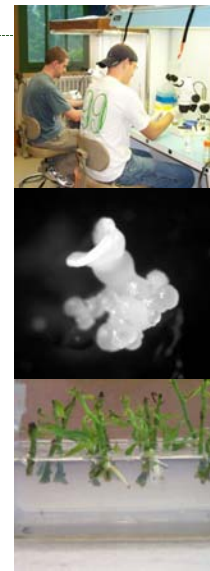
- Discovered in France
- Virus that infects the blight fungus, reducing virulence
- Causes “big ugly” cankers
- Infection requires compatibility



Transgenics and Cisgenics



- *Agrobacterium*-mediated transformation
 - Used to insert putative resistance enhancing gene
- Embryos are moved to a medium containing antibiotics that will kill the bacterium
- Utilize green fluorescent protein marker
- The transformed embryos are then multiplied and regenerated into whole plants
- Initial genes of interest came from wheat, now also focusing on genes from Chinese chestnut



Mapping Chinese Chestnut for Blight-Resistance



- Genetic Marker/Mapping
- Physical Mapping and DNA Libraries
- DNA Sequencing
- Comparative Genomics
- **Total = Gene Discovery**



Chestnut Restoration: No “right” way

American chestnut restoration efforts have relied on a variety of approaches



- **Traditional breeding for blight resistance**
- Hypovirulence and research on the fungus (biological control)
- Transgenic and cisgenic technologies to confer blight resistance
- Genetic mapping to identify genes for blight resistance



Lessons Learned



- Germplasm reservoir
 - American chestnut germplasm is not lost by cutting
 - Other methods may be more applicable to other species
 - ✦ Seed banks
 - ✦ Germplasm conservation plantings
- Multiple approaches
 - American chestnut has benefited from a variety of approaches and levels of follow-through – and still does
 - Continued interest in species restoration



RESTORING THE AMERICAN CHESTNUT



Goals:

- To develop blight-resistant American chestnuts
- To ensure regional adaptability
- To ensure long-term resistance



TACF Backcross Breeding Program



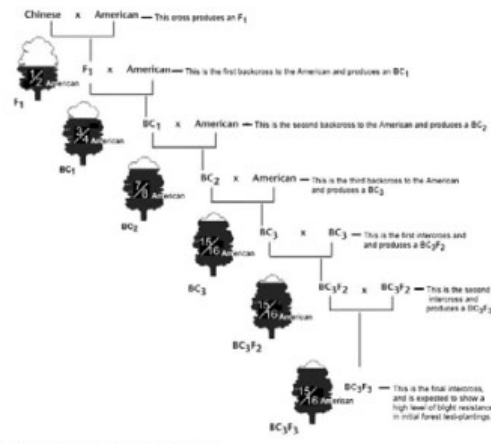
TACF's Breeding Plan

Expect to produce a tree with a high level of blight-resistance and American chestnut character and the ability to survive and compete in the forest.


**THE AMERICAN CHESTNUT FOUNDATION
 BACKCROSS BREEDING PROGRAM**


ADDITIONAL AMERICAN CHESTNUT CHARACTERISTICS ARE REGAINED WITH EACH BACKCROSS

TACF expects a high level of blight resistance and American characteristics to be present in selected BC₃F₃ seed orchard parents. Their BC₃F₃ progeny will be extensively tested by TACF for blight resistance and ability to compete in the forest.



Note: In each step, the backcross is selected for resistance. Trees indicate average fraction of American genes with no selection.







Meadowview Research Farms

Location:
Meadowview, VA

Support:
TACF's Chief Scientist Dr. Fred Hebard, research assistants and farm staff



- Completed 4 generations of breeding since 1989
- Started with 2 advanced sources of resistance
 - 'Clapper' - 1st backcross from USDA breeding program
 - 'Graves' – 1st backcross from CAES breeding program
- Adapted cultural methods to reduce generation times
 - Flowering and selection size achieved in 2-4 years
- Currently home to 34,000 trees on over 150 acres





Goals:

To develop blight-resistant American chestnuts

To ensure regional adaptability

To ensure long-term resistance





TACF Backcross Breeding Program

TACF State Chapters

Currently 17 state chapters from Maine to Georgia



- Carry out the TACF breeding program locally
- Tasked with inventorying local, wild American chestnuts
- Conducting controlled pollinations and harvesting nuts
- Planting and maintaining orchards
- Educating the public
- **All-volunteer** with regional staff support

Goals:

To develop blight-resistant American chestnuts

To ensure regional adaptability

To ensure long-term resistance



TACF Backcross Breeding Program



TACF has been producing potentially blight-resistant nuts at our Meadowview Research Farms since 2007

Progeny testing began in 2009

This effort will require a range of partners to complete





Progeny Testing – Forest and Orchard

OPPORTUNITIES FOR PARTNERSHIP



PROJECTS AT THE LOCAL AND RANGE-WIDE SCALE








New England Chapters

Four chapters covering all six New England States:

- ME
- VT/NH
- MA/RI
- CT



Chapter Orchards



Breeding Orchards	Seed Orchards
<ul style="list-style-type: none">• <u>10-15</u> year commitment• Last backcross generation of breeding – trees are moderately resistant• Mostly complete in New England –<ul style="list-style-type: none">○ MA/RI – Nanking○ VT/NH and CT – possibly one more site	<ul style="list-style-type: none">• <u>30-45</u> year commitment• First intercross generation - produce potentially blight-resistant trees• Actively seeking sites in MA/RI, VT/NH and CT

American Germplasm Conservation



- Most current germplasm conservation is addressed by including trees in the backcross breeding program
- Germplasm Conservation Orchards are a way to preserve additional native germplasm
- Increase efficiency
- “Perpetual orchard”
- Flexible design



Progeny Test



- Used to test the trees in the seed orchards to better refine our breeding stock
- Guided by TACF's Testing Task Force Protocols
 - Installing both forest and orchard progeny tests
- Aim to include 20 B3F3 families (or more) plus controls
 - Plantings are often 500-1,000 trees, but dependent on site size and material availability (1-2 acres, on average)
- To test all families from Meadowview Research Farms will require a minimum of 270 progeny tests
 - Approximately 25 in the ground thus far, another 20 or so planned for 2014 installation

Reintroduction Trial



- Forest plantings aimed at evaluating silvicultural techniques for reintroduction of potentially blight-resistant material
 - Similar planting materials as used in Progeny Testing
- Forests have changed dramatically since the loss of American chestnut
- Design will be variable, based on the goals of the test



Restoration Plantings



- Planted with material proven to have sufficient blight-resistance through progeny testing
 - Will not likely begin installing these plantings until 2020 or 2025
- Expect a majority of the trees to have a high level of blight-resistance
- Original range of American chestnut is 180 million acres
 - Estimated currently about 95 million acres of suitable habitat, 24 million acres of optimal habitat
 - Replanting that acreage, even within the next 100 years, is a HUGE task...but we'll do our best!

Chapter Partnerships

A quick glance at some of the partnerships our TACF Chapters have established...



- **ME:** US Forest Service, Penobscot County Conservation Association, University of Maine, Viles Arboretum, Merryspring Nature Center, Northeast Wilderness Trust, Unity College, The Nature Conservancy, Small Woodland Owners Association of Maine...
- **VT/NH:** US Forest Service, University of Vermont, NH Div of Forestry, Merck Forest and Farmland Center, Beaver Brook Association, Public Service of New Hampshire, Green Mountain Power, National Grid, Bartlett Tree Experts, St. Anslem College, VELCO, VT State Parks, Asplundh, Davy Tree...
- **MA/RI:** MA Fish and Wildlife, MA DCNR, Old Sturbridge Village, National Grid, NSTAR, NRCS, University of Massachusetts, University of Rhode Island and URI Master Gardeners, Smith College, South Kingstown Land Trust...
- **CT:** University of Connecticut, Woodbridge Land Trust, Northern CT Land Trust, Great Mountain Forest Corp, Litchfield Hills Audubon, Town of Middletown, Middletown Garden Club, Bartlett Tree Experts, Connecticut Light and Power/Northeast Utilities, Town of Guilford...

Got Chestnuts?

Kendra Gurney

TACF New England
Regional Science
Coordinator

705 Spear St
USFS Northern
Research Station
South Burlington, VT
05403

kendra@acf.org
802-999-8706 (cell)



• TACF Website

www.acf.org

• Tree Locator Form

http://www.acf.org/find_a_tree.php

○ Report an existing tree or sprout