

# Chestnut Tree

## The Pennsylvania Chapter of The American Chestnut Foundation



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### Spring Members and Growers Meeting

This meeting will kick-off our new County Programs with the emphasis on tree pollinating. Members and county coordinators are invited to participate in this program around Pennsylvania. Our goal is to have one or more counties participate within each of the thirteen regions of PA. New Jersey, Delaware, and Ohio members are also invited to participate.

The meeting is scheduled for the Milton B. Hershey School, Environmental Center in Hershey, PA on March 20. There will be coffee and donuts during registration starting at 8:00 a.m. and the program will begin at 9:00 a.m.

Our agenda will include our keynote speaker, **Dr. Paul Sisco**, TACF Southern Regional Scientist who will make a presentation of "How Cytoplasmic Male Sterility (CMS) works and its application in Chestnut tree breeding". Also invited is Chapter member **Dr. Gary Alt** who will present information about the deer population progress in PA. **Ann Leffel** will review the art and technique of hand pollination that will be so vital to this year's county program. Our workshops include seed and supply distribution; tree pollen drying; Tree location and locator forms; Chestnut harvesting and storage; and inoculation techniques. Lunch will be served in the Environmental Center. Members are asked for a \$5 donation to help pay for registration snacks and lunch. We plan to have a pay-as you-go dinner after the meeting at a local restaurant.



### A Message from the President

- *Bob Summersgill*, (908) 647-5864

The Pennsylvania Chapter has been working with chestnut enthusiasts in neighboring states for many years. In Maryland, we helped plant the first orchard at ThorpeWood in 1999. By 2003, their organization had developed enough to become a fledgling chapter of TACF. This past year, we started working with The New Jersey Conservation Foundation. **NJCF** is doing an outstanding job buying land throughout the state to preserve it for future generations. In a unique agreement, they offered land to plant the first BC3 orchard in New Jersey. **PATACF** may use the land for the life of the trees and they will mow the fields. A second piece of **NJCF** land called the Jarboe site consists of 29 acres and will be planted with TACF Graves BC3F2 material. This is the generation in which one out of every 64 trees should be blight resistant. Also, the NJ Forestry Service has offered their nursery facilities and will put up deer fencing when we plant.

**Dr. Emile DeVito, Dr. Louis Cantafio and Jon Wagar** of **NJCF** also put us in touch with the Morris County Park Commission. After a meeting with **Dave Armstrong** and I, the Park Commission offered **PA-TACF** orchard sites on Morris County Park land. The current plan is to use Morris County's hilly park sites for the cytoplasmic male sterility (CMS) Program (see page 2). **Charley Zafonte and Rob Jennings** work for the park commission and are very enthusiastic and are working out details.

Mount Paul Memorial Park in Chester, New Jersey is the first site selected. The park department will mow the fields, purchase a 12 X 18 foot shed for our supplies and make the trees available to TACF volunteers for the life of the trees. The only cost to our chapter will be supplies such as tubes, herbicide and fertilizer, plus a lot of effort on the part of volunteers. A mower and heavy-duty lawn cart have already been donated to the chapter. Volunteers in NJ and PA will help oversee the care of these future backcross trees.



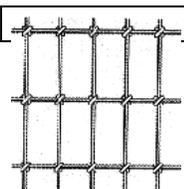
There will be a PA-TACF Board meeting at the MBH School Horticulture Center At 3:00 p.m. on March 19<sup>th</sup>.

### DIRECTIONS TO MILTON HERSHEY SCHOOL

From US 322, turn onto Meadow Lane. Continue on Meadow Lane for 0.5 mile to Crest Lane. Turn right to the Environmental Center.

### FENCING AVAILABLE to CHESTNUT ORCHARD GROWERS

See page 6 for details!!



### Calendar of Events:

- |                |   |
|----------------|---|
| March 20       | Spring Meeting at Milton Hershey School |
| June/July      | County Pollinating                      |
| August 17-19   | PSU Ag Progress Days, Rock Springs, PA  |
| Sept/Oct       | County Harvesting and Nut Storage       |
| Oct 16         | PA-TACF Fall Meeting, Pittsburgh, PA    |
| Oct 29-30      | TACF Annual Meeting in Asheville, NC    |
| Jan 8-15, 2005 | PA Farm Show                            |



## Orchard Signs

This year at our Spring Members meeting on March 20, we will be distributing orchard signs. We plan to distribute to our larger orchards first and then to some to the smaller orchards. Please see Dave Armstrong at the meeting.

## The County CMS Program and Hand Pollinating Chestnut Trees

- Dave Armstrong

**County Breeding Programs:** The PA Chapter is entering a new phase of chestnut tree breeding that we believe will produce more adaptable Pennsylvania trees with multiple sources of blight resistance. The program relies on local coordinators and volunteers to do tree location, pollinations, harvesting, seed storing and planting within each county. The program also recommends smaller orchards than we have planted in the past. It utilizes genetic emasculation, Cytoplasmic Male Sterility (CMS), that occurs in interspecific hybridization of chestnuts. The incorporation of that phenomenon into a chestnut breeding program was proposed by our Chapter Scientist, Dr. Bob Leffel and his colleagues.

We refer to this County project as **the CMS program**. By crossing American trees female flowers (burs) with Chinese male flowers (catkins), we produce seed for a male sterile tree (F1). The great advantage of the CMS Program is that, after the first hand cross (F1), subsequent generations (BC1, BC2, etc) may be produced via open pollination in isolation from all chestnuts except American. Open-pollination is a superior method: one can produce more nuts without the labor-intensive bagging/hand pollinating process.

We recommend that each County do their own CMS program with assistance, education, supplies and support from the Chapter and your county coordinator (List available at [http://chestnut.cas.psu.edu/maps/PA\\_regions.html](http://chestnut.cas.psu.edu/maps/PA_regions.html) or by contacting the Chapter). Our goal this year is to have one or more county CMS programs organized in each of the 13 PA regions. To participate in the launching of this project, be prepared to locate and pollinate trees this spring. Beginning a program can be done by a county team or by an individual. **Start-ups will be a feature of the Spring Meeting, March 20, 2004.**

The following describes the chestnut tree hand pollination process:

**Tree Location and Locator Forms:** Over the past several years, we have collected over 350 tree Locator Forms throughout PA and the Mid Atlantic region. This information is available to our coordinators and members upon request from **Sara Fitzsimmons** at: (814) 865-7228 or email:

## BOARD ELECTIONS

Final tallies of votes from the 2003 PA-TACF Board elections were held at the end of last year. Here are your new Board members:

**Tracey Coulter  
Chandis Klinger  
Frank Brouse**

There will be four total positions up for grabs in 2004, including that of PA-TACF Vice-President. We encourage you to start thinking of your nominations now!

More on the Board to come in the June issue of *The Chestnut Tree*.

[sff3@psu.edu](mailto:sff3@psu.edu).

Identification and recording American and Chinese tree locations is the first step to the County CMS Program. For those trees not already cataloged, please send a locator form with leaf and twig sample to **Dave Armstrong** at the PA Chapter. We will immediately advise you of our analysis of the species. Locator forms are available on our web site: [www.patacf.org](http://www.patacf.org) or call Dave at: (717) 632-8669 or email: [darm@blazenet.net](mailto:darm@blazenet.net).

To begin, you will need to locate and verify 2 old ( $\geq 30$  years) and blight-free Chinese trees and 4 pure American trees for hand-pollination. Male catkins from the two Chinese trees, we'll call them A & B, are used to pollinate female burs of two of the American trees, call them C & D. With those trees, you will make two sets of crosses: C x A, and D x B (ladies always listed first). Place 20 protective bags on each set of burs. Bags are available at spring meeting or from Dave Armstrong at the Chapter. Note the location of other two American trees, call them E and F, and harvest 20 or more nuts from each. Keep seed lots separate and label with name of tree and collectors name and phone number.

**Timing of Pollinations:** Placing bags on the burs of the American tree female flowers (burs) prevent unwanted external pollination (planned parenthood). Observation of the American trees, at least twice a week starting in early June, is the key to timing when the burs are ready to bag. Watch for the emergence of the styles at the top of the bur, which is white or yellow. The styles will be fully emerged when it is 3 to 5 mm (about 1/8 inch). It is safe to bag for only 5 days after full emergence. Burs should be approximately size of tip of little finger.

**Bagging, Marking and Recording:** Prepare 20 bags per tree by numbering them 1-20 and randomly select two as check bags. The check bags are marked with a Red X and they are not removed during the pollinating process but used to check for contamination from other pollen (Since you will not be pollinating the flowers in the "check bags", if you find seed in your check bags during harvest, you'll know that some amount of pollen contamination occurred).

With scissors or clippers remove the male flowers (catkins) and leaves around the bur to clear the limb for bagging. Cut

*Figure 1. The male and female flowers of a chestnut tree.*

male & female flowers  
(bisexual catkin)

styles

female flower (bur)

male flowers  
(on a catkin)



off the end of the catkin above where the bur resides (bisexual catkin). Try to leave the terminal leaf and several small leaves to keep the bur healthy and prevent aborted burs. Place the bags over the limb/burs, leaving about 1" head space, and seal with a twist tie or large paper clip. We recommend using a different colored twist tie on the check bags as an extra measure to prevent reopening. Using prepared chapter record sheet, record pollination information (sheets are available on-line at <http://chestnut.cas.psu.edu/forms.html>, or you may request them from Dave Armstrong or Sara Fitzsimmons.

**Pollinating:** In general, pollinate 10 to 12 days after bagging. Remove the freshest (whitest) catkins from the Chinese tree and cut them to 2-3 inch lengths. In a carpenter's apron or a shirt pocket, place a container (a small plastic cup works well) with the catkins. On the American tree, remove the bag and swipe a length of catkin over the style 4 to 5 times. Do not open the two check bags until harvest time. Before moving to another tree, wash your hands thoroughly and clean the pollen container with alcohol. Use second Chinese tree pollen for the second American tree. Enter required info on record sheet.

(SILVICULTURE, continued from page 8)

of a 2.5-foot tall tree shelter began to out-compete similar seedlings grown with a shelter. By age five the difference was significant. This trend mirrors the direct-seeded 5-foot tree shelter study results presented previously. Again, the tree shelters appear to cause a disadvantage to seedlings in height growth after a period of four to five years. There was no height difference between seedlings grown in different container sizes and without shelters at age five, although survival was better with the seedlings that were started in the larger container. Also note that the seedlings started in both the large and small containers, but without tree shelters, were similar in height to 1-0 stock of the previous year (i.e. the same age). In addition, both containerized stock grown without shelters and 1-0 stock were taller than 1-1 stock of the same age. For example, mean height for seedlings started in a large container and grown without a tree shelter at age five (2003) was similar to 1-0 stock of the same age in 2002; both were 6.5 feet tall. 1-1 stock at age five (2001) was 5.3 feet tall. The significance of this requires further testing as climatic factors of varying years were limiting and not excluded from these results.

Again, at TSF, the story changes for differences between containerized stocks due to excessive deer pressure. Sheltered stock started in large containers tended to out-compete

all other treatments, including nursery stock, after the first year, although sheltered seedlings started in small containers began to catch-up by age five, as did a diminished number of 1-0 stock. Note that the heights of the sheltered stock are not much above the height of the shelter itself (2.5 feet). All heights are well below those at SV at similar ages. In addition to the deer pressure, the abundant tulip-poplar crop had rapidly redeveloped and was causing significant competition and shading the already battered chestnut. Some chestnut individuals had escaped the deer browse line and were competing fairly well with the tulip-poplar, typically just behind them in height. These will continue to be observed with great interest.

Overall it appears that transplanting of both nursery and greenhouse containerized stock can be accomplished with great success, especially in the absence or control of deer pressure. These stocks also show that they compete well with surrounding natural regeneration if that regeneration is reset at the time of field planting, and perform better without the aide of 2.5-foot tall tree shelters where deer browsing is not a factor. It appears from this study that 1-0 nursery stock and containerized stock may be more suitable than 1-1 nursery stock for transplanting in terms of cost efficiency and better performance. Additional container sizes and growing media also need to be tested. These trials will continue to be monitored until most trees succumb to blight.

#### **NOTE TO CHESTNUT GROWERS:**

Note to CMS Orchard Growers: Some F1 seed lots may fail completely. Some may produce seedlings that are weak and die in the first year. This is not uncommon when two species or sub-species are crossed. Unsuitable site, lack of adaptation, inadequate cultural practices, extreme weather, etc. may also be responsible. Do not be discouraged by losses of trees. Plant more. If you wish to enter your orchard in the chapter CMS data base, and have not already done so, please report the following information including American and F1 trees planted in 2001, 2002, and/or 2003, for each PA seed lot you received:

For each tree: PA Seed Lot #, tree position, how it was planted (seed vs. seedling), whether it germinated, and when it was recorded dead. Send the information to Sara Fitzsimmons [sff3@psu.edu](mailto:sff3@psu.edu) or contact her at 814-865-7228. Data on the program are available upon request.



### Pollinating Method

by: Ann Leffel

from selected breeding trees – Planned Parenthood. It has been customary for 10% of the bags to be left unopened and unpollinated. These bags are called “checks” or “controls”. If, at the end of the season, they contain nuts, then you know you were too late with bagging and the seed lot has been contaminated with seed produced from unwanted pollen.

When making controlled crosses, glycine bags are placed over emerging female flowers prior to their being receptive to fertilization in order to protect the female from unwanted airborne pollen. When the female is receptive, the bags are removed long enough to hand hybridize the female with the pollen

**Dr. Henry Gerhold**, School of Forest Resources, Penn State U., questioned our checking procedure several years ago. He thought we should apply two types of checks. Control #1 – Unopened Bags – bags not opened and no pollen applied. Control #2 -- Opened Bags – bags removed for length of time it takes to apply pollen, but no pollen applied; bags returned. This check would tell you if contamination occurred during the process of pollination. The chapter has done only #1 while making third backcrosses because we did not want to loose another 10% of potential nuts. Adequate numbers of flowers are hard to come by and we need to produce at least 100 nuts on each American mother tree that we cross.

This past year, I did use both types of checks. It was not a particularly good year to make such a test. The blooming season was late and unpredictable because of the late spring and cool rainy weather. But I went ahead. There are two sets of data. The results are shown in Tables A and B:

Table A. Set A - This table shows the results from the pollination of four American trees, utilizing pollen from one Japanese and three Chinese trees.

♀ Type	♂ Type	POLLINATED		CONTROL 1		CONTROL 2	
		Bags	Seed	Unopened Bags	Seed	Opened Bags	Seed
American	Asian	68	253	8	12	8	9

Table B. Set B - This table shows the results from the pollination of two F1 (American x Chinese) trees, utilizing pollen from nine different American trees.

♀ Type	♂ Type	POLLINATED		CONTROL 1		CONTROL 2	
		Bags	Seed	Unopened Bags	Seed	Opened Bags	Seed
F1	American	148	160	18	0	18	0

### Conclusions:

Set A - Contamination occurred prior to bagging. The bloom was not late; I was!

Set B – There was no seed in either type of control bags. Based on this sample with 148 pollinated bags and 36 check bags, there was no contamination. There does not seem to be a need for the second set of checks. It would be wise to replicate the experiment.

### Public Chestnut and Chestnut Blight Research in the U.S.

- Dr. Bob Leffel

Current Research Information System (CRIS), Cooperative State Research, Education, and Extension Service (CSREES), U.S. Department of Agriculture (USDA) has provided fiscal data and the CRIS Reports on chestnut and chestnut blight research for the past eight years. In FY 1998, CRIS adopted a revised classification scheme. Total funding in thousands of dollars for FY 1995, 1996, and 1997 was 2045, 1544, and 2020, respectively. Appropriations for 1998-2002 are below:

STATES within regions	Funding in thousands of dollars for FY:				
	1998	1999	2000	2001	2002
Michigan	119	90	98	106	127
Nebraska	55	55	58	68	70
Connecticut	154	199	210	214	202
Massachusetts	85	102	195	192	346
New Jersey	208	248	156	115	149
New York	189	193	94	122	143
Pennsylvania	9	10	4	48	152
West Virginia	196	210	192	129	150
Alabama	29	34	17	1	
Georgia	54	70	59	6	
Kentucky	147	132	8		
North Carolina					112
South Carolina	1	5	5	2	
Tennessee	91	60	167	189	218
Texas	220	203	58*		
Virginia	78	55	40	54	53
California			175	208	210
USDA-ARS**	46	58	71	71	
<b>NATIONAL TOTALS</b>	<b>1681</b>	<b>1724</b>	<b>1608</b>	<b>1525</b>	<b>1933</b>

\* The Texas work was relocated to CA in 2000  
 \*\* ARS = Agricultural Research Stations

Field of Science	Funding in thousands of dollars for FY:				
	1998	1999	2000	2001	2002
Biochemistry & Biophysics	10	9			
Physiology	82	90	84	36	29
Cellular Biology			175	208	210
Molecular Biology	233	218	133	87	69
Developmental Biology				32	29
Biology (Whole Systems)	81	131	140	175	241
Ecology	58	63	53	59	106
Genetics	253	218	345	295	410
Virology	18	5	5	2	
Mycology	107	69	6		
Other Microbiology	43	30		44	231
Entomology & Acarology	4	4		5	5
Pathology	748	829	613	529	538
Epidemiology	38	49	51	52	59
Chemistry	8	9			
Economics			2	2	6
<b>National Total</b>	<b>1682</b>	<b>1724</b>	<b>1608</b>	<b>1525</b>	<b>1933</b>



## Selection and Experimentation Within CMS F1 Hybrid/ American Chestnut Orchards

By: Bob Leffel – Chapter Scientist

Cytoplasmic Male Sterility (CMS) occurs in interspecific crosses of chestnut and apparently occurs in American x Chinese (A x C) F1 hybrids, but not in the reciprocal (C x A) F1 hybrids. CMS is a method of genetic emasculation, eliminating the requirements of thousands of controlled crosses for each backcross (BC) generation, when conducted in isolation from all undesired sources of chestnut pollen. Seed production via open-pollination will be greater and much more efficient than with controlled hybridization.

Several CMS orchards were first established in Year 2001 and additions and/or replacement seedlings within each of these orchards were made in 2002 and 2003. Each CMS orchard consists of: (1) putative A x C or A x J (Japanese) F1 hybrids and their reciprocals, and additional A x C F1 hybrids; and (2) putative American chestnut trees established from seed of PA American chestnut trees. Objectives of the studies with these orchards include: (1) the determination of the prevalence of CMS in A x C F1 hybrids vs. C x A reciprocal crosses; and (2) the possible utilization of CMS as methodology for breeding blight-resistant American chestnuts as described previously (Chestnut Tree 7:(2) pg. 7). Sept 2001 and, in more detail, in a Draft of 24 Feb 2003, still under review).

Results in 2003 indicate that some putative A x C F1 hybrids are A's, some C x A F1 hybrids are C's, and some A's are A x C F1 hybrids. Why? Because our methodology in producing the seed lots is subject to possible outcrossing, before or after bagging, i.e., the pollination of trees serving as females by trees other than the assumed male parent. Thus selection for hybridity among F1's and for typical American characteristics among A's should be accomplished ASAP. This should not be difficult. All C x A F1 hybrids, and all male fertile (MF) or partially MF A x C F1 hybrids, if such occur, should be removed from the orchards as soon as MF or male sterility (MS) is identified for each tree. This will require almost daily examination, beginning with the first appearance of catkins and continuously throughout the blooming season. The only trees remaining in the orchard subsequent to culling and selection will be: (1) MS F1 A x C hybrids; and (2) American chestnut trees. Such an orchard in proper isolation will produce the first backcross generation (BC1) seed, (A x C) x A, on the MS F1 A x C trees, via open-pollination (OP). Seed produced on A trees will be the result of intercrosses among A trees only, a germplasm pool of A chestnuts.

When to inoculate and screen for resistance, and what should be the intensity of selection for resistance? Solomon-like advice is needed here! Assuming that our older, blight-resistant C trees differ in genes for resistance and possess adequate resistance to chestnut blight, I suggest no selection for blight resistance until the BC1 generation, and subsequent to selection for MS among BC1 trees. We don't know the number of C genes conditioning MS in A cytoplasm: one to three genes are reported. We can hope for prevalence of one gene control, and eliminate cases of multigenic control? The BC1 tree 'Graves' apparently possesses a single dominant gene when used as pollen parent on A trees: the resulting BC2 trees segregated 1:1 for MS:MF. Control of CMS by a single, dominant gene will be essential for efficiency in the proposed breeding methodology. The planting by PA-DCNR in 2003 of a BC1 population, [(A x C) x A], should provide data for this specific BC1.

Some trees seeded in Year 2001 in CMS orchards produced catkins and/or bisexual flowers in 2003. Selection within, and utilization of, these orchards must begin in 2004. An A x J F1 hybrid tree in 2003 exhibited bisexual flowers only with MS, a female tree to date! The 11 burs produced 19 OP seed, indicating good female fertility. In Year 2003, in the 300-tree orchard established in 2001 (150 F1 A x C and C x A hybrids and 150 PA-A's), 15 A x C F1 trees producing catkins were MS and all C x A trees producing catkins were MF.

Until all culling and selection within an orchard is completed, managers may pollinate selected MS F1 A x C trees with selected A trees within the nursery, providing BC1 populations for study and utilization. Also, managers of all other orchards (BC, American, etc.) should check for MS F1 hybrids, and if present, such MS trees can be backcrossed to A trees for BC1 populations.

Yes, we're operating in the dark – but in the nature of chestnut breeding to date?! The only way to determine the feasibility of CMS as breeding methodology is to attempt to utilize same. The resulting populations will provide a much broader germplasm base for both resistant non-recurrent and American recurrent parents than is currently available.

### Do you have an idea for a newsletter article?

If you have a topic you'd like to see written about, or would like to write an article yourself, simply contact Sara Fitzsimmons by e-mail at [sff3@psu.edu](mailto:sff3@psu.edu) or by phone at (814) 865-7228. Next deadline is May 10.

**PATACF 1995 - 1997  
CHESTNUT  
ORCHARDS REPORT**

*A Review of Inoculated and  
Selected BC3 Orchards*

A DETAILED REPORT WAS  
PREPARED BY BOB  
LEFFEL. A SUMMARY  
FOLLOWS:

The 1996-97 Brogue, Dornsife, and Reels Corner and 1995 Reineman BC Orchards were reviewed by Dr. Fred Hebard, Dave Armstrong, and Bob Lef-fel 19-22 Nov 2002. A detailed report on the review has been prepared. A summary follows:

Subsequent to May 14, 2002 culling, 12 of 92 inoculated

BC3 trees in Br96 and 96A orchards remained. Three of these 12 trees were dug with 4' spade 4 April 02, ball and burlap, moved to Penn State University at Rock Springs Ag Farm, but died. Scion wood collected from 7 of the selected trees produced 19 potted trees grafted on American rootstock were planted at Rock Springs November 12, 2002 but failed to survive (Miller). One tree of the 9 remaining produced male and female bloom in 2002 and six of these trees produced female and male flowers in 2003. The only tree rated "excellent" for all characters on November 29, 2002 appears to be dying. The remaining trees are vigorous and growing well despite the blight, exhibiting moderate resistance as expected.

Of the 203 inoculated BC3 trees in Br97 orchard, 15 trees currently survived 2001 inoculation and culling. All BC3 trees remaining in the three orchards are possibly adequate in resistance rating. Final selections should be on basis of American characteristics – and survival! Dr. Hebard recommends evaluation of resistance 1 year after inoculation as most accurate, but six of those BC3 trees in Br97 orchard have died since selection in spring 2002 as "keepers".

At Dornsife, 63 BC3 trees were inoculated in Do96 orchard and 5 were selected 19 Nov 2002 for retention. Of the 112 inoculated trees in Do97 orchard, 8 were selected for retention.

At Reels Corner, 49 BC3 trees were inoculated in RC97 orchard 9 June 2002 and scored by Hebard in November. A severe ice storm in October had previously destroyed many trees in this orchard. Tom Pugel re-evaluated the survivors in May 2003 and did a preliminary selection, keeping 19 trees in hopes that some of them recover before making any further selections.

Year Seeded	BC3 Cross Code	Orchard			Total
		Brogue	Dornsife	Reels Corner	
1996	CrYo x CL 287	1			1
1996	OrYo x CL 287	8	5		13
1997	OrYo x GR 226	6			6
1997	DoNo-1,2 x CL 53	2	1		3
1997	JoSc x GR 210	7	4	16	27
1997	RcSo-m x GR 210		3	3	6
<b>Grand Total:</b>		24	13	19	56

**F E N C I N G**

Through the efforts of PA Chapter member and orchard grower **Mike Shanshala** of Warren, PA, we now have 4 ft woven wire fencing available to our growers.

For details and more information contact:  
Tim Phelps  
Ph: (814) 865-7228  
E-mail: phelpst@psu.edu

Dr. Hebard recommended open-pollination among selected trees, attempting to have the same number of trees per line, if possible. This would require elimination or emasculation (removal of catkins) of all but the selected trees. But all orchards herein have other chestnut plantings to be retained, and there are nearby chestnut trees as well, so controlled pollination by hand will be required.

The three orchards have several BC3 lines in common. Table one below shows the selected BC3 trees in the 3 orchards as of August, 2003.

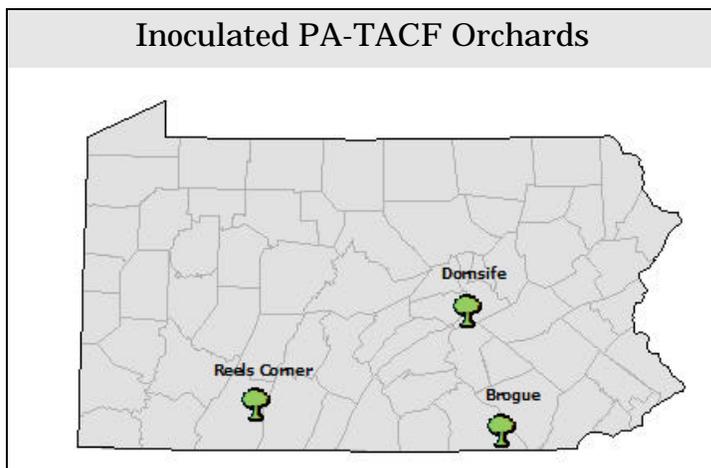
**The Next Step: BC3F2 Generation**

By controlled pollination, PA-TACF produced 331 BC3F2 (2 single crosses) seed utilizing 4 trees in Br96 and 6 trees in Br97 orchard in 2002, avoiding sib-matings. Year 2003 single crosses total 376 bags plus 42 check bags among the flowering selected trees remaining in the three orchards (Table II).

Genetic Lines Crossed	2002	2003
(OrYo x CL287) x (JoSc x GR 210)	234	489
(OrYo x CL287) x (RcSo x GR 210)	0	20
(OrYo x GR226) x (CrYo x CL287)	97	0
<i>Total</i>	331	509

Table II. Amount of BC3F2 seed produced by PA-TACF

The BC3F2 seed produced by PA-TACF is planted in the Arboretum at Penn State University (University Park, PA).





## Chestnut Silviculture Studies, part II

By Tim Phelps and Kim Steiner  
Penn State University

In the previous issue of the Chestnut Tree, results were shared from a study investigating early height establishment of American chestnut trees grown from seed and administered three treatments; vented and unvented five-foot tree shelters and no tree shelters. It was generally concluded that in the absence of deer pressure five-foot tree shelters did not provide a distinct advantage in early height growth beyond four years of age. In fact, trees grown without the aide of a tree shelter were significantly taller after the fifth and sixth growing seasons in this environment. Of course, in areas where deer pressure is significant, the protection of trees is essential until they reach above the deer browse line (a height that may forever and always be disputed). However, according to these results it may be better to use another means of protection if it is more efficient and affordable (eg. fencing). Please keep in mind these results pertain to forest plantings and do not necessarily justify management decisions for backcross plantations.

In this issue, focus shifts to alternate planting methods other than direct seeding. While we acknowledge that direct seeding will likely be the most efficient and cost effective planting method for reestablishing chestnut into the central Appalachian landscape, many growers may prefer to use established seedlings for transplanting into their woods to insure higher survival rates per seed and have more control in plant placement. Transplanted seedlings may also be competitively superior against the understory vegetation often encountered on forest sites. It will also likely be that the first several batches of blight resistant American chestnut seed will be small, and we will not want those precious seeds to go to the squirrels!

Nurseries that grow various types of trees in outdoor field beds have been the predominant producers of hardwood seedlings for reforestation projects. Seedlings are generally grown in a seedbed for one, two, or three years and then transplanted in the field or put in a transplant bed at the nursery for an additional one to two years. The age of “nursery stock” can be identified by the number of years grown in the seedbed and transplant bed by its two digit code, where the first number relates to the number of years it was grown in the seedbed, and the second number relates to the number of years it was grown in the transplant bed. For example a 1-2 nursery stock seedling would have been grown in a seedbed for one year

followed by a transplant bed for an additional two years making it a three-year-old seedling at the time of planting in the field where it is to remain for the rest of its life.

The greenhouse industry has long been a producer of conifer seedlings, but new technologies are beginning to catch on for the hardwood market as well. Advancements for containerized greenhouse-grown seedlings are largely centered on growing media and container features. A future segment will look at some of these differences in more detail as they pertain to American chestnut propagation.

Trials for the nursery/containerized stock study were established at the Stone Valley (SV) and Tuscarora State Forest (TSF) sites adjacent to the previously mentioned direct seed tree shelter studies to compare the growth of seedlings grown in two sizes of containers raised in a greenhouse for three months (mid-February thru mid-May) versus older transplants originating from two nursery stocks that were grown at nearby DCNR Penn Nursery. The large and small containers used in the greenhouse measured 10 inches in length by 2.5 inches in diameter (40 cu.in.) and 8.25 inches in length by 1.5 inches in diameter (10 cu.in.) respectively. One hundred of each type of containerized stock was planted at each site, half of which also had a 2.5-foot tall shelter treatment for limited deer protection. The nursery stock consisted of 20 each of 1-0 and 1-1 stock at each site. Each site was established in a completely randomized design.

The SV and TSF sites differed in harvest treatment, levels of competing vegetation, and deer pressure, as well as in other possibly relevant respects. The SV site was within a shelterwood cut prior to the 1997 tree shelter trial. The competing vegetation consisted of oak, maple, and birch seedlings (~ 2 ft. tall), and *Rubus* species (blackberry and raspberry). A six-strand electrified fence had been installed but not maintained as it became obvious that deer pressure was very low.

In contrast, the TSF site was situated in an older clearcut (harvested early 1990s) where natural regeneration was slow to get going likely due to heavy deer pressure despite an eight-strand electric fence around the perimeter of the harvest area. In fact, the TSF district forester said that deer hunters actually preferred to hunt inside the fence than outside. Competing vegetation at this site consisted predominantly of a very abundant crop of 2 – 3 ft. tall tulip-poplar seedlings.

(See *SILVICULTURE*, continued on page 8)

Table 1. Survival (n) and mean height (feet) of the 1999 containerized/nursery stock study at Stone Valley Experimental Forest. Means with different letters within a column are statistically different at the 0.05 level.

Stone Valley Treatment	1999		2000		2001		2002		2003	
	n	mean	n	mean	n	mean	n	mean	n	mean
1-1 stock	19	1.6 a	19	2.8 a	19	5.3 a	19	7.3 a	17	9.4 a
1-0 stock	19	1.3 b	18	2.5 a,b	17	4.6 b	18	6.5 a	18	8.2 a
Large w/ shelter	49	0.6 c	47	2.2 b,c	47	3.2 c	44	4.0 b,c	38	4.6 c,d
Large w/o shelter	49	0.6 c,d	47	1.9 c,d	47	3.5 c	47	4.9 b	43	6.5 b
Small w/ shelter	48	0.5 d	47	1.7 d	46	2.8 c	45	3.6 c	32	3.9 d
Small w/o shelter	49	0.5 d	45	1.6 d	40	3.3 c	39	4.7 b	39	5.9 b,c

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*(SILVICULTURE,  
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The trials for each site were established in early to mid-May of 1999. Existing vegetation was cleared to the ground with a circular saw mounted on a weed eater in an effort to reset competition. An auger with a 6-inch bit was used to plant the seedlings. Measurements were recorded at the end of each growing season to present.

Tables 1 and 2 show the results through age five for the seedlings at SV and TSF respectively in terms of survival (n) and mean height (mean). Based on fairly consistent survival rates, it appears that all planting stock transplanted well with better than 90% survival across all treatments and sites after two years in the field. More individuals began to die-out after the third year as other limiting factors (eg. deer pressure, competing vegetation) became increasingly severe.

As expected, heights of the older 1-1 and 1-0 nursery stock were significantly greater than the containerized seedlings after the first year in the field. This trend continued through age five at SV where there was no deer pressure. Also, there was no difference between 1-1 and 1-0 nursery stock after age three which would indicate that the

extra year at the nursery is not necessary. So far, the extra resources of these stocks (i.e. more developed root/shoot systems) are still providing a height advantage to the nursery-produced seedlings compared to the younger containerized material.

At TSF the story is vastly different due to significant deer pressure. While the nursery stock held height advantages over containerized material after the first growing season, 1-1 and 1-0 stock exhibited only a very meager rate of growth in following years. Because these seedlings were unprotected by shelters, the deer continually kept these trees from growing, and it appears that many of the trees could no longer survive after three years of abuse as indicated by a marked decrease in survival (n) for 2002.

Containerized stock at SV was fairly consistent in height growth across treatments through 2001. At age four the seedlings grown without the aide (see SILVICULTURE, cont. on page 3)

Table 2. Survival (n) and mean height (feet) of the 1999 containerized/nursery stock study at Tuscarora State Forest. Means with different letters within a column are statistically different at the 0.05 level.

Tuscarora Treatment	1999		2000		2001		2002		2003	
	n	mean	n	mean	n	mean	n	mean	n	mean
1-1 stock	20	1.4 a	19	1.8 b	20	1.9 b,c	11	1.7 c	9	1.7 b,c
1-0 stock	18	1.0 b	18	1.7 b	17	2.3 b	11	2.5 b	8	3.4 a
Large w/ shelter	50	0.8 c	48	2.6 a	49	3.0 a	40	3.3 a	34	3.7 a
Large w/o shelter	50	0.7 c,d	50	1.3 c	45	1.5 c	31	1.5 c	25	1.8 b,c
Small w/ shelter	49	0.6 d,e	48	1.8 b	47	2.1 b	38	2.5 b	29	2.8 a,b
Small w/o shelter	48	0.5 e	48	1.2 c	39	1.5 c	29	1.4 c	16	1.5 c