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Editor's Note

Chestnut trees once accounted for more than 25% of the hardwoods in the Appalachians. Because there must have been millions of self-seeded wild trees well suited to the various ecological conditions of the region, one might think that the fundamental things needed to breed new blight-resistant varieties - such as pollination and growing the trees from seed would be virtually foolproof.

If the letters we receive are any indication, members of the American Chestnut Foundation are an intelligent, well-read and industrious group who, like Dr. Charles Maynard ("Chestnut pollen collection and handling," p. 101) and your editor, have found a hundred and one ways to kill pollen seeds, and little trees. Even Patrick Chamberlain ("A practical way for the layman to participate in breeding resistance into the American chestnut," p. 76) has lost some of his carefully nurtured seeds to mice and rabbits.

This issue focuses on the basics of the breeding program. We have added a special section with articles on pollination and planting, addressing rnany of the problems readers have reported. Background articles are to be found throughout the 'Notes" and "Memories" sections.

The backbreeding program is only one of several ways the American Chestnut Foundation and chestnut workers throughout the world are rnaking progress in the battle against *Endothia parasitica.* The next issue of this *Journal* will include an overview of developments in these various scientific fronts.

Letters to the Editor are always welcome. Your suggestions, thoughts, complaints, questions, disagreements, pointers, and praise are all a part of the ongoing process "Toward the Restoration of an American Classic." Share them!

The *Journal of the American Chestnut Foundation*
seeks volunteers to participate in the editing process.
If you have access to a Macintosh computer and
some editorial experience, please drop the Editor a note.

President's Message

April 1991

Philip A. Rutter

I hope you like the changes The American Chestnut Foundation is going through. We are growing - as we must if we are to provide a secure future for our work.

At the moment, the growth is a little painful. Having decided it was realty necessary for the Foundation to make the stretch and hire an Executive Director, we ran smack into a recession. The charitable organizations that fund projects like ours are suddenly experiencing an increased demand for their help. Just when we needed to expand our budget, the money has become harder to find.

We are having to work overtime just to keep afloat. Consequenily, when our editor called for me to submit my President's Message for this issue of the *Journal,* I found myself struggling to find the time.

So I decided to take some "politically correct" action, and "recycle" something I wrote for a different audience. While it talks about many of the familiar reasons for bringing back the chestnut, I think you may find a few that are less commonly discussed. Yes, I love our tree. But there are even deeper reasons for our work.

The following essay was one I was invited to write for the *Newsletter* of the Natural Resources Council of America. The NRCA is an "umbrella" group that every conservation organization you can think of (literally!) belongs to. It provides a way for the executives of these groups to communicate directly with one another. For most of those folks, this was the first time they had heard of the ACF. I hope you like it. I'd be glad to have you write to me with your reactions to it.

We *Can* Restore The American Chestnut
Editorial, *Newsletter* of the Natural Resources Council of America, Jan., 1989)

Imagine the furor if an explorer, poking into a sheltered but remote bay in southern Greenland, discovered a small but healthy population of the long-extinct Great Auk. International headlines would surely result, and dozens of meetings of government commissions ond censervalion organizations, as the world mobilized to protect, preserve, and eventually restore this fascinating bird. It is hard to conceive any other response - we *could* never ignore such a species in need. How is it, then, that the American chestnut tree, once the most productive component of the eastern hardwood forests, has been an orphan for decades, and pro)ects to restore it are still unacknowledged and desperately underfunded?

The American Chestnut Foundation is a new organization created to deal with *on* old disaster. The great chestnut blight of the 1920s and '30s was possibly the most destructive forest disease episode in history, and was certainly one of the formative influences for the entire conservation movement. But much of the story has now been forgotten, and the species has long been considered beyond all hope of saving. It is always dangerous to *forget* history, and doubly So in this case: not only is there a real danger of repeating past mistakes, but an opportunity to remedy old problems with new solutions could all too easily be ignored. The chestnut, however, was simply too important a species for us to miss any chance that it might be restored. Our Foundation intends to see that the full restoration of the tree to its place in the forest communities becomes a reality-

The American chestnut tree was destroyed by a human-imported fungus disease. Prior to the epidemic this tree was probably the single most important tree in the eastern hardwood forests, both from the standpoint of human economics, and in regard to its keystone role in the hardwood ecosystem. Its natural range covered some 200 million acres, and in the Appalachian regions from New England to northern Georgia it is estimated that a full quarter of all the hardwood trees in those vast forests were chestnuts.

Its unique importance to the ecosystem, including humans, lay both in the fact that it grew desirable wood 30-50% faster than oaks (to which it is related), and in that it alone of the forest trees produced generally reliable crops of nuts. Virtually everything in the forests ate those nuts, and the availability of a large, dependable food source in late fall may have accounted for much of the abundance of game

reported by early explorers and settlers. Imagine a forest where a quarter of the trees could he counted on to produce one to three bushels of nuts every fall; and then imagine the impact on the entire ecosystem.

Biologically the loss of those trees was staggering, and there is evidence that the chestnut tree is not ecologically replaceable by the many species which now grow in its place. Economically, a similar loss might be felt if Douglas fir were to suddenly disappear. The extent of the damage was realized at the time, and disasters of this magnitude do not go unaddressed. The eager young men recruited into the infant Forest Service by Gifford Pinchot tackled the problem vigorously, but without effect. The fungus killed more than 99.99% of the chestnut population; an extraordinarily efficient pathogen. Billions of trees died; the blight was far more destructive than the currently familiar Dutch elm disease.

By the 1950s, American chestnut was reduced from a dominant species of giant forest trees to a residual population of understory shrubs, sprouting from the stumps of trees killed by the blight. At about the same time, government research on the species was halted as a waste of money. The chestnut joined the list of species exterminated by human folly - it has now long been considered an issue as dead as the passenger pigeons that once fed on it.

The chestnut, however. is far from extinct.

The stump sprouts, as a part of the unusual biology of the tree, continue to survive. They now go through cycles of growth and renewed epidemic, and since the blight has now so much less to feed on, and the spore population is consequently so much less than it was during the pandemic, it is common for trees to survive for as many as 25 years before the blight knocks them down once more. Only rarely do new seedlings get started, however, and the natural attrition of the years continues to erode the remaining sprout populations. Extinction is not here, but is a definite possibility in the foreseeable future.

The great hope stems from the fact that in the past few years several new approaches to the possible restoration of the species have been suggested, ranging from biological control of the fungus to breeding resistant trees using modem techniques to biotechnological manipulation. Virtually all scientists intimate with the tree and the fungus are highly optimistic that one or a combination of these methods could actually result in a complete recovery of the chestnut, restoring one of this continent's most productive natural resources. Govemmental and business interest in funding the necessary research, however has been slight.

It is not hard to understand why. Government funding of forest research has been decreasing recently, and with a shrinking pie, it is easy to see that administrators would not be eager to try to cut yet another piece from it, particularly not for a project with a history of failure. Business has perhaps less excuse, and yet it is not difficult to guess that a project with no probable pay-back for at least 30 years would not be very attractive in the absence of compelling reasons to undertake it.

Perhaps it is just as well. The history of governmental research on the chestnut has been one of repeated fits and starts: individual researchers have maintained their research programs by force of personality, but when they retired, years of work were lost when a successor's true interests lay elsewhere. In the worst cases, research plantings have actually been destroyed to make way for other projects. Realistically, research on forest trees must be able to count on expanses of time well in excess of the average scientist's career. Given the predictable changes of priorities the political process generates, we must consider the possibility that research on forests and forest trees, in institutions which rely on the contemporary political administration for their funding, will always be in some jeopardy.

The American Chestnut Foundation was created with these thoughts in mind. In order to supply the continuity of research necessary to work with a species with a 400 year life span, we have as our sole task the pursuit of projects that can lead or contribute to the restoration of the chestnut. We will not be sidetracked, nor will we quit. No forest disease problem is insolvable, unless abandoned.

We have the support of the national scientific community involved in chestnut research. We also seek public members from the entire nation, since the problem is one that does not respect state lines.

So far, however, we are badly underfunded by the standards of most national organizations. Our consultants tell us that the reason, in part, is that our project is slightly "different." It covers too large a region to immediately catch the eye of local philanthropists. The effects of the restoration would also be so large that they are not easily categorized as conservation, preservation of biological diver-sity, or economic development, a siwation which can leave potential donors uncertain about whether we fit within their "guidelines."

The sweeping impact of restoring such a key species, however, is precisely why we are determined to succeed. This is not, and was never conceived as, a "single species" project. What makes the restoration worth attempting is the very fact that the health of the entire ecosystem, humans included, could be strongly bolstered by this uniquely preductive component.

There are several reasons why our project should be of special interest to the conservation community.

First, there is the fact that the tree *is* a "nature," "conservation," "preservation," and human "economic" project. The tree was a mainstay for game species; it also supported several specialized insect species, some of which have not been seen have not been since the blight. And it was the economic backbone of the Appalachians. This one tree brings all these concerns together in a particularly clear fashion - it can help make it plain, to those who still do not understand, that these concerns which sometimes seem to be at odds with one another are deeply interconnected. When the forests die, it isn't just the wildlife that suffers, nor just the tender-hearted. We humans have a cold, hard, cash interest in seeing that our ecosystems stay healthy. When the tree was hurt, so were we; and badly. As a tcol to teach that lesson, the American chestnut is unexcelled.

Second, the chestnut focuses many high visibility environmental concerns in one species. Reforestation to counteract the greenhouse effect is an idea which has captured the public interest- what better species to plant than this biological powerhouse? But forests everywhere are declining; could trees withstand the already degraded environment well enough to make a real impact on atmospheric Cq? While the proper answer to acid rain is to make the rain less acid, not find species that will tolerate poor conditions, in the meantime, chestnut does tolerate highly acid soils much better than many other species. Also, chestnut trees appear to be among the survivors of the gypsy moth infes-tations. Perhaps this tree stands some chance of withstanding these threats to forests, when others are failing.

Third, what possible better justification for the environmental conanunity's concern for the preservation of biological diversity could there be than the restoration to productivity of such a key species? Until recently we have not appreciated the unique roles the chestnut played in the Appalachian forests. Only now, 60 years after the blight, can we see clearly that the productivity and health of the entire ecosystem have suffered irremediable harm. Luckily, with this sPecies, restoration is still a possibility; but for thousands of other species there will be no restoration from extinction, and no chance to reverse unrecognized damage.

Fourth, there is the hope that with the success of a project like restoring the American chestnut, we may begin to form a new habit- one of planning for the long term. Even in national policy making, habits are crucial. The first action of any administrator faced with a new problem is to see how similar difficulties have been handled in the past. As we are all well aware, far too often these days temporary patches are put on problems crying for real, long term resolutions. If temporary patches are the only examples available, then we are likely to see the pattern repeated. Restoring the American chestnut cannot be accomplished in our own life times. We hope and expect to make recovery inevitable, but the forests cannot resemble the originals in less than 300 years. We are no less determined to do it. Perhaps this may make it ever so slightly easier in the future to plan in terms of 150 and 200 years. With practice, maybe we could make it a habit.

And lastly, it looks like this is one we can win. The disease is not incurable, and the tree is a fast grower and vigorous colonizer of existing forests. If we can provide a leg up, the tree may carry out the restoration on its own. As rainforests and species continue to disappear; while groundwaters grow more foul; when even the once limitless oceans and atmosphere show unmistakable signs of damage, the chestnut blight may be a disaster that can be put right. That may be critical.

Hopelessness is one of our greatest enemies. If the majority of mankind comes to the conclusion that we are powerless to alter the apparent downward spiral of our biosphere, support for the difficult and endless tasks necessary to preserve the health of the world may waver. There is no substitute for the *will* to put things right, and to keep them that way. And there is no better brace for our collective will than a big win; something to demonstrate that we can, really can, make a difference. Just possibly, restoring the American chestnut might be that big win. But it is clear that we are going to need the support of the conservation community to make the restoration, and the win, a reality.

ACF Hires Executive Director

Your Foundation has recently hired John Herrington, of Bennington, Vermont, as executive director. Herrington comes to the Foundation with a strong background in forestry communications and non-profit association management.

Prior to joining ACF, Herrington served for seven years as regional manager for the American Forest Council, consecutively managing the New England, Mid-Atlantic and North Central regional offices. He was responsible for overseeing the implementation of the American Tree Farm System and the Forestry Council's internationally acclaimed environmental education program, Project Learning Tree, in twenty east and midwest states. Herrington is familiar with the forestry and environmental community in this large region, knows the media, and has experience orchestrating the revitalization of national programs at the local level.

In the mid 197Os and early '80s Herrington spent ten years as executive director of the New Hampshire Timberland Owners Association. Hired as that organization's first professional staffer, he built a fledgling nonprofit into a strong, viable entity.

For many years John Herrington has wanted to be instrumental in conserving and protecting natural resources. He says that he joined the American Chestnut Foundation for several reasons: because of the challenge; the noble goal of bringing something important back to the environment for future generations; the professionalism of the research scientists; and the commitment of the people involved.

John Herrington can be reached at ACF's executive office in Vermont: P.O. Box 4044, Bennington, VT 05201; telephone (802-447-0110). The Foundation's member services office will rernain in West Virginia: P.O. Box 6057, 401 Brooks Hall, West Virginia University', Morgantown, WV 26506.

NOTES

Chestnut Loses Two Friends in Michigan

Dennis W. Fuibright
Michigan State University

Two key players in the story to bring back the American chestnut tree recently passed away in Michigan. Both Jim Comp and George Unger played key roles in bringing good news about the American chestnut in Michigan to scientists and the American public.

Comp was 90 years old and according to his son Jack, "lived a productive life very few of us could match. He had an unrivaled work ethic and was tireless in pursuit of the things he believed in." Lucky for us, Jim knew the value of chestnut. He was born in Maryland just before the blight struck and at an early age moved to West Virginia. Blight started killing the trees in West Virginia and as he moved through Ohio and on to Michigan the devastation brought on by the blight followed him. In the mid 1970s, Jim found unpollinated American chestnut trees in Cadillac, Michigan, and wanted them to produce chestnut. He began looking for American chestnut trees producing nuts in northern Michigan so that he could plant seedlings near the non-bearing trees to enhance their chance of pollinization. The scheme worked and two years ago the lone chestnut trees began producing nuts. In the meantime, his nut collecting forays got larger with more and more volunteers helping. Volunteers consisted of other retirees, Consumers Power Company, Boy Scouts and the local soil and water conservation district. This loose group of volunteers recently became a nonprofit organization called the Nortwest Michigan Chestnut Council. It still collects and distributes thousands of American chestnut seedlings each year. These seedlings have been sent to individuals in nearly every state (except those with quarantines) and scientists working on chestnut. On page 9Ian excerpt from a talk given by Jim Comp at an American chestnut meeting held in Michigan in 1981.

George Unger played a different role in the chestnut story. Having emigrated from Austria with his family as a young boy, George lived most of his life on a 104-acre farm in Grand Haven, Michigan. The farm must have been difficult to work since the soil is nearly pure sand from the dunes along Lake Michigan. The homestead was blessed with an American chestnut orchard which consisted of about 50 trees. The canopy of the orchard was Sc) thick that George told the story of a pheasant that could not escape by flying through it. He also told of gathering the nuts and shipping them to market in Chicago and later locally in Grand Haven George was very proud of the trees and was saddened when they became blighted in the 1940s. "All the experts told me to cut them for firnwood because they would all die anyway, he told me. And he did start cutting them. He noticed a curious thing in the 1 960s. The bark on the trees started "exploding out." Even though the trees looked bad they never died. The power company came to remove some of the trees from the right-of-way that crossed his land. He remembered begging them not to cut the trees because "they were fighting so hard they're fighters." They didn't cut the trees that day or any day since.

What George Unger saw and do& umented was the biological control of chestnut blight called hypovirulence. These were among the first trees in North America to develop natural biocontrol. While it took sometime for scientists and nature enthusiasts to begin to take him seriously, meny chestnut scientists have visited the Unger's location and had their pictures taken with Mm under his large surviving trees. I asked him how many of the original 50 trees actually died of blight. He said that only one died of blight; the others he had cut down for fire wood before he noticed they were recovering. Some of the stumps have sprouted and are now becoming trees.

The last few years were difficult for George 'with land tied up in probate court. The land has now been sold to a developer and, so far, has been denied protection by the state of Michigan because the trees were planted instead of naturally seeded.

American Chestnut Foundation 1990 Nut Harvest

Frederick V. Hebard, Philip A. Rutter, Mark Widriechner, Larry Inman

First, we would like to thank Sandra Anagnostakis of the Connecticut Agriculture Experiment Station for providing access to flowering hybrids; without her assistance, our harvest would have been quite meager. We would also like to thank the White Memorial Foundation of Litchfield, Connecticut, for providing access to flowering hybrids.

Last year, the ACF had its largest nut harvest to date. In this paper, we present a summary of the harvest broken down into the various crosses we made (Table 1) and the yield per bur and bag pollinated (Table 2). This tatter information should be useful for planning crosses in the future. In order to explain why we made certain crosses, it will be helpful to review our current strategy for breeding blight-resistant chestnut trees, which is to use the backcross method to transfer the blight resistance of Chinese chestnut into American chestnut.

The backcross method for chestnut entails crossing American and Chinese chestnut to obtain hybrid frees which are 1/2 American and 1/2 Chinese chestnut (F1s). The hybrids are then backcrossed to American chestnut to produce trees which are 3/4 American, 1/4 Chinese (B1s). Those B1 trees which test as most blight resistant are selected for a second cycle of back-crossing and selecting, to produce trees which are 7/8 American, 1/8 Chinese chestnut (B2s). A third cycle produces frees which are 15/16 American (B3s). The last step is to intercross the B3s to produce lines which breed time for high blight resistance (B3-F2s). Selected B3-F2 trees will produce seed for reforestation. Based on experience with other crops, the 15/16 American chestnut trees are expected to be indistinguishable from pure American chestnut, except for their blight resistance.

Our main objective is to advance the breeding as rapidly as possible. Currently. we have large numbers of B2 (7/8 American) trees growing, and we are producing more in additional American chestnut backgrounds (Table 1). Our goal is to produce a population of American chestnut trees for reforestation. We cannot reforest the Appalachians with only one tree. We will produce blight-resistant trees in at least twenty separate American chestnut backgrounds. That is one reason we continue to produce F1s and B1s (Table 1). The F1s also are used as controls in screening the various backcrosses for blight resistance.

First hybrids and backcrosses are intermediate in blight resistance between Chinese and American chestnut. Previous chestnut breeders believed they had to cross hybrids a second rime with Chinese or Japanese chestnut to obtain trees with high levels of blight resistance. In contrast, we believe we merely have to intercross

A Quick Guide to
Chestnut Breeding Terminology
The Editor with help from Fred Hebard

American x Chinese	- Fl
F1 x F1-	F2
F2 x F2-	F3
American x F1-	B1
American x B1-	B2
B1 x Fl-	intercrossed B1

the hybrids or backcrosses to recover highly blight-resistant trees. We believe this because current

evidence indicates that resistance is controlled by two pairs of genes. Thus a second objective of our research is to confirm how many genes control blight resistance. The F2 progeny in Table 1 were produced to achieve this second objective; they also will demonstrate that intercrossing of hybrids and backcrosses yields highly blight-resistant trees. Once we know for sure how many gene pairs control resistance, we also will know how many progeny are needed for each type of cross.

As indicated previously, Table 2 presents data on the number of nuts produced per pollinated bur and bag in 1990. These figures should serve as rough guides in anticipating future yields while bagging flowers for controlled pollinations. In general, about one nut was obtained per two burs pollinated, while a bit more than one nut was obtained per bag. The number of nuts obtained per bag varied, of course, with the number of burs enclosed in each bag. The pollinations in Iowa were done with dried pollen, while those in the east were done with fresh catkins. There was little difference between Iowa and the east in the yield per pollinated bur, although the highest yields were obtained with fresh catkins. This suggests, in the absence of data from controlled experiments, that pollination with fresh catkins is the best method, but good yields can he obtained with dried pollen.

Now, if we can get trees from 75% of these nuts, we will be doing quite well!

Table 1
Number of nuts harvested and number of
American chestnut backgrounds for controlled pollinations
by the Americen Chestnut Foundation in 1990.

Cross	Number of Nuts	Number of Backgrounds
American x [(Chinese x American) x American]. B2	449	9
American x (Chinese x American), B1	132	2
Chinese x American and *vice versa,* Fl	561	8
(Chinese x American) x (Chinese x American). F2	299	*
[(Chinese x American) x American] x [(Chinese x American) x American]. B1-F2	485	*
Total	1926	

*Not relevant

Table 2

Nuts produced per bur and per bag for controlled pollinations at different locations by the American Chestnut Foundation in 1990

Location	Nuts	Nuts' per Bur	Nuts per Bag	Pangs	Range
Virginia mountains	366	0.5	0.0-1.4	1.0	0.2-2.0
Virginia orchard	151	0.6	0.3-1.5	1.3	0.6-3.9
Connecticut	784	1.5	1.1-1.9	2.1	1.3-3.7
Northeastern Iowa	577	0.5	0.2-1.1	1.4	0.7-2.1
Central Iowa	41	0.3	0.2-0.5	1.0	0.8-1.2
Total**	1919	0.7	0.0-1.9	1.5	0.2-3.7

*Burs were counted at the time of harvest in Virginia and Connecticut but counted at the time of pollination in Iowa. Fresh catkins were used to pollinate in Virginia and Connecticut, while dried pollen was used in Iowa.

**The total harvest for Table 2 differs from that for Table 1 because additional pollinations lacking bar and bag counts are included in Table 1

NOTES

1990 Rutgers Chestnut Breeding Report

John E. Kuser
Cook College, Dept. of Environmental Resources
State College of New Jersey

Last year's 58 American x Chinese nuts produced nine seedlings. Chipmunks destroyed three, and the remaining six were overwintered in a chipmunk-proof wire cage. These seedlings appear intermediate between their parents, having twigs that are reddish-brown like those of American chestnut, but slightly fuzzy like those of Chinese. They are also intermediate in ease of culture here, less vigorous than Chinese but not as prone to chlorosis and sunscald as American.

This year we again used the lOm American chestnut at New Egypt as female parent, after cutting off two blighted branches and curing a trunk canker by mudpack treatment. We pollinated 136 burs, using pollen of either the tall Chinese chestnut (TC) at the Turf Farm, or Dunstan chestnut (D), or Heritage (H). This produced a total of 61 filled nuts: 23 American x TC; 32 American x D, and 6 American x H. We also picked 117 open-pollinated burs which yielded 126 filled nuts. We were able to reach the top of the tree with the help of a new 14 foot stepladder and a long pruning pole.

The hybrid nuts, along with 210 open-pollinated American and 114 openpollinateel TC, have been planted in Leach cells and put in the 34° F coldroom where they stayed until February.

We are beginning to work on strategies to induce early flowering of the hybrids so that they can be hackcrossed to American. At the same time, we need to develop a blight-resistance test capable of identifying resistant clones in less than the several years necessary to field plant and screen them. Dr. Peter Uedker is working on a test.

Plans for 1991 include a reciprocal cross of TC x American, in case we have any problem (such as male sterility) with American x TC; and possible use of an additional American female parent to see if it produces nuts with a higher germination rate than those from the New Egypt tree.

Meadowview Notes

Frederick V. Hebard, Superintendent

Nineteen-ninety was another good year at the Wagner Research Farm in Meadowview, Virginia. Rainfall again was abundant, giving good growth to the trees.

Our 1989 plantings did well, but the survival of grafts over the winter of '89-'90 was disappointing. Details are shown in Table 1. A main cause of graft failure may have been lack of nitrogen. Leaves began showing chlorosis in July of 1989; foliar analysis results implicated nitrogen. The standard recommendation for transplants is little or no fertilization the first year, but our experience indicates that transplants at the Wagner Fann need more fertilization.

Survival in the remainder of the orchards planted in 1989 (Table 1) was good except for the KY-Iowa B1s and the Test Plots. The Kentucky plants had been inoculated with the blight fungus in the greenhouse in Kentucky before trans-planting. An unexpected soil-borne phase of blight developed in the transplants, and has killed many of them. We did not expect blight to be a problem because it normally does not attack the roots of chestnut trees.

The Test Plots were started from seed of Chinese and American chestnut. Rodent predation was a main cause of mortality, and we have taken steps to alleviate this in our 1990 plantings.

Emergence of nuts planted in 1990 averaged 54%. Among seedlots, emergence ranged from 92% to 0%. The details are in Table 2. The seedlot with 0% emergence was a first hybrid of Chinese and American chestnut. In general, most seed lots of first hybrids showed poor emergence. In 1991, we will test various methods of starting first hybrids and compare their emergence to that of nuts of pure species.

Seedlings from nuts planted in 1990 averaged 17 inches in height, ranging from 5 to 41 inches. Seedlings of nuts planted in 1989 averaged 25 inches in height, ranging from 9 to 37 inches. Trees transplanted in 1989 averaged 42 inches in height, ranging from 14 to 76 inches.

Our pollination results for 1990 are given in detail elsewhere in this issue. The ACF obtained 1926 nuts from controlled pollinations in 1990. That is 607 more than the combined annual average of the old USDA and Connecticut Agricultural Experiment Station programs! We are beginning to get this breeding program off the ground. I hope you are encouraged to continue supporting our efforts. I would like to thank Sandra Anagnostakis of the Connecticut Agriculture Experiment Station and the White Memorial Foundation of Litchfield Connecticut for providing access to flowering hybrids.

Many people who responded to our Foundation's membership survey indicated an interest in helping out with the research. There are several ways to do this, including growing chestnuts on your own, coming down to work on the farm, and helping organize a core of volunteers in a state chapter of the ACF to run and staff a chestnut breeding program.

Eventually, we hope to replicate the Meadowview breeding program every few hundred miles along the Appalachians from Maine to Georgia and west to Michigan and Mississippi. The reason for this is that American chestnut trees from the Meadowview area may not grow well in Pennsylvania or Georgia. But we wilt not know this until we have blight-resistant trees to test in the various locales. If blight-resistant chestnut trees from Meadowview grow poorly in New York, it may take another 25 to 50 years to adapt them to New York. During that time, there quite probably will not be as many native American chestnut in New York as there are now with which to breed. To avoid that risk, we hope to start breeding soon in all the states. The Meadowview farm will be the prototype.

To help people get started in that direction, this issue includes two articles: on locating flowering American chestnut trees; and on growing chestnut from seed. If you are interested in seeing first-hand the techniques described in the articles, write me at Rte 1 Box 17, Meadowview, VA 24361. I can show you around the farm or put you to work most any time of year! Peak planting times are mid-February and mid-May. Our main times for pollinating are from June 10 to July 10. We especially need all the help we can get during pollination season.

Table 1
Survival of chestnut trees planted at the

ACF Meadowview Farm In 1989

Orchard	Number Planted	1989 Surviving	1990 Surviving	Percent
Minnesota B1s *	35	34	28	82
KY-lowa B1s *	150	100	46	31
Exotics **	26	26	20	77
grafts ** *	24	21	3	13
Chinese **	19	19	18	95
grafts ***	13	12	7	54
Test Plots	80	73	37	46

* Planted the completely randomized design.

** Planted in a randomized block design.

*** Twenty-four of the exotics were grafted trees. two were seedlings. The stocks of most of the grafts survived but we lost the grafted tops on all but three Likewise, 13 of 19 of the trees in the Chinese Orchard were grafts. end only seven of the grafted tops survived, white all but one of the sucks lived.

Table 2
Emergence of chestnut seed planted
at the ACF Meadowview Farm In 1990

Orchard	Number Emerged	Number Percent	Planted
Clapper B2s *	327	231	71
B1s *	110	28	25
Chinese B1s *	216	187	87
F1s *	55	13	24
F2s *	180	87	48
Age-Pathogenicity **36		11	31
Seedbed	162	24	15
Tubex test **	24	15	62
Test plot	20	16	80

* Planted in a completely randomized design. Control frees for resistance screening veers six each of Chinese and American chestnut, their first hybrid, and highly blight-resistant 'Nanking' Chinese chestnut.

** Planted in randomized complete blocks.

MEMORIES

The Legacy of Luther Burbank

Monahan Bernard
California State Coordinator, American Chestnut Foundation

Luther Burbank, one of America's foremost plantsmen, left us seven chestnut trees and a chinkapin. These are planted on the few remaining acres of his Experiment Farm in Sebastopol, California.

Four of these remnants of Burbank's chestnut work are big and old; two are petite and old; and the remaining chestnut tree is large, low headed and spreading. The chinquapin is tiny. This portion of his Research Farm is being preserved and restored by the Western Sonoma County Historical Society. I have joined, and new members are welcome. But let me tell you more about Luther and the chestnut trees.

Luther was a Massachusetts boy who went West to Santa Rosa, California. He, Henry Ford and Thomas Edison were the foremost creative talents who led the United States into the twentieth century. Luther was born in Lancaster, Massachusetts on March 7, 1849. He was the third child (the first two died in infancy) of the third wife (Olive Ross of Sterling, Massachusetts) of Samuel Walton Burbank. This made him the thirteenth child, but seven did not survive their teens.

He was a shy boy of slender build. He lived on a farm and was exposed *to* horticulture at an early age. At five, he watched his older half brother graft the apple orchard. His father was English, his mother Scotch. His mother Olive was an avid gardener and her father was the originator of several new grapes. He must have inherited "gardener's genes" from both sides of *the* family, and as a youth he also read a lot.

When Burbank was ten years old, Darwin first published On *the Origin of Species.* Dreyer cites Burbank's comrnent that he later bought the hook but never read it.[1] What lit Burbank's light was Darwin's *Variations of Animals and Plants Under* Domestication, *(2* Vols.), which he obtained from the library at Lancaster. He read this when he was nineteen (1868). He then bought Darwin's *Cmss and Self-Fertilization in the Vegetable Kingdom.* He became a disciple of Darwin, and Darwin's concept "Variation" formed the basis for his plant bneeeling plans. Although Cregor Mendel published his work in 1865 and a copy of it, in German, was at Harvard's library 30 iniles away, Luther was unaware of it until 1900, *when* Hugo DeVries rediscovered Mendel's process. By this time Luther was over 50 years old. The techniques he had developed were proven moneymakers and he was very reluctant to change a winning game. He did acknowledge Mendelism as an outgrowth and an explanation of Darwin's process of "Variation," but he advised, "After you have read Mendel, go back and read Darwin!"

In 1868, when Luther was 19, his father died and his cousin bought out Luther's mother's half of the farm. Two years later Luther bought 17 acres in Lunenburg Township which he truck farmed. He learned the economics of plant production and sales and never forgot them.

Even at this time, he was actively attempting experiments to improve his seeds and the quality of his produce. He crossed white (sweet) corn with yellow (field) corn; however he did not appreciate the need to propagate through the second generation. (He learned this after he moved to California.)

During this period he had a Rose potato go to seed (an extreme rarity). He saved the seeds and replanted them. One seed produced much larger, different potatoes, and thus Burbank's Russet or Idaho potato was created, and Luther's vocation was decided.

He took ten of his potatoes and followed his brothers to California in October 1875. Two years later his mother and sister followed. They settled in Santa Rosa, where his mother bought a house on four acres, and immediately leased 2.5 to Luther. He opened a nursery and was an immediate success.

In the 1860s California had been the Wheat Bowl for the Pacific and the British Empire. Wheat could be shipped economically around the Cape of Good Hope to England. Then, on May 10, 1869 the transcontinental railroad opened and presented the eastern produce markets now only a week away. Soon the demand for fruit and nut trees would start. Burbank thought he was in heaven.

He created 20,000 prune trees for a customer in eight months. He began crossing walnuts. Chestnuts followed. He had chestnuts shipped to him from back East and from Europe (the European shipment included a Chinese

Chestnut). Several people had already imported plum trees from Japan. Burbank had a shipment of fruit trees and nuts sent to him from Yokohama in November 1884. The shipment contained 25 large chestnuts which he called "monster nuts." The plum trees died in shipment. Undaunted, the next

year he ordered more, including a blood plum of Satsuma, which he had heard a sailor describe. These imported plum trees led to his great success as a developer of fruit trees. If there had been agricultural patents, he would have become a multimillionaire.

The chestnut trees were a different story. Walter Howard's Bulletin 691, *Luther Burbank's Plant Contributions,* March, 1945 lists six chestnut trees. They are:

"China" (1888) "Similar to the European ohly more dwarf" was listed in his circular. When Burbank found a new ("Novelty") plant he checked to see that it bred true and then marketed it. If he developed a new plant, he was much more conservative and often went 10 years before he marketed it. He continually tried to market plants and their rights to wholesalers, but he produced so many that he repeatedly was forced to market to the general public

The "Coe" Seedling (1893) One of three unnamed seedlings sold to Judge A. J. Coe of Meridan, Connecticut who called one of them the 18-Month, presumably because of its precocity. When the judge died, J. H. Hale of South Clastonbury, Connecticut bought the trees and sold the "18-Month" tree as the "Coe" and the other two as the "Hale" (1897) and the "McFarland" (1897) Other names were Coe's Early, Coe's Mammoth, New Japan Mammoth, and Sweet Japan. All of these trees were pre cocious and dwarfish.

The "California Golden" (1894) were selected seedlings of Castanopsis chryso *pylla,* the evergreen chinkapin, native to California.

The Miracle (1918), is a later developed precocious dwarf. Woodroof in Vol. 1 of *Tree Nuts, 1967, on* page 236, discusses Japanese chestnuts. He writes, "Plants of the Miracle type preduce nuts at two and three years of age when they are five feet tall. At Manheim, Pennsylvania, 35-year-old trees have trunks more than 2 feet in diameter." Are these Burbank's chestnut trees?

It is odd that Burbank worked with chestnut trees for 40 years and only marketed six varieties. Let us delve into this mystery further. We know that after Burbank's death in 1926, the Stark Bros. tock over the Sebastopol Farm and shipped several chestnut trees back to Missouri. All died in shipment. Were these Luther's latest chestnut developments, or were they part of production and not his motherhood parental stock? And how did they relate to the trees that remain?

The trees that remain are in different locations. One is located on the top of tho hill; it is big and has had its main trunk and several branches sawed off. It has at least two grafts on it; the chestnut's larger fork pollinates later than the smaller fork, and has slightly different leaf forms.

Then there are four trees planted in a row on the side of the hill, with the fifth tree offset by three feet downhill. The three big trees are planted at 20-foot spacings and now form a continuous canopy. The first big tree is close to the street (Bodega Avenue) at the edge of his property. Each of these trees is big and old, gnarled and multiply grafted. The middle tree has its trunk sawed off, and numerous limbs bear sawed surfaces. Generally, you can recognize topworked grafts, by the change of cross section; however grafted buds are sometimes difficult to detect: The discernible grafts are 20 to 30 feet high. Mter the three big trees comes the offset tree. It is old but less than a foot in diameter. Its leaves are small and narrow. The next tree is back in line and also is small. I could not detect any grafts on the two small trees.

The seventh tree is located in a different area of the farm. It is planted amid walnut trees. It is low headed with many branches and spreading. possibly this is one of his later developments. The remnants of nuts on the ground around this tree (the ground is fairly flat) seem to be of the same color and form, and the leaves are similar, with the catkins active at the same time which implies an ungrafted tree.

The chinkapin is small and barely alive. It is not a golden chinkapin, which is evergreen. It looks a lot younger.

We know that Burbank bought more land in Santa Rosa in 1884 for his expected shipment from Japan. He could have planted his 25 "Monster Nuts" there or on his original Santa Rosa acreage. He did not buy the Sebastopol Farm until December 28, 1885, just when the second shipment arrived. To transplant the trees after one year would cause a setback, so it is doubtful that this occurred. It seems more likely that he would let them grow until the scions he cut from them were established on his larger trees, which were in Santa Rosa. Thus the four big trees at Sebastopol are either from the second

shipment or from even later developments. Since the small trees closely follow the big trees, they must have been planted at the same time. They could be crosses of his original Japanese chestnut trees.

When you use a *Variation technique* many seeds and many plants are required. If one in a hundred shows the desired variation, you need one thousand to get ten variants. Often Luther Burbank started with 8,000. To mininize the work involved, Burbank grafted his scions on common stock; usually he let the insects do the pollinating; then he harvested the seeds for inspection. The nuts that he rejected (probably 70%) he sold to an Italian grocer.[2] The nuts he liked were planted; when they sprouted, he inspected them and rejected probably 70% again. The remainder were planted in closely spaced rows. A year later his choice of these would be grafted on older trees. The others went on the bonfire pile.

The burs and nut remnants under the three larger trees confirm this. Some have six nuts to the bur; some have multiple kernels per nut, some long and thin, some fat and round; some are tan, some dark chestnut brown. As a guess I would say that Luther topworked each of these trees with at least six different grafts, some of which have been removed.

Are all the sawed surfaces his work? Are these the result of pruning, or were these varieties that were superseded and got the axe? We know he didn't keep losers because he was famous for his bonfires. For months he would pile rejects until the pile was twenty feet high and thirty feet deep and then in the evening, he would torch them. It must have been spectacular!

But as of now the mystery remains unsolved. Do these trees contain his six marketed varieties? Were these trees in development? What is their ancestry? Since he was concerned with the future and not with the past; he kept limited lineage records. And with his pollinizing technique, sometimes he didn't knew their sires! Ah, yes, but what was his plan for these trees? And is there any latent value there?

This has been a good year, there was late rain in May, so there may be a good crop of nuts. Anyone interested may write:

Western Sonoma County Historical Society
P.O. Box 816
Sebastopol, CA 95473

1 Peter Dreyer, *A Gardener Touched With Genius: the Life of Luther Burbank, page 23.*
2 Ken and Pat Kraft, *Luther Burbank: The WizaH and the Man, page 162*

MEMORIES

Natural Bonsai Chestnut

Fred Paillet, Department of the Interior, Denver

The old forestry textbooks describe chestnut as only moderately tolerant, or only successful in becoming established under tight shade. A similar class of tolerance is attributed to northern red oak, a familiar forest tree throughout the range of American chestnut. Despite these descriptions in the old, pre-blight literature, some of us have been documenting the amazing degree of shade tolerance associated with longestablished chestnut clones. These are the small, bushy sprouts from old chestnut root systems. Blight kills the stems every now and then, but the root systems survive even in the dense understory of old-growth woodlands. Here are a couple of extreme examples from my field notes.

The small chestnut sprout drawn in Figure 1 is located in northern Virginia. I first noted this miniature tree in early 1986. Everything about the little tree looked ancient, right down to the densely furrowed and flaky bark. The stem was barely one inch in diameter and about five feet tall. The presence of a dead and partially decayed chestnut stump within a few inches of the base of the stem indicates that this little tree originated as a basal sprout from a former full-sized tree. The original tree was probably killed by the first appearance of blight in this part of Virginia sometime around 1925. The little tree looked old enough that one could suspect it had originated in 1925 and has survived for 65 years in the shade of the pines and oaks that now dominate the forest. Of course, I was not about to damage its tiny trunk with an increment boring.

Instead, I measured the stem and set to watch its progress over time. Of special interest was the new shoot growing from the base of the old stem. Would this new shoot become a replacement stem in the coming years, and how would the mobilization of resources needed to build a new stem under low light conditions affect the existing miniature tree?

The answers weren't long in coming. The old stem was clearly failing in 1987, and was dead by the summer of 1988. At the same time, the new stem grew rapidly to about tleree feet in height. The first thing to be done was to section the dead stem and see how old the small chestnut tree was. The rings were far too closely spaced to count with the naked eye. A wood microscope was used to make the count and showed that this tree was 40 years old in 1987. Growth had been uniformiy slow at the astoundingly small rate of about half a millimeter of diameter increment per year. These tiny growth rings surrounded an initial core repr& senting the original basal shoot establishing the stem. The size of the inner cylinder indicates that this original shoot was about the same size as the basal shoot I saw in 1986. However, the ring counts shows that this stem did not originate in 1925, but 1946. This means that the old stem was already a second generation sprout, and the new shoot established after 1986 is the third generation. One wonders how long this kind of growth can continue. The ability of these established root systems to survive makes one hope that many chestnut root systems can survive into the next century when genetic engineering can release the potential of this residual diversity in chestnut.

The second example of chestnut survival in difficult conditions (Figure 2) comes from the Harvard Forest in Petersham, Massachusetts. This poorly formed stem is not remarkable in any way, except that it was found growing in the dense shade beneath a stand of hemlocks. The light levels are so low in this location that very little grows except for a few patches of moss. Even such shade tolerant plants as wintergreen and partridgeberry are clearly sparse and suffering from the shading. The only deciduous trees estab lished here on the forest floor are a number of beech seedlings (which may actually be root sprouts) and a couple of chestnut sprouts. My detail of the terminal twigs on this little chestnut show how slowly this stem has been growing under these very difficult conditions. Of course, the density of chestnut sprouts here is much less than in the surrounding deciduous woods. The shading produced by the dense hemlock overstory has probably removed all but a few of the hardiest chestnut sprouts. The long-dead chestnut stump illustrated in my figure is just one of many underneath the hemlocks. These indicate that the the present hemlock stand arose after removal of a chestnut dominated canopy. Harvard Forest records show that this happened when stand was harvested in 1890, and not when blight arrived about 1920.

None of the old chestnut stumps show living sprouts from their periphery-It is likely that many of the sprouts from these stumps could not compete with the released hemlocks. However, at least some of the

sprouts from seedlings could remain alive. My data from many sites in New England show that sprouts from seedlings tend to be somewhat more hardy than those from the bases of large trees. The survival of even a few chestnut sprouts in one of the most densely shaded areas one can find indicates just how tough and tenacious some of these "old seedlings" can be.

Interning for the Summer at the ACF Wagner Research Farm in Meadowview, Virginia

Andrew John
University of Tennessee, Knoxville

I first came across the American Chestnut Foundation when a friend of the family who lives in Emory, Virginia, mentioned to my mother an opening for a summer intern on the Wagner Chestnut Farm. I decided that nearly anything would be better than working at Little Caesar's again, so I looked into it.

The weekend before Easter my father and I drove up to the farm for a tour. Fred Hebard showed us his prized demonstration plots, the barn and equipment, and the new trees still covered with protective aluminum cones to keep them moist, warm and free of mice.

When I finally started work the Monday after final exams we transplanted grafts into pots with lighter soil than the thick, silty Virginia red clay. By the end of the first day I could tell the difference between Chinese and American chestnuts.

Working on a farm and in a laboratory offers a variety of chores most other summer jobs do not offer. A job with so much variation tends to produce a sense of pride. My pride was strengthened by the fact that I worked to bring the American chestnut back into the eastern American forests.

The schedule for the summer depends on the flowering time of the chestnuts. Elevation and latitude play the most important role but other factors like wind, blight, and rain stress the trees and cause them to bloom early. Working around the flowering time the summer course of events went as follows: plant the grafts, measure canker growth and check for hypovirulence, open aluminum cones, cut out dead branches and trees, collect tissue from blighted trees, grow isolated fungus in agar, inoculate wild trees with blight, pollinate, and collect nuts in October. Throughout the summer many tasks like fertilizing and bush-hogging need to be done.

Although pollinating wild American chestnut trees in Mount Rogers became quite boring after a while, it did not take forever and it can also be considered as a learning experience. Fred with his vast knowledge of plants and fungi showed me how to identify more than 200 different species easily: scarlet oak, sourwood, pokeweed, jacko-lantern mushroom, chestnut oak, chinkapin, as well as how the blight affects scarlet oak, how to distinguish a buzzard from a golden eagle, and how long it takes a Frazier fir to grow to Christmas tree size.

I would highly recommend this job to anyone in high school or college interested in understanding the biological world. I got to meet the president and officers of the American Chestnut Foundation and many of the foresters who work in the southwest Virginia forests. Fred Hebard is an intelligent and well respected person, and he is fun to work with.

MEMORIES

A Practical Way for the Layman to Participate in Breeding Resistance into the American Chestnut

Patrick Chamberlain.
Cussewago Chestnut Farm, Edinboro, Pennsylvania

The use of the Backcross Breeding Method for breeding blight resistance into the American chestnut tree as proposed by Dr. Charles R. Bunnham can be easily adapted by anyone who has the desire to be actively involved in restoring this valuable species to its rightful place in our eastern hardwood forests.

Basically, all you need is an open space in a semi-protected area devoid of any foreign species of chestnut, where there is room to plant two trees 10 or 20 feet apart. Suburban as well as rural areas contain a vast number of lawns which would be ideally suited for this project.

At the time I became aware of Dr. Burnham's proposal, I had already started an attempt to cross breed chestnuts, although I didn't have a clue as to what direction my venture would take. By coincidence, my modest beginning dovetailed perfectly with the proposed Backcross Breeding Method.

Some of my ancestors immigrated to this country from Ireland and eventually made it to Northwestern Pennsylvania in the late 1700s. My great-great grandfather settled in what is now Cussewago Township in Crawford County where he built a log cabin and a small barn on a gently sloping woodland hill. To this day a tiny stream meanders between the two buildings, both of which have been reduced to piles of fieldstone rubble. All traces of the original log walls have vanished. An old hand-dug well is covered by a great flat rock, and various rusted iron implements of long ago can still be uncovered from the leaf litter if one cares to dig for them.

One of my earliest memories as a child is of my grandmother talking about chestnuts and of how the blight killed all the giant trees back in the 1930s. We would sit in her kitchen drinking tea as she reminisced about how she would have to go up to the pasture to fetch the cows while trying to avoid stepping on any bum with her bare feet. I would try to imagine what those great trees must have been like.

Time passed. I was in high school when one day a girl brought in a sharp, spiny, green bur clinging to a small branch she said was from an American chestnut tree. I was immediately reminded of the stories my grandmother had told. Now that I had a visual reference, I started searching, and within a week I located a tree on my parents' farm. I was hooked for life on learning about the American chestnut.

During my wanderings in the next few years I found perhaps a half dozen isolated, flowering small trees or sprouts which never bore anything but unfilled nuts. Then, one early summer day I decided to explore the area near the ruins of my great-great grandfather's homestead where my grandmother used to pasture the cows. The area had since given way to young second growth forest and patches of thornapples. As I was passing through a small clearing I suddenly detected the distinct aroma of flowering chestnut catkins, and upon looking found two flowering trees about 50 feet apart. My excitement at the chance discovery lingered throughout the summer as I continued to check on the trees frequently.

When fall arrived, I was able to rescue seven or eight good nuts from the squirrels and promptly planted them under a mixture of sand and rotted leaves alongside my parents' house. Around 19701 transplanted the two surviving seedlings to a permanent location. One of these was subsequently eaten by a rabbit, but the other seedling took hold and flourished.

About 12 years later I found an article in a national gardening magazine which dealt with grafted hybrid chestnut trees and the nurseries which sold them. After making inquiries of several of the nurseries, I obtained a grafted selection in 1983 named the Douglass #IA, which is purported to be one half Chinese by one half American chestnut. This graft was planted next to the American tree with the hope that the two would naturally cross pollinate. At this point I had no plan on what to do if viable nuts were produced. I realized that any progeny resulting from the cross might not have much blight resistance. I was simply hoping to produce an American type tree which would at least last a few years longer than the standard pure American chestnut. Then, shortly after my hybrid was safely in the ground, I learned of Burnham's Backcross Breeding Method.

Briefly, for the benefit of those who are new to chestnuts, the first step in backcross breeding is to cross a full Chinese chestnut with a full American chestnut. The most resistant of the resulting CA hybrids are then grown and crossed back to a full American tree. This backcross results in seedlings which will be 3/4 American and 1/4 Chinese. Again, the most resistant seedlings of this batch are grown and crossed with another American tree. This creates seedlings which are 7/8 American and 1/8 Chinese. One more such backcross will create chestnut seedlings owing 15/16 of their genetic makeup to American chestnut parentage.

At this point the most resistant 15/16 American trees are crossed with each other. The result will be some seedlings with essentially pure American characteristics which also possess full blight resistance due to the original Chinese parent.

Now that I already had a CxA hybrid planted next to a pure American, all I had to do was wait. In 1984 the Douglass #lA had a few male catkins while the American developed four burs, all of which proved to be infertile. In 1985 the Douglass #lA put on quite a show with male catkins. The American was also starting to put forth numerous catkins and eventually developed about thirty burs which were again infertile. This puzzled me at the time but I decided that perhaps the hybrid was simply not shedding sufficient quantities of pollen yet.

Things were looking great in 1986 as the Douglass #lA developed 8 female flowers and was loaded with male catkins. However, for the first time I noticed that the Douglass #lA was shedding its pollen about twelve days earlier than the American was. This meant that any female flowers on the American might not be receptive until after the Douglass #lA pollen was gone. I was particularly troubled because, as the summer wore on, nearly 400 burs could be counted on the American tree!

As September approached I kept a close eye on both trees, and my worst fears were realized when the American's burs began to open prematurely and drop unfilled nuts. However, my disappointment soon diminished. In the end I collected 21 good nuts from the Douglass #lA and eventually did get 15 from the American tree.

Since I had poor luck in previous years storing refrigerated nuts due to mold and spoilage, I elected to bury them outside for the winter. To deter the squirrels and mice, 1 dug a hole eight inches deep in a well drained area and placed a 12"x12" sheet of quarter-inch wire mesh on the bottom and bent the four sides of the mesh up about two inches high. I filled this makeshift box with earth, set the chestnuts in the earth, and placed another sheet of wire mesh on top. The hole was then filled and a layer of dead leaves was spread over the top and held in place with an old piece of pine board. The following spring the nuts were just starting to germinate in late March when I dug them up and planted them at a depth of one inch in a raised bed which had been prepared the previous fall.

During the winter after that first harvest I tried to think of a way to convince the Douglass #lA and the American tree to flower at the same time instead of nearly two weeks apart. An idea occurred: perhaps manipulating the soil temperature at the site could have a positive effect. So, in January of 1987 I hauled three bales of hay to the Douglass #lA tree and spread a thick layer all around the hybrid extending out past the drip line. I figured come springtime the hay would insulate the ground from the warming rays of the sun and retard the bud break a bit. Conversely, in an attempt to create a greenhouse environment, that April I spread several sheets of clear plastic extending beyond the drip line under the American tree. Not until four weeks later, in mid-May, did I remove it. While I never did check the temperature under the plastic, the tremendous growth of the weeds underneath was a good indication my idea was working at least to some extent. I left the hay under the Douglass #lA tree. Eventually it turned to compost.

By the end of June that year it seemed that my experiment had worked. The American tree started shedding pollen only three days later than the Douglass #1A. But at harvest time, while the nine burs on the Douglass #lA dropped with filled nuts, it was obvious that the female flowers on the American tree had still not been receptive on time. Although there were over 800 burs on the tree that year, the grand total of nuts collected from the American was 18.

All of the nuts gathered in 1987 were stored in the same manner as those in 1986. In the spring of 1988 a total of 61 seedlings were transplanted to their permanent location at a spacing of six feet apart in rows also six feet apart. There were two reasons for planting them so close together. Because these seedlings owe _ of their genetic makeup to American parentage, only about one in four will have the resistance required to be retained for the breeding project. Therefore, three out of four will eventually be

removed, which will allow more room for the remaining trees. The second consideration was efficiency. It is much easier to care for a smaller plot.

That summer I used a rototiller to break up and pulverize the sod to a depth of 10 inches between the rows of seedlings. Although tilling was time consuming, it was the best thing I could have done. The seedlings responded by more than doubling their growth the following year.

In this way I discovered that the elimination of all sod cover from the immediate vicinity of small chestnut seedlings is very important. There is little point in becoming involved in the breeding program if the seedlings produced are not allowed to grow and push their roots through a loosened soil devoid of competition from weeds and grasses. Seedlings grown in sod will either wither and die within two years or at best will have their growth badly stunted for five or six years.

As of the end of 1990 most of the 53 surviving hybrids, which have completed their third and fourth growing seasons respectively, range in height from four to seven feet. Several have had cankers which have disappeared, but it is too early to tell if this is due to blight resistance, hybrid vigor, or whatever. Subtracting the "scrubs" and the slow growers leaves 33 which I feel are suitable candidates for growing and testing for blight resistance

To create the 7/8 backcross the most resistant hybrids will be crossed to one of two pure American trees also growing on the site. One of these is a 10-year-old transplanted sprout and the other is a 2-year-old graft from a tree near Erie, Pennsylvania, which reached about 70 feet before it started to decline. The sprout had had male catkins the last two years.

The prospects for producing more 3/4 American nuts in the future are good. Although the original American parent of the hybrids has died and was cut down in 1988, in its place as of the end of the 1990 growing season are about a dozen sprouts each reaching 14 feet high. This will be a tremendous advantage. Essentially, where there was once one American tree there are now 12 separate crosses. When the blight hits again they won't all die at once. As some of the stems succumb others will be flowering, and since the area receives full sunlight, the root crowns will be continually sending up new sprouts to replace the ones that die off. Some of the older sprouts should be producing pollen again in a year or two.

The Douglass #IA graft is now seven years old (nine counting the rootstock), and is 26 feet high. Due to a lack of pollen because of the death of the American tree, the 110 burs produced this year by the hybrid were infertile. However, since the female flowers of the Douglass #IA are so receptive to the American pollen, the coming years could be very productive indeed.

If one is serious about becoming actively involved in the breeding project it is easy enough to get started.

Locate an American sprout in the woods, tie a ribbon on it, get permission to dig it up, and then do so after it goes dormant. Plant it in an open area with good drainage where it will get plenty of sunlight. Contrary to popular belief, chestnut trees do not prefer to grow on rocky ridges; they simply possess the capability to do so. The truth of the matter is that they will do very well (actually better) in good fertile topsoil.

Since there are good hybrids available, there is no need to pair off the American sprout with a pure Chinese tree. The question arises as to which hybrid to plant next to the American tree, and whether it should be a seedling or a graft. Most of the hybrid seedlings available today are from open pollinated trees in orchards which contain a veritable smorgasbord of chestnut germplasm. While these orchards can and do produce some very good seedlings, In many cases it would be difficult to tell what the exact parentage is. To lessen the risk of planting a dud, it would probably be wise to plant up to 10 or 20 hybrids in close proximity to the American sprout, and after six or seven years save the one exhibiting the roost resistance and best timber form for breeding purposes. Of course, this still does not guarantee that the remaining hybrid will be a good tree.

I believe the better approach is to go with the grafted tree, although this also entails some risks. Craft incompatibility, in which the rootstock rejects the scion, can be a problem the first two years after grafting. The way to get around most of this problem is to buy material which has already been growing for a year or two in a reputable nursery. As long as the scion has been growing vigorously, graft incompatibility should not be a problem.

The late Earl Douglass of Red Creek, New York, is responsible for producing the Douglass #IA hybrid. What makes his achievement so remarkable, in my opinion, is that the Chinese parent he used

has very poor form, even for a Chinese tree. However, when he crossed it with the American parent many of the seedlings which resulted eventually proved to be bonafide timber trees with higher blight resistance than one would expect for FI hybrids. Possibly the most resistant, best timber type tree Earl produced is the one named the Douglass #IA. One of the few nurseries where this grafted selection can be obtained is the Crimo Nut Nursery in Niagara-on-the-Lake, Ontario. Ernie Crimo grafts the Douglass #IA scions onto Douglass seedling rootstock which greatly reduces the chances of graft incompatibility.

John Cordon of North Tonawanda, New York, has some interesting hybrids in his plantings. John rarely sells grafted chestnut trees, but he does sell scion wood and seedlings. One of the Douglass selections he has is the Douglass #4 which is a larger fruited variety than the Douglass #1A. One of the better timber potential trees he has is something called the Layeroka X Watertown #3. The Layeroka parent of this hybrid was created by the late J.V. Cellately of British Columbia. It is a complex hybrid containing both Chinese and European stock, and is an upright heavy-bearing selection. The Watertown #3 half is from a group of trees purported to be American from near Watertown, New York, which seem to have a bit of resistance. I have a young graft of the Layeroka X Watertown #3 and it does seem to have good timber potential. There are quite a few other nurseries which sell hybrid chestnut trees. I have only mentioned the two I am most familiar with and can recommend from past experience. I would suggest contacting a good number of them to find out exactly what is available.

After deciding on a grafted variety, plant it next to the American sprout and protect it by placing a wire mesh cage and two or three stakes around it. Under optimum conditions a grafted tree will begin to bear two or three years after planting. Depending on the age of the American sprout, it should begin flowering in two to five years. When nuts are produced they should be protectively stored during the winter and then dug up in early spring before germination begins. The seedlings should be grown for at least two years in a raised bed before being transplanted to a permanent location. For the first three or four years they will need to be watered during dry spells. At the time the nuts are placed in the raised bed it might be a good idea to cover the bed with a mulch of dead leaves to help prevent weed growth and guard against possible frost damage.

At the present time I am hesitant to distribute any material from my hybrids simply because they are a bit too young to be tested for blight resistance. However, once I do know which trees are carrying the resistant genes, I would be happy to distribute scion wood to anyone who might be interested in participating in the breeding project. We need to work together to promote a free exchange of ideas and material in order to fully exploit the potential benefits that are possible to achieve.

Increasing the number of people working on this noble experiment can only enhance the prospects for bringing back the finest tree God ever decided to create the American chestnut.

MEMORIES

A Minnesota Story: Restoration of the American Chestnut

Charles R. Burnham
A memoir by a founder of the American Chestnut Foundation

In December, 198O, I found Frank Kaufert's publication on "Prospects for the American Chestnut in the Upper Mississippi Valley" in the list of University of Minnesota publications in the bulletin room on the St. Paul Campus. I read it over the Christmas holidays and learned that blight resistant American chestnuts had not been produced by the United States Department of Agriculture (USDA) breeding program. Why not? What happened?

My first job was at West Virginia University in Morgantown (1934-1938), where I had seen large, almost dead American chestnut trees that often produced flowering branches. Hence, pollen was still available.

I knew vaguely that the USDA had a chestnut breeding program, but not until 1959 did I write asking about the program. Fred Berry responded by sending me the 1954 Farmers' Bulletin No. 2068 on chestnut blight, and also a 1955 report on the status of chestnut breeding in the U.S. given at an international meeting in Rome. The *Farmers' Bulletin* had a picture of a row of tall 15-year-old H hybrids between the blight resistant Chinese (C) and the American (A) chestnut, with the statement that the hybrids were more resistant than the American but less resistant than the Chinese chestnut parent. Resistance was incompletely dominant, obviously, but not stated. I remember mentioning to someone that in backcrosses to the American chestnut, the moderately-resistant segregates could be selected for the next backcross to American chestnuts, this to be followed by successive selection and backcrossing to American chestnuts. Hence, I was surprised to learn about the failure of the program.

The Old Approach, and the Genesis of a New One

I read the references given. I could not believe what I was reading. Others reacted similarly. In the USDA chestnut breeding program, the backcross method had not been used in the manner required to produce American chestnuts with blight resistance.

Most of the backcrosses made were to the resistant parent, though they should have been to the American chestnut. Only a few were from a backcross to the American parent. None were beyond the first backcross. When I explained to Carl Moha in the Forestry Department what had happened, he responded by saying backcrossing should succeed. Colleagues in my department, in Plant Pathology, and Horticulture agreed.

The following is from the 1986 review, "Breeding blight-resistant chestnuts" by Burnham, et al.: "Many pollinations of native species were made at the USDA between 1894 and 1911 using European and Asiatic species then available (Van Fleet, 1914). When the blight disease appeared in their plantings in 1907 and the American chestnut and its hybrids developed the disease, work continued only with hybrids involving the European and Asiatic species and native American chinkapins." Three resistant selections were given Plant Introduction numbers and were used, along with new importations of Chinese and Japanese chestnuts in the breeding program that was resumed in 1922 by the Office of Forest Pathology, Bureau of Plant Industry, USDA. The first crosses were between the blight-resistant Japanese chestnut and the American. When they teamed the Chinese chestnut was more resistant than the Japanese, the JxA hybrids were crossed with the Chinese chestnut. Following a suggestion by D.F. Jones, the Cx(JxA) hybrids from that cross were crossed with the American chestnut.

Thousands of hybrids were produced by hand, controlled, pollination. Most of them were obtained by crossing the various blight-resistant species with each other and with the American chestnut, and then crossing the most promising hybrids. A similar program, begun by Arthur Graves while he was employed at the Brooklyn Botanic Garden was transferred to the Connecticut Agricultural Experiment Station at New Haven in 1947. I realized much later that the goal of both programs was a single tree that would be

blight resistant, and have the desired timber-type growth form. It was to be propagated clonally as for apple cultivars. Based on his experience with hybrid corn, D.F. Jones believed it would be a rare combination among thousands of hybrids (see the *Journal of the American Chestnut Foundalion* 1:8-11, 1987). Hence their goal was not to restore the American chestnut by adding blight resistance, but to produce a blight resistant chestnut with timber form that would replace the American chestnut. The USDA program had been terminated about 1960, possibly because they believed that they had attained their goal.

We now realize that a single hybrid clonally reproduced will not restore the American chestnut.

What could be done to put the program back on track? Although I had read that the American chestnut was extinct, I soon learned there are flowering American chestnut trees growing in the University of Minnesota Landscape Arboreturn. Frank Kaufert's publication described additional ones in Minnesota, Iowa, and Wisconsin.

At one time there was also a Chinese X American Fl hybrid at the University of Minnesota Arboretum, but it died back every year due to winter injury. It had finally been removed when Dr. Brierley, the one person in Horticulture interested in nut crops, retired. A European chestnut at the Arboretum still dies back every year. New shoots come from the roots.

The beginnings of a New Approach

We needed the cooperation of those who had worked on, or were still working on the chestnut problem; hence we had to explain the backcross method and why it should be used. With the help of Drs. David French and Thor Kommedahi, I submitted a Letter to the Editor which was published in the international journal *Plant Disease* in 1981, describing the backcross method, how it could be applied to produce blight-resistant American chestnuts, and also requesting information on Chinese x American hybrids that might be used for crosses on the American chestnut trees growing at the University of Minnesota Landscape Arboretum.

A news item with similar information was released April 1, 1981 by the University of Minnesota Extension Service. Responses came from several countries. Al Newhall at Cornell University sent male catkins from a chestnut tree at the home of L.H. McDaniels. A year later, at the 75th anniversary of the Plant Breeding Department at Cornell University in 1982, Dr. Will Provine went with me to McDaniel's home. The tree was flowering, but the lowest branches were out of reach.

The tree was at least six inches in diameter. Dr. Provine wrapped his arms and legs around the trunk and pulled and hunched himself up to the branches and cut off several. I brought catkins back for use at the Arboretum. A scion from an American chestnut had been grafted on a Chinese chestnut stock. The graft had died after growing to a diameter of about 6 inches. The living tree *was* a Chinese chestnut.

Earl Douglass responded to my "plant disease" letter by sending a brochure about his chestnut hybrids in New York, plus some large nuts. They were from a cross between a Manchurian (Chinese) chestnut and the American chestnut. But were they F1s, or from open pollination between the hybrids and other chestnuts growing there? Later, some were identified as probable F1s. A few other responses were received, but none with information on usable hybrids.

Search for Usable Hybrids

When I explained the backcross method and the reasons for earlier failures, a group of local scientists and others became interested in making an attempt to restore the American chestnut. The recurrent parent would be the American chestnut. A search began for hybrids suitable for beginning the backcross breeding program. The first crosses in this new breeding program were made in 1981. When I wrote to Richard Jaynes in 1981 about the backcross breeding program, he was skeptical about its prospects for success, but sent pollen-shedding catkins from tree WdSL Row 11T7, a [Cx(JxA)]xA cross.

That pollen was used by Harold Petlett and his staff on two American chestnuts at the University of Minnesota Landscape Arboretum. Tree #58681C produced 2 nuts from the cross. Tree #68242A also produced 2 nuts. (Note: the first two numbers: 58.. and 68.. are the years in which the seedlings were planted.)

Tree WdSL Row 11T7? was undoubtedly a promising survivor, but its ancestry is complex. First, three different species are involved. Hybrids from the Cx(JxA) cross would have received half their

inheritance from the C parent, but for the other half they received from the JxA hybrid there are many possibilities. Tree WdSL Row 11T7 received half of its inheritance from A, but for the other half there are also innumerable possibilities.

Many of the reports on the breeding work had been published in the Annual Reports of the Northern Nut Growers Association. Most libraries have incomplete sets. Some were missing from *the* University of Minnesota forestry library. In December 1981, our University library borrowed missing ones for the years following 1925 from the State University of Iowa library at Ames. I photocopied all the pertinent articles on chestnut breeding before returning them. Over the Christmas holidays I read, among others, Dr. John Shafer's 1966 paper that described his results from a planting in Indiana of 100 Chinese X American chestnuts (CxA) F2s he had obtained from Diller in the USDA. He also discussed what might be accomplished if large numbers of F2s were grown in blight areas. Nature would select those that were blight resistant.

I wrote to him in January, 1982. He replied that a few of the F2s had survived and were producing nuts. These are F3s possibly the first ones ever grown. He also wrote that he had obtained Chinese chestnuts from Arthur Graves, one of those involved in the Connecticut breeding program, had crossed them with the American chestnut, and had flowering F1 hybrids at Logansport, Indiana, and in Tennessee.

Dr. John Shafer received a Ph.D. degree in plant physiology at Cornell University with a minor in plant breeding. He helped Marcus Rhoades at least one summer with corn pollination of the Maize Genetics Cooperation stocks. His first job was at Virginia Polytechnic Institute, Blacksburg, Virginia, but he left it to manage the family lumber business in Logansport, Indiana.

Mr. Philip Rutter (currently President of the American Chestnut Foundation) came into the picture at about that time. Dr. Phil Riegel from the University of Minnesota was visiting my older daughter in Chicago. She told him what I had discovered and what we were doing about the American chestnut. He told her that a friend of his in Southeastern Minnesota was growing chestnuts. I finally obtained Rutter's address and telephone number from Phil Riegel. I convinced Rutter that backcrossing to the American chestnut was the only way to go.

In 1982 pollen from John Shafer's CxA F1 hybrids was used for crosses by Dr. Harold Pellett and his staff on American chestnut trees at the University of Minnesota Landscape Arboretum; and also by Rutter on isolated American chestnut trees that he had located in northern Iowa. The Iowa flowers did not need to be bagged since isolated chestnut trees rarely produce nuts. They are self-incompatible. Pollen from other chestnut trees is required to produce nuts.

Also at about that time I told Dr. Lawrence L. Inmen the same story. He had received a Ph.D. degree in 1957 under me with a thesis on a cytogenetic problem in maize. He had a B.S. degree in forestry from Iowa State University (1947) and had prepared a review of the literature on tree breeding for Dr. Arthur Wilcox in Horticulture here at the University of Minnesota. He now says he knew the American chestnut story, but believed he was in no position as a student to do anything about it, and did not know that I had any interest. Inman's first job had been on a Ponderosa pine tree breeding job in Idaho. After completing various foreign assignments, he was operating a farm at Danvers, Minnesota. He became interested immediately. He has helped since then with pollinations at the University of Minnesota Arboretum. Inman's familiarity with the tree breeding literature, his firsthand experience in a tree breeding project, and background training in forestry, genetics, and plant breeding have been a valuable resource. We have had numerous discussions about goals of the chestnut program to establish populations of blight-resistant American chestnuts adapted to different growth zones. Discussions by Inman of strategies to accomplish this are in two issues of the *Journal of the American Chestnut Foundation.* He drove to Connecticut to help Fred Hebard with bagging in preparation for the 1989 crossing and to exchange ideas and information with Sandra Anagnostakis and Philip Gordon. He also visited Paul Galloway who has an excellent American chestnut tree in New Hampshire that is being used for crossing.

The nuts produced in 1982, first backcrosses to A, were all sent to Dr. David Benzing at Oberlin College, Ohio. Dr. John Shafer has provided pollen several times since then. He has also sent us F3 nuts from his F2s.

Rutter and I discussed the possibility of giving a talk at the 73rd Annual Meeting of the NNGA in 1982. Since the meetings are in mid-August, usually at the peak of my corn pollination, Rutter gave the talk we prepared. The published paper is a revision of that talk. The talks and other articles are published there

without peer review.

The "Clapper" Tree

The USDA (Diller) and CT (Graves) had established forest-type test plantings of chestnut hybrids in 15 sites in 13 states from 1947 to 1955. They had been evaluated several times, the last time In 1978 by Fred Berry.

Fred Berry, one of the last workers in the USDA chestnut program, had subsequently moved to Delaware, Ohio, on another program. He had published in 1980 a general summary of his 1978 observations on those plantings. He sent me copies of his field observations on the survivors in those plantings and their pedigrees where available.

One hybrid Berry found in the 1949 Carterville, Illinois, forest-type test planting was from a first backcross made by Diller in 1946. That tree, designated the "Clapper" hybrid, was described as the most promising of all their hybrids, the longsought, rare hybrid with blight resistance and excellent form. Since it was a first backcross (F1), its resistance could have been no greater than moderate, similar to that of the CxA F1 hybrid. In fact, the "Clapper" the tree had survived the blight for about 25 years, but finally devetoped a large canker at the base and died. Contrary to expectations, it was not widely propagated (Diller and Clapper, 1969). Since these would be clones of a single tree, nuts would be rare. Pollen from other chestnut trees would be needed for nut production. If their recommendation the planting of an elite Chinese chestnut as pollinator had been followed, the progeny would have been backcrosses to the Chinese chestnut, useless for the restoration of the American chestnut.

Blight resistance was believed to be dependent on many genes, and the other desired traits were certainly complex in inheritance. In fact, the problem was considered to be so complex there was no chance of success in the breeding program (Jaynes, 1960). Research turned to other means of control. Only a few crosses were made in Connecticut in the 1970s. Most research beginning in the early 1970s and continuing to the present in Connecticut and several other states has been on hypovirulence as a possible method of blight control.

None of the other survivors in those forest-type plantings were ideal for beginning the breeding program. They were complex hybrids or from backcrosses to the resistant parent, not to the American chestnut.

Dr. Jaynes and others were skeptical about the prospects of success of abackcross program, but provided information on some of the missing pedigrees and answered questions. Records of the breeding program and reports of Mr. Liu and others who had sent material from China had been transferred from the USDA in Washington, D.C. to the Forest Products Laboratory in Madison, Wisconsin. I visi ted there and borrowed the record books and later also the Plant Introduction cards for chestnuts.

The Clapper tree had died. No sprouts developed from the tree. Only two clonal sources have been located. Grafts on the Chinese chestnut were in the Lesesne State Forest in Virginia, but Tom Dierauf, Virginia Division of Forestry, checked and found the Clapper scions had been winter killed.

Then, late in 1982, Richard Jaynes remembered that 3 Clapper grafts on a Chinese chestnut were growing in a Connecticut Experiment Station planting near Hamden, Connecticut. In 1983 he crossed them on a Scientists Cliff American chestnut and also with pollen from an American chestnut identified as being somewhat resistant to the blight this is the Floyd American tree (Griffin, et al. 1982). The nuts produced from these crosses are second backcrosses. Clapper pollen was used also on the American trees in the University of Minnesota Arboretum and also by Rutter on American chestnut trees in Iowa.

The nuts were stratified here in Minnesota and the sprouted nuts were sent as follows: nuts from the Clapper x Floyd American to Blacksburg, Virginia, the others divided between Morgantown, West Virginia; Great Smoky Mountain National Park, and here at the University of Minnesota. Grafts of some of Shafer's F3s on chestnuts at West Virginia University, Morgantown, failed (William MacDonald in a recent communication). Clapper pollen was used again in 1985 on American chestnuts in the Arboretum and in Iowa. These are growing at the University of Minnesota. The Clapper clones in Connecticut are still flowering and were used again in 1989 for crosses in Connecticut.

The seedlings from the 1983 crosses were to be cloned at each location so that they could be sent to the other locations. Each location would have a complete set. American chestnut leaves have only a few simple hairs on the underside. Chinese chestnut leaves have a woolly met of simple and stellate hairs. There are some exceptions. The leaves of CxA F1 hybrids are intermediate in numbers of hairs, but these

usually appear in seedlings only at the end of the second year of growth. Under conditions favorable for rapid growth this has occurred at the end of the first year. The leaves on the Clapper clones have only a few simple hairs, an occasional stellate hair. Stronghold, Inc., at Sugarloaf Mountain near Washington, D.C., has seven flowering and fruiting trees from open pollinated nuts from the Clapper tree. These are near their headquarters building. The leaves on branches I obtained in 1987 when I visited there show that for all but one the hairiness is intermediate, and must have come from crosses with other hybrids or with the Chinese chestnuts in the Carterville planting. Nuts were mature at the time of my visit in October. The burs were just beginning to open. All had three large nuts per bur.

Founding the Foundation

Some funds for the work had been received for the project, but it became obvious that these would not be sufficient. Rutter proposed that a Chestnut Foundation be established. With the help of Mr. Donald Willeke, a Minneapolis lawyer with an active interest in trees and the Chestnut work, the American Chestnut Foundation was established in 1984 in Washington, D.C., but the first officers of the Foundation were all in Minnesota: Philip Rutter, President, David French, Treasurer. The University of Minnesota Agricultural Extension Service sent a newsrelease about the Foundation, on March 8, 1984, in a list of local papers and other publications in the states within the natural range of the American chestnut plus the Experiment Station's Horticulture list.

Beginning December 1988, Dr. William MacDonald, West Virginia University became Treasurer.

Micro-propagation

Yang Qui-guang, a Chinese scholar who had worked with chestnuts in China, spent a little over a year (1985-86) in Dr. Paul Read's Horticulture Department lab working on tissue culture propagation of the American and European chestnuts. He was successful in getting proliferation of microshoots. Rooting percentages varied from 2 to 8 in 10-microshoot tests. He also was able to establish them in soil in pots (Yang et al., 1986, *Hort. Science 21:).* Attempts to repeat the final procedures, transfer to soil, since then have failed.

Chestnut Workers

The immediate goal is to complete the transfer of blight resistance to American chestnuts as rapidly as possible with the backcrosses made in 1982 to 1986. These blight-resistant American chestnuts can then be used in the later backcrosses in the long-term program described above. these will not encounter the difficulties that probably are occurring in the progeny from crosses between the American and Chinese chestnut species in the first steps in the backcross program.

The ultimate goal is to establish breeding populations of blight resistant American chestnuts, each of which will be adapted to a different growth zone in the natural range, as described by Inman in 1987 and 1989 in the *Journal of the American Chestnut Foundation.*

The Foundation also hopes to manage and document existing sprout populations and the occasional larger American chestnuts so that they can be used in crosses to introduce blight resistance.

Beginning in 1987, pollen from the hybrids being used in the 15 backcross programs was sent to several people who have fruiting American chestnut survivors in the natural range. Those now involved in this program include Tom Dierauf, Virginia Division of Forestry, Charlottesville; Tom Hall, Tennessee Tech University, Cookeville; Scott Schiarbaum, University of Tennessee, Knoxville; John Kuser, Rutgers University, New Brunswick, New Jersey; Philip Gordon, Yale University, New Haven, Connecticut; Sandra Anagnostakis, Connecticut Agricultural Experiment Station, New Haven; Charles Maynard, State University of New York, Syracuse; John Kelley and Alan Newhall, Cornell University, Ithaca, New York; and Paul Galloway, Walpole, New Hampshire.

American chestnuts homozygous for the genes for blight resistance can be established in each of the different growth zones, the progeny from crosses between them and American chestnuts in the same zone that are heterozygous for those genes will range in resistance from moderate, like that of F1s, to full resistance.

Addendum:

This story would not be complete without acknowledging the many chestnut workers who have

provided information through correspondence and telephone conversations. They have shared not only their own experience, but also their information from unpublished work of others, e.g., that of the late Bruce Givens, through John Elkins. I have learned much from those involved in the earlier breeding program, i.e., Fred Berry, Richard Jaynes, Hans Nienstaedt, and Jack Elliston, and from many people who are currently active in chestnut research such as William MacDonald, Gary Griffin, Tom Dierauf and John Elkins. Sandra Anagnostakis has used the plans for the original chestnut planting plans to locate and identify the various species and hybrids, and Philip Gordon is locating, identifying and cataloging the chestnut sprout populations and older trees now growing in Connecticut and in several other states.

Thanks to the Wagner family in Washington, D.C., the Foundation now has the use of part of their farm near Meadowview, in southwestern Virginia, for chestnut research. Research there is being conducted by Dr. Fred Hebard. In 1989 Hebard utilized the White Memorial Foundation plantings of CxA and AxC F1 hybrids and Chinese chestnuts at Litchfield, Connecticut for crosses. These Litchfield plantings were made by Graves and Nienstaedt in 1944 and 1953. Hebard also used the Clapper clones for crosses with Chinese and American chestnuts in 1989.

Every *Castanea* species is now represented in those plantings. Included are winterhardy versions of the blight-resistant Chinese and Japanese chestnuts. These are being used for crosses with American chestnuts to produce F1 hybrids for which both parent trees are known. There are two excellent Japanese chestnut trees in Connecticut that were planted about 1876. Chinese chestnut cultivars were crossed also with American chestnuts in 1987 to provide F1 s with known parentage.

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MEMORIES

American Chestnut in the Northwestern Lower Peninsula of Michigan

Jim Comp

(from a talk given at the Michigan American Chestnut Workshop, 1981)

I was six years old when we moved to West Virginia, and probably picked my first chestnut at that time because it was in its heyday. There were chestnuts every-where and everybody had a grove of chestnuts.

The woodlot was chestnuts. I don't suppose that anybody here today ever stole somebody's watermelon, corn, black walnuts or chestnuts. Now in that chest-nut grove, you might get a little buckshot. We could get in and get out and, if you've never felt buckshot, I have, and I was down on my hands and knees when it happened, and that buckshot felt as big as a baseball thrown at my back. But they didn't break the skin, so I often say, if I had to serve time, that's not too bad.

Chestnut was king of the forest! You were raised with this thought and little did I know about what was killing our trees. But they started to die right after the First World War and by 1925 practically everything was dead in the state of West Virginia. We moved to Ohio in the Metropolitan Park area around Akron in 1928. We used to have our Boy Scout camporee in Metropolitan park and we found a lot of chestnut trees there and we gathered the nuts. Then these trees began to die. I know of no trees that were left in Ohio when we came to Michigan and moved to Cadillac in 1937

When I came to Cadillac, the American chestnut was on my mind. Of all the hours we spent in the woods, not only gathering chestnuts - we gathered walnuts and hazelnuts as well - 1 never saw a free t didn't love, no matter how homely it was. These trees are homely because they have struggled. You see, the American chestnut has struggled to overcome this disease that came in 1904 and they are still struggling. They're still shooting up and saying "Somebody help me!"

You have these men here today who are going to do something about it. Pennsylvania alone spent $275,000 in 1912. That was a lot of money then. They cut down every diseased tree they could find. They burned it to try to control the disease. It didn't work so they lost their money.

Well, when I came to Cadillac, I found a chestnut tree back on the alley which was producing nuts and all at once it quit. I didn't know why it quit. I didn't know until I met somebody, probably Dr. Bill MacDonald, who told me there are male and female. You have to pollinate. I said, "What do you mean?" He said, "Well, you've got to have them within 1300 feet." That is a long way. I don't believe it will go that far. But it's got to pollinate. It lost its buddy. Five blocks up the street had been two other chestnut trees. Now these didn't die with the disease. They just died. But this poor tree. He couldn't make it alone. He did everything but produce seed. He blew. He burned. And you got all blanks. Later on, I found three more trees. later on, two more. And later on, a few more and they were producing nuts. I ran across one that didn't produce nuts. Well, I began to realize then that he didn't have a buddy. We made a covenant with ourselves that here was a loner. Someday we were going to see to it that they had a buddy. That day is coming true. That is, in Cadillac, there is a single tree where they built the new apartment houses. When they laid the drive out, they laid it out to take out that tree. I happened to ... just happened to ... go around to the superintendent and say "Hey, you know what you got? That is an American chestnut." He says, "Wait until I get the owner." They changed the drive to save the tree. They are pruning it and it is beautiful. And we have, within a quarter of a mile, a tree about 12 years old. Three or four more years and it will pollinate that tree and I hope I'm here to see it when it happens.

Well, Chet Arnett, of the *Detroit Free Press,* thought that he and his wife were experts on the American chestnut. He said they were extinct and he could remember when he gathered chestnuts when he was a little boy and they boiled them. So I sent him a pound of chestnuts and burs and told him I had news for him, that we have chestnuts. He was very kind to write back and say that he and Betty sat down that night and roasteel those chestnuts.

Then the Forest Service came out. This is the organization that is so sham, you know, and they know everything about growing trees, accelerated growth, all this stuff. It is amazing to me what they can do

when they make a pine tree grow in 12 years what nature does in 30. More power to them. But they came out and said there weren't 100 American chestnut trees left in the United States. So I wrote to, I think, every state from Maine to Alabama, to the Departments of Natural Resources. We got lots of answers back from them, not that they knew where there were any chestnut trees or of any hopes or anything like this. But they were concerned and wrote back and then more and more articles came out that no more American chestnut existed. The forest is dead. By that time we were up to maybe 2,000 or 3,000 trees. Now, we stand between 7,000 and 8,000 in eleven counties in Michigan, aged from 123 years down to yearlings.

POLLINATION AND PLANTING

Quick Guide to Making Controlled Pollinations of Chestnut

Philip A. Rutter

This set of quick instructions is condensed from A *Chestnut Pollinator's Handbook,* and will put you on the right track if you want to make your own hybrid nuts. There are plenty of quirks to chestnut flowers, however, so he aware that the *Quick Guide* does not cover all the facts and possible problems; the *Handbook,* which is now up to 18 pages, tries to. We hope to have it ready soon.

These tasks are presented in the order in which you will probably have to perform them. In the northern half of the country, you should start looking at your trees in early or mid-June, so you can judge when you will need to go to work; in the southern US, trees can he receptive in May.

1. Records

Write down everything you do: how, when and where. Any nuts you make will he unidentifiable and worthless otherwise. Always check the species identification of your frees; mistaken identifications are *very* common.

2. Bagging

Even on isolated trees, chestnut flowers must he "bagged" to exclude random airborne pollen from fertilizing or blocking them. There are four problems in bagging chestnut flowers: heat, wind, moisture, and soft twigs. Most other trees flower in cool weather, and bags put on them can he fastened onto well-hardened wood. Chestnuts flower in hot weather, on fresh shoots that may he soft. Leave some flowers unbagged so you can judge the correct time to pollinate.

a) When: Bag female flowers as late as possible to decrease damage and mortality of flowers. Try to bag just as soon as the first chestnut trees in the area start to shed pollen, but not earlier. The female flowers should not he receptive yet.

b) *What:* The bag to be used should have no pores that can pass pollen, but should "breathe" to decrease moisture buildup. Brown paper grocery bags can be used to enclose entire branch tips, and can be fastened on hardened wood. Smaller white "corn shoot" bags specifically designed for pollination (Lawson Bags #421) keep the flowers cooler and may be less susceptible to wind damage, but may have to he fastened around soft wood.

c) *How:* Remove the male catkins that would he included inside the bag, including the male portion of the bisexual catkin; use small sharp scissors. With small bags, it may help to cut off the tips of some of the leaves, or remove them, so the flowers are not too crowded and don't get too wet in the bag.

Wrap the place on the twig where the bag will he fastened with an unwound cotton or polyester ball (available at any drugstore). This will prevent abrasion, allow some air in, and keep moisture out without allowing pollen in.

Secure the bag around the branch, making certain the bag is closed and no pollen can enter. I use a common stapler and several staples; at least one staple going through some of the cotton. If you use wire, be careful not to get it too tight.

2. Pollen Collection & Preservation

If you are making controlled hybrids, you need to have someone send you preserved pollen from an appropriate tree; if you are gathering your own pollen, read on. It is possible to collect fair amounts of pollen from chestnuts, but if you have collected pollen from any other tree species, you will he surprised at how *little* pollen chestnuts produce. If you are collecting directly from trees, try to pick a calm day; early

morning may he your best bet. Pollen is available on cool or hot days, morning or afternoon. Don't try to collect it if there is dew on the trees.

a) *Fresh Catkins:* If you can pick fresh, early-mature male catkins to use as a pollen source, this works fine. You can use them like brushes to apply pollen to your receptive female flowers within 3 hours. If you have to hold them longer than that, try to keep them dry and refrigerate them.

b) *Flowering branch in a bucket: An* old nutgrower's trick for unbagged isolated trees is to pick a small flowering branch of the male tree, put it in a container of water (add a little 7-Up if you want to go high tech) and hang it somehow in the crown of the female tree. You should get some nuts this way.

c) Fresh pollen on a glass microscope slide. This is now my favorite methed for using fresh pollen; I think I get better results even than with the fresh catkin. Hold a scrupulously clean microscope slide (or similar small piece of glass) under the male catkin, either on or off the tree. Tap the catkin lightly on the slide, three or four times. If there is good pollen in the catkin, you should be able to see it on the slide, as a *very* faint, fine, dust. Several loaded slides can he carefully packed up, so their sides won't touch anything, carried to the female free and used to apply the pollen to the females. Use them within 2 to 3 hours if possible, keep them protected from wind or dust, out of the sun, dry, and cool.

If your male parent has only a few catkins available to you, this way you can gather pollen from the same catkins for days.

d) *Collecting larger amounts of pollen. If* you want to store pollen for use next year, or to send pollen to someone in another state, you'll need to collect more than is possible using the previous methods. Pollen can he collected either directly from the trees, or from branches brought inside to ripen; which method you choose will depend on how far away your trees are, how high the flowers are, and your own preferences. Outdoor catkins will produce more pollen if they are bagged some days ahead of time. This excludes wind and insects, also pollen from other trees. Dick Jaynes recommends this, and suggests using Malathion dust (insecticide) in the bags to kill the small insects that are constantly grazing on the catkins.

Method 1: Gather mature catkins and put them on trays in a windless, dry room for 2-3 days. Put the partially dried catkins through a fine sieve, using a toothbrush or something similar, and dry the sieved material as described below.

Method 2: (Currently my favorite) Get a clean, smooth piece of window glass. Just before collecting pollen, wipe it down with new lens cleaning paper, but no liquids. This eliminates dust and unwelcome pollen from other trees. Don't cut yourself on any sham edges. Hold the glass under a bunch of ripe catkins, and lightly strike the catkins against the glass, several times. Chestnut pollen sticks to glass well, but it can be knocked off the glass into the air or onto the leaves if you continue striking too long.

Hold the glass up to the sunlight after several hits if you can see a faint haze on the glass, collect the pollen before continuing. Chestnut pollen is very tiny; you *cannot* see the individual grains. There will be plenty of visible "trash" on the glass, including anthers, anther filaments, and bugs. If you have plenty of catkins to collect from, you may want to tilt the glass and tap it to dump the trash off; most of the pollen will stay on the glass. Leaving the trash on will probably not hurt anything if you pick out the bugs.

Using a single-edge razor blade, scrape the pollen together. If your catkins are good, you will collect small piles of brilliantly yellow dust (pollen). Scrape these into new or carefully cleaned vials. The vials should seal tightly, and he suitable for long term storage. Don't forget to label them. Repeat the collection procedure until you have the pollen you need. One group of catkins may he good for 2 or 3 collections before you harvest another group.

e) *Preserving pollen: As* quickly as possible, (not more than a few hours if unrefrigerated) put the opened vials into a tightly closed jar, at room-temperature, with silica gel or calcium chloride. (Silica gel can he bought at craft/hobby stores; it is used to dry flowers. Calcium chloride is used as sidewalk de-icer.) About a half a cup of fresh drying agent, placed loose in the bottom of a 1 to 2-quart jar, should dry

several vials of pollen nicely. Make sure the vials can't tip over, and don't mix pollen from different trees in the same drying jar. Dry the pollen for at least 4 hours, more if there is a lot of pollen, but not more than 24 hours. After this the pollen can he safely mailed to other pollinators. Pollen should simply he refrigerated if it is to he used in the next week or so or frozen at OûF if it is to he saved for next year. Do not freeze fresh (undried) pollen. Do not store fresh pollen in high humidity or at room temperature any longer than absolutely necessary.

3. When To Pollinate

The long, creamy, bottle-brush catkins are male (staminate); anthesis means "pollen shedding." The little green female flowers are found a! the tip of the branch, and the receptive stigmas are pinpoints at the very tip of the finger-like styles. Pollinate your trees when most of the flowers look like this:

(missing picture)

4. Pollen Testing

It does no good to pollinate your tree with dead pollen. It is best to test pollen if possible, but you will need a microscope. Chestnut pollen is easily germinated if floated on drops of 1% table sugar (or glucose) in nonchlorinated water, and held at 85û-90û F for one hour. Examine under a microscope at lOO-4OOX magnification. Good pollen should show l5-60% of the grains with tubes (as long as the grains) growing out of them.

5. Pollen Application

The *only* receptive part of the female flower is at the very tip of the style; the pollen must be put there. Chestnuts have one of the smallest stigrnas (receptive areas) of any tree. [See R.E. Harrison, "Determining the location of receptive stigmatic surfaces on page 111 of this issue.]

Pollen can be applied with a camel hair brush, a fresh or correctly dried catkin, or with my (current) favorite glass slide. To use the first two, just drag a catkin or pollen loaded brush over the stigmas. The glass slide is a little more complex to use, but has the outstanding advantages of using an absolute minimum of pollen, and of putting pollen exactly, certainly, and visibly, right on the stigmas. The following describes using slides in detail, but parts of it are relevant if you are using other methods, too.

a) Get several clean glass microscope slides. Clean them with lens paper before using.

b) If using preserved pollen, separate your pollen supplies into "stock" and "working" vials. You will probably drop your working vials, and slides, in the process of working through the branches, and it would be disastrous to lose your entire supply. Keep your stock secure.

c) Take a clean slide, open your working pollen vial, and cover the mouth of the vial with the slide. Holding vial and slide tightly together, turn the vial upside down, and shake pollen onto the slide. Turn the vial right side up, shake all the pollen you can *off* the slide and back into the vial. A film of pollen will remain on the slide, which is now "loaded". Recap the vial.

d) Carefully remove the bag from the branch to be pollinated. *The bag will have to be replaced immediately when you are through pollinating.* If it is in good shape, the same bag can be reused, otherwise a new bag will have to be put on.

e) Take the loaded slide, and drag it lightly over the stigmas of the flowers. Try to hit *all* the stigmas. 2 or 3 drags are plenty; more might conceivably damage the stigma. You may be able to see the stigmas leaving "tracks" in the pollen film. *BE SURE you touch the LOADED side of the slide to the flower!* This seems obvious, but the pollen film is nearly invisible, and it is easy to lose track of which side has pollen, even in full sunlight. A mark with a crayon or grease pen will help (or put pollen on both sides).

f) After 5 to 10 flowers, reload the slide with more pollen. After 50 to 60 flowers, get a new slide or

completely clean the old one. It is likely that the old one will be accumulating "self" pollen from your tree, which might interfere with fertilization. If possible, repollinate the tree in 4 to 6 days, since this can increase the number of "takes."

6. Labels

Branches should be labeled with information about what pollen or treatment was applied. Aluminum or waterproof cardboard tags can be used. Both tend to disappear over the summer wind, birds, and curious people take them off. Write a description of what branch was pollinated with what in your notes, as a backup.

7. Controls

Always remember to leave a few of your branches as "controls": Bag them, take off and replace the bag when you pollinate, but *don't* pollinate them. If they develop nuts, there has been a slip somewhere.

This is important. Don't neglect it.

8. Removing Bags

Remove the bags as soon as pollen is no longer shedding from any of the nearby chestnut trees; or if that is too long and you are worried about heat damage to your flowers, when all the male catkins of your tree have dropped.

9. Wait

You are done pollinating; now you have to wait and see if it worked. Chestnuts are frustrating to pollinate because the bur will develop whether or not it has been successfully fertilized. You probably won't be able to tell if the burs have good nuts in them until a week or so before they drop they stay green longer than empties, and get a little bigger at the end. Good luck!

Acknowledgments:
Special thanks to Dick Jaynes, Mark Wiilrlechner, and Fred Heburd

POLLINATION AND PLANTING

Locating Flowering American Chestnut Trees

Frederick V. Hebard

There are several ways to generate a breeding population of American chestnut trees. You will be able to order trees from us and grow them in your backyard or in an orchard, or you can go out in the woods and find some flowering American chestnut trees. The advantage of the second approach is that you can start breeding immediately rather than waiting for the trees to grow. This article describes facts of chestnut biology pertinent to locating flowering trees.

The American chestnut tree is very common in many parts of its range. I estimate there are between 100 million and 100 billion living sprouts. As Eyvind Thor said, "It must be almost as common as the passenger pigeon was once." Like the passenger pigeon was once, the American chestnut also is threatened with extinction, in this case by the blight disease.

Most American chestnut sprouts are located in wooded areas of the Appalachian mountains and foothills. Fre quently, they will be found just below the top of a hill or on mid-slope benches. Even a small hill only 50 to 100 feet high can have chestnut sprouts on it. When you do locate a chestnut sprout, look directly uphill and downhill for more. Remember that chestnut trees spread by dropping chestnuts, which tend to roll downhill.

Sprouts are not found at all the sites where the tree was originally located. They disappear when a woodlot is grazed continuously by cattle or browsed heavily by deer. Chestnut also tends to be absent from moist coves characterized by rhododendron and/or tulip tree. However, it is common on dry sites characterized by mountain laurel and huckleberry. Chestnut also prefers welldrained, acidic soils, such as sand; you will tend to find more chestnut around red and scarlet oak than around white oak.

Chestnut oak is a distinctive marker for American chestnut. The sharply defined, broadly-angled bark ridges of chestnut oak make it easy to spot. When you enter an area with chestnut oak, American chestnut sprouts frequently will be hiding somewhere nearby.

You may wonder if these chestnut sprouts are surviving because they are resistant to blight. on the contrary, almost all of them are quite susceptible. They are merely escaping infection by the blight fungus because they are small. In mature forests, chestnut sprouts grow under the shade of larger trees of other species. The lack of light keeps them small to the extent that a 1-inch diameter sprout, 6 feet tall, can be over 40 years old. Pencil-sized sprouts with only five or ten leaves can be 20 years old. Most small trees in mature forests escape infection by the blight fungus because there are not enough spores to infect very many of them The blight fungus produces spores in cankers, or lesions on the bark of stems. The surface area of a canker determines the number of spores that can be produced, and the circumference of a stem limits the maximum area of a canker. Additionally, smaller stems are encircled and killed more rapidly by blight cankers than larger stems; after a stem is killed the fungus stops producing spores. On small sprouts in mature forest, the blight fungus does not produce enough spores to create more than one new canker on the surrounding sprouts.

When chestnut sprouts become exposed to full sunlight, they grow rap idly; their diameter can increase 1/2 inch per year and their height 2 to 5 feet. In cutover areas, 10-year-old sprouts are 4 to 5 inches in diameter. Cankers on such large sprouts are ten times bigger than cankers on small sprouts growing in the shade of mature forest, and the blight fungus in each canker produces ten times as many spores able to form new cankers. Consequently, almost all large chestnut sprouts in cutover areas are blighted by 10 years after cutting. In contrast, only 20 percent of small sprouts in mature forest have blight.

But sunlight does more to chestnut sprouts than nourish their growth. It also encourages them to flower. In addition, blight itself stimulates flowering in chestnut. Thus many sprouts in cutover areas flower between 6 and 10 years after cutting, at a relatively young age. That is where we collect much of our reed of native chestnut and where we do much of our pollinating. When pollinating in cutover areas, you do have to avoid trees with advanced blight, as they may die before bearing. 'hey generally are characterized by a superabundance of early flowering catkins and burs.

If there is a federal or state forest in your neck of the woods, the foresters there should be glad to direct you to cutover areas. This also holds true for wood lots owned by private companies. Be sure you have permission from the land owner before you start pollinating!

One also can find flowering American chestnut trees in areas where gypsy moths have killed most of the oaks, between 5 and 20 years after the devastation. Gypsy moths do not kill all the trees in a mature forest, so most chestnut sprouts are not exposed to full sunlight. Consequently, they grow more slowly than sprouts in cutover areas, and the blight does not spread as rapidly, leading to longer lives for the flowering trees. Unfortunately for us, the trees also tend to be quite large before they flower, making them more difficult to pollinate than trees in cutover areas. The lower amounts of sunlight and blight are the probable causes of this delayed flowering.

In Connecticut, where most areas are cut selectively rather than clearcut, chestnut sprouts also persist for long periods, in a fashion similar to areas devastated by gypsy moths. The patchy nature of the amount of sunlight reaching chestnut sprouts in selectively cut and gypsy-moth-damaged areas leads to great variability in tree size. In Connecticut, it is not uncommon to find 1-inch-diameter trees growing near trees 8 to 10 inches in diameter. In comparison, most trees in cutover areas are killed by blight before they reached such a large size. Only rarely is there enough light in undisturbed forests for sprouts to attain diameters of more than 3 to 4 inches.

A final place to locate flowering American chestnut trees is along roadsides and power lines in wooded areas. Invariably, the agency in charge of the right of way has to clear out encroaching trees. As indicated above, this can lead to rapid growth and flowering of chestnut trees. Be advised that flowering chestnut trees are not frequently encountered along roads, so be prepared to do a lot of driving if you chose this last approach. The best roads to search may be those which have been constructed, widened or cleared 5 to 15 years previously.

Flowering American chestnut trees arc most easily located while they are in full bloom, in the weeks around the Fourth or July. The great mass of conspicuous white catkins on larger trees is visible at great distances. The odor is also quite distinctive, especially on still mornings and evenings. Later in summer, bur-laden trees are fairly obvious. In early fall, chestnut leaves turn yellow' sooner than the leaves of many other deciduous trees with yellow leaves. In late fall, the brown leaves tend to stay' on the trees. To locate flowering trees in fall and winter, look on the ground underneath them for fallen burs. In spring, look for the thin green catkins which emerge soon after the leaves.

American chestnut is not the only species of chestnut you will find in the woods. Chinese, Japanese and hybrid chestnut also have been planted in many areas of the country. Dr. Phil Cordon wrote a key to chestnut species for the last issue of this Journal, Volume V, Number 1. If you are unsure of the identity of your trees, send leaf samples to the ACF Wagner Research Farm, Rte. 1 Box 17, Meadownview, VA 24361. In winter, you can send brown fallen leaves collected from around the base of trees.

Chestnut Pollen Collection and Handling

Dr. Charles Maynard
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[Editor's Note: The Foundation's external grant program is an aggressive, many-sided eftort to hasten the American chestnut's return to the wild forest. Those who receive ACF grants are asked to write a report on their results for consideration by this jounal. This report describes some of the many ways a competent scientist's work can be frustrated by chestnut pollen's fragile nature.]

Introduction

With the development of a back-crossing strategy for breeding blight-resistant chestnut trees by hybridizing

American chestnut *(Castansa dentata)* with Chinese chestnut *(Castanea mollissima)* and then using *C. dentata* as the recurrent parent (Burnham 1982, Rutter and Burnham 1982), interest is re-emerging in methodologies for making controlled crosses in chestnut. Because this interest, like the remnant populations of American chestnut, is scattered across the world, methods for easy and reliable collection, storage and shipment of chestnut pollen are needed. This report presents a series of preliminary studies attempting to develop these methods.

Much of what we learned happened by accident rather than design, and thus represents uncontrolled observations rather than the planned studies outlined in my original proposal. Much of what we learned was also negative results and thus probably would never see the light of day without the American Chestnut Foundation and its *Journal.* My presentation at the 1988 Chestnut meeting was subtitled "A Hundred and One Ways to Kill Chestnut Pollen." I think this summarizes what we learned very well.

Nevertheless, knowing what can go wrong is very valuable in an applied breeding program and this report should provide a starting point for further research.

POLLEN COLLECTION AND STORAGE STUDIES

Pre-flowering collections

The author has successfully used the cut-branch forcing method to collect viable pollen from various *Populus, Picea,* and *Pinus* species as much as three months before normal pollen shed. The major difference between these species and *Castanea* is that flower buds are formed the year before pollen shed rather than on the new growth.

Objectives

To determine whether the cut-branch forcing method would work on *Castanea dentata,* allowing out-of-season collection of pollen.

Procedures

Five branches approximately 2 feet in length were collected at approximately weekly intervals beginning in early June, 1988 until natural pollen shed in early July. Cuttings were placed in plastic *bags,* stored in a cooler with crushed ice, and upon return to the laboratory, placed in water. A fresh cut was made on the end of each cutting every three days until anthesis or catkin abscission.

Results

At the first collection dates, the anthers were completely closed and hard. When placed in water, they would elongate but did not open. Eventually, the cut branches began to dry, despite repeated fresh cuts, and the catkins died.

At later collection dates, especially as natural anthesis approached, anthers on cut branches could be induced to shed pollen, but only in extremely small quantities and not more than a week before normal

pollen shed.

Further research with various forcing solutions might be fruitful, but the complete lack of success indicates that a useful technique will require a great deal of effort. I think that emphasis should be placed upon improving storage techniques.

Comments

Freezing injury to fresh anthers: Because of a refrigerator malfunction, an inadvertent pilot study was conducted on the susceptibility of freshly collected anthers to mild freezing. Freshly collected dehiscing anthers were stored overnight in a refrigerator set at slightly below O°C. When the anthers were spread out to dry the next morning, no freezing injury was noted, but tetrazolium staining of the freshly collected pollen showed zero viability. We know the pollen was viable when collected, because we processed part of the anthers the same day they were collected and shipped them to Dr. Mark Widrliechner at the plant Introduction Station, Ames, Iowa. His tetrazolium test and his use of the pollen in controlled crosses both showed good to excellent viability.

This indicates that freshly collected catkins must be handled carefully. Even our ice-filled cooler used to transport the anthers back to the lab may have caused some damage to those catkins in direct contact with the crushed ice.

Drying injury to fresh anthers: In another collection, fresh anthers were spread out to air dry on laboratory benches at a temperature of approximately 28°-30°C. The catkins were left overnight and pollen collected the following morning. As in the inadvertent freezing study, tetrazolium staining showed zero viability. Subsequent collections were air dried for a minimum of two and a maximum of four hours before refrigeration or frozen storage.

Initial air drying recommendations: We found that freshly shed pollen cannot be frown immediately, but rather requires a period of air drying. We also found that overnight drying, at least at the high temperatures we used, was too long. At this time I would recommend a minimum of two hours and a maximum of four.

Desiccator cap containers: The perishable nature of chestnut pollen is amply demonstrated by the two catkin drying studies described above. We felt it desirable to design a storage container for the pollen that would maintain a low moisture content in the pollen while being shipped, and if possible even while the pollen was being used for pollination work. Several years ago I purchased some drying containers called 'Top Hat Desiccators' (D-Hydro Container Corp., Hauppauge, New York). They are screw-on lids that fit on 100ml brown-glass bottles and have a dessicator compartment built into the lids. They were too large for the few grams of pollen we would be typically shipping, but the idea was appealing. We found that several different sizes of vial lids are actually molded in two pieces and snapped together. We found that a hot wire could be used to burn small holes in the bottom of the cap and that approximately 1/2 to 1 g of the granular dessicant "Dryrite¨" could be placed in the cavity.

Down-sized shipping containers: We also felt that that the large shipping containers we had used in the past were excessive. These foam-lined cardboard boxes have approximately 1/4 cubic foot capacity and even with several refrigerant packs, require a great deal of packing material simply to fill the empty cavity around the few vials typically included in any one shipment.

I obtained a catalog from a manufacturer specializing in shipment and storage of perishable materials and found that indeed, small shipping containers with built-in ice compartments are available (Polyfoam packers Corp., Wheeling, Illinois).

Freezer storage vs. refrigerated storage vs. ambient temperature storage

Storage of pollen from season to season is a common procedure in many plant species. It allows for more careful planning of crosses to be made, based upon known quantities of pollen from known trees. It allows pollen to be shipped in cool weather rather than potentially riding around for a day in the back of a scorching mail delivery van in 100° weather. It also allows pollen to be collected from late-flowering trees

or regions and used to pollinate trees in early-flowering regions the following season. Unfortunately, pollen viability is usually lost with extended storage.

Objective

To develop a successful storage method for *Castanea dentata* pollen.

Procedures

In early July, 1988 large quantities of dehiscing anthers were collected from *Castanea* trees located on the property of Mrs. Earl Douglas, Red Creek, New York. We collected approximately 1/4 to 1/2 a grocery sack full from each of six different trees. As described earlier, we split open the paper bags and spread the catkins out to dry for 2 to 4 hours. The anthers could then be removed by rubbing the catkins between the fingers while holding them over a piece of glass. The pollen/anther mix could be collected by scraping the glass with a single-edge razor blade. We then weighed and subdivided the pollen/ anther mix into small preweighed vials approximately 200-300mg per vial. The total weight of vial plus pollen was recorded and vials from each clone were assigned to four different treatments: oven drying, air drying, refrigerator drying, and frozen storage drying. (The oven drying procedure was not intended as a preparation method for storage, but rather to obtain estimates of the initial moisture content of the samples.) Oven drying took place at 90° C for 24 hours while air drying took place on a lab bench at ambient temperature and humidity (approximately 28° C and 75-80% RH) for 24 hours. Samples to be dried under refrigeration were capped with the previously described dessicator caps and placed in a refrigerator at approximately 5° C. Samples to be frozen and dried simultaneously, were placed in a dessicator in the freezer compartment of a standard household refrigerator at approximately –10° C. After 24 hours, vials were reweighed and the moisture loss or gain was calculated.

After one year, samples of refrigerated and frozen pollen were removed from storage and tested for viability. A sample of the pollen/anther mix was spread over the surface of a glass slide and stained with tetrazolium. A sample was also dusted onto the surface of a petri dish containing 1% agar to test germination.

Results

As expected, the four different treatments significantly influenced the moisture status of the pollen. Somewhat surprisingly, differences among clones were also highly significantly different (Table 1). The treatment-clone interaction was also significant, but because any comparison among drying treatments and clones would be based on only two degrees of freedom, I felt it better to simply report that the interaction was significant but not try to explain it.

These results again point up the ephemeral quality of chestnut pollen. Twenty-four hours of air drying resulted in near-total loss of moisture and presumably a parallel loss of viability (Table 2). Immediate freezing of samples may also be inadvisable. It appears that freezing samples immediately after collection and extraction of anthers from the catkins slowed evaporation to the point that there was no drying of the samples taking place.

Interpretation of moisture loss differences among clones is also problematic (Table 3). Because all of the samples for this study were taken from a single collection date, it is impossible to determine if these moisture loss differences among clones reflect differences in maturity of the catkins collected during a particular week, or if they reflect true clonal differences. If the latter, it could explain the commonly observed differences in pollen viability and longevity in storage among clones and would make it imperative to test the moisture content of each freshly collected sample.

RECOMMENDATIONS

Cut-Branch Forcing

The cut-branch method will not work for forcing early pollen shed in chestnut.

Catkin Collection

Collect as catkins begin to shed through maximum anthesis.

Collect white to light-cream-colored anthers. However, even browning catkins will have some live anthers.

Collect lots of catkins, since a grocery bag of catkins may yield less than 25 grams of anthers.

Catkins shed over an extended period when attached to the tree but dry out quickly when detached.

The moisture content of fresh catkins is greater than 75%. They are, therefore, highly subject to freezing, which will kill the pollen. They are also highly subject to desiccation, which will also kill the pollen.

Pollen Handling

Fresh anthers collected by rubbing catkins still have a very high moisture content and cannot be frozen directly.

Fresh pollen will not dry properly at sub-freezing temperatures even if stored over dessicant. It must be air dried first.

Fresh pollen is highly subject to desiccation and cannot be air dried for more than a few hours or it will die.

Pollen Storage

If at all possible, use pollen within a few days of collection.

If storage is essential, make sure the pollen has been air dried to 5 to 7% moisture content (a best guess), refrigerate overnight over dessicant, then place in freezer.

Best storage technique is to use many small vials, each containing only a few milligrams of anther/pollen mix and each with its own dessicant cap. Store multiple samples of pollen from a single male parent together.

When removing pollen from storage, remove only one vial from each pollen source and immediately replace the remaining vials in freezer. (Repeated freezing and thawing has been shown to reduce long-term viability of seeds. I assume that pollen is at least as subject to damage.)

Pollen Shipment

Remove anthers from catkins and place in vials with dessicant caps.

Pack in insulated shipping cartons with gel refrigerant packs (Bluelce¨ or similar) and foam 'peanuts.'

Ship immediately via overnight mail.

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Table 1

Analysis of variance of moisture loss as affected
by different drying conditions and clones.
(r2 = 0.93, C.V. 33.36, overall mean moisture loss = 4.76%)

Sources of variation	Degrees of freedom	Sums of squares	F	PR > F1
Treatment	3	1117.20	147.39	0.0001
Clone	6	118.92	7.84	0.0001
Treatment-clone	18	96.53	2.12	0.0253
Error	38	96.01	2.53	
Corrected Iota!	65	1428.68		

1 The probability of an F value M this size occurring by chance.

Table 2
Mean moisture loss or gain upon drying
for chestnut pollen/anther mixes.

(Moans followed by the same letter do not differ at the 0.5% probability level, based upon Duncan's multiple range test.)

Treatment	N	Moisture loss (% of fresh WI.)
Oven dry	13	9.6a
Ambient	13	9.0a
Refrigerated	26	3.5b
Frozen	14	1.4c

Table 3
Percent moisture lose (trash weight basis) among
7 clones and 4 drying treatments for cheatnut pollen

Clone[1]	Oven	Air	Refrigerator	Freezer	Overall Mean
A	11.2 (0.38)2	9.1 (-)	5.5(0.12)	-2.1(0.64)	5.5
Al	5.9 (0.17)	9.1 (0.23)	2.4(0.21)	-1.2 (0.14)	3.9
B1	11.3(0.72)	9.7(0.05)	3.1(0.13)	-1.0 (0.37)	5.5
D	14.3 (2.27)	8.4 (0.43)	4.4 (0.35)	-0.9 (0,04)	6.1
El	5.4(5.4)	7.8(0.24	1.9(0.36)	-1.9(0.14)	3.0

F	8.1 (1.6)	7.2 (0.55)	1.9(0.14)	-1.7 (0.04)	3.5
G	12.6 (-)	12.0(2.69)	5.1(0.13)	-1.1 (0.32)	6.1

1 Clones are located on the property of Mrs. Earl Douglas, Red Creek, NY. They are a mixture ot Fl hybrids between American and Manchurian chestnut and backcrosses to the American parent. A map showing the full pedigree of each tree woo not available

2 Numbers in parentheses are standard errors of each mean.

Determining the Location of the Receptive Stigmatic Surfaces in Castanea dentata Borkh.

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One of the main goals of the American Chestnut Foundation is to develop American chestnut trees resistant to Endothia *parasitica,* the devastating fungus which has blighted *Castanea dentata* Borkh. to near extinction. Modern plant breeding procedures are currently used in the development of resistant trees. However, problems with simple procedures often stand in the way of progress. Such problems include the collection of pollen and the making of controlled pollinations.

The quantities of viable chestnut pollen available to the breeder has been small due to the difficulties of obtaining pollen from trees. Moreover, current pollination practices use relatively large amounts of pollen for each cross. The pollination procedures involve dusting pollen over the entire bur, or female flower structure, in order to insure that the stigma surface is covered. The precise location of the receptive surface in *C. dentata* has not been clearly documented in the literature. Locating the receptive surface will enable the breeder to devise improved methods of pollination.

Heslop-Harrison et. al. (1977) reported on the characteristics of receptive surfaces of nearly 100 species. Included was a description of Fagaceae, including *Castanea,* as having a dry, non-papillate, stigma with the receptive cells concentrated in distinct ridges, zones, or heads. *Castanea* was also used as an example of extreme restriction of the receptive stigma area to a very limited number of cells (Heslop-Harrison, 1984). Each of these articles mentioned *Castanea* only briefly with no reference to individual species.

Mattsson et al. (1974) found that stigmatic surfaces have an external proteinaceous coating. This coating, referred to as the pellicle, has certain properties which led the researchers to believe it important in capturing and hydrating pollen grains. They also found the pellicle was consistently associated with esterase activity (Mattsson et al., 1974). They used the non-specific esterase staining procedures of Jensen (1962) to determine the presence or absence of enzyme activity.

The purpose of this study was to look specifically at *Castanea dentata* to determine the location of the receptive area of the stigma, when receptivity is initiated, and its duration.

Materials and Methods

Three pure American chestnut *(C. dentata)* specimens, located at the University of Minnesota Landscape Arboretum, were used to supply the female flowers for this study during the summer of 1988. Just prior to anthesis, white ear bags (Lawson No. 421, used for corn pollination) were placed over the stems containing selected burs to prevent pollination. All catkins which would have been inside the bags were removed.

Anthesis began on approximately June 17 and continued until July 15. Four weekly bur collections were made. The first collection was made at the beginning of anthesis. At this stage the styles (finger-like projections) were just beginning to emerge from the burs, and most of these were between 0.5 and 2 mm in length.

The next collection was made during anthesis. The stigma and styles had pushed further out of the burs and most of these were between 2 and 4 mm in length.

The third collection was made near the end of anthesis, and the fourth one week following anthesis. The development of the stigma and styles were similar at each of the final two collections and were all greater than 4 mm in length. For each collection, stems containing the selected burs were removed from the trees, placed into water, and moved to the lab where the stigmas were tested for esterase activity.

The top one third of each bur was removed with a scalpel. The object was to remove the majority of the bur tissue, but leave enough tissue attached to the stigmas and styles so that the structures would remain in their natural positions.

The terminal one third of each bur, including the attached styles, was removed and treated either by submerging it into an esterase stain or into a control treatment of distilled water for 10 minutes. The esterase stain used in this study was a colorimetric stain used for electrophoresis (Cheliak and Pitel, 1984). The stain contained 25 ml 0.25 M phosphate buffer adjusted to a pH of 6.4; 25 mg each of a- and b- naphthyl acetate dissolved in 2.5 ml of acetone; and 50 mg of fast blue RR salt. Following the staining procedure, the styles were rinsed in distilled water and observed under a dissecting microscope at 60X and 250X. The tissue with esterase activity was stained dark blue or purple.

Results

All styles from the last three collections during anthesis, near the end of anthesis, and one week following anthesis responded positively when treated with the esterase stain.

In every case the area that stained positive for esterase activity was limited to a small point at the top of each style. A few of the styles from the first bur collection stained positively, but most did not. Those with no esterase activity were just beginning to push out of the bur tissue and were less than 1 mm in length. No color changes were detected on the stigmas in any of the control treatments.

The data suggests that the pellicle appears to be secreted 24 to 48 hours after the styles break through the surface of the bur or when the structures are approximately 1 mm in length. Once the esterase activity had begun it showed no signs of diminishing throughout the collection period. Although no data was taken later than one week following anthesis, it is probable that the esterase activity continued until desiccation of the styles occurred.

The reactions of *Castanea dentata* in this study appear to be consistent with those found in *Castanea* by previous researchers (Heslop-Harrison et al., 1977; Heslop-Harrison, 1984). It is likely that similar staining patterns would be found throughout the genus. The location of the receptive site in this experiment also reinforces the observations of Velguth (1987) in which pollen tubes were noted entering the stigmas of *C. dentata from* only the tips of the styles.

Conclusions

In this study of *Castanea dentata* flowers, the stigma pellicle was secreted soon after the stigma-style structures began to develop outside the bur tissue. Once these structures had emerged approximately 1 mm from the burs, all styles stained positively for esterase activity. This suggests earlier receptivity of the stigmas than originally thought.

Previous studies have concluded that the pellicle is involved with capturing and hydrating pollen grains; therefore it may follow that the receptivity of the stigma would be closely associated with the secretion of the pellicle. However, this proposed association of the pellicle secretion and receptivity was not tested in this study. This research also found esterase activity, believed to be associated with the receptive site of the stigma, to be restricted to the extreme tip of each style in *Castanea dentata.* These findings indicate no reason for breeders to pollinate any portion of the style except the tip.

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Growing Chestnut Trees from Seed

Frederick V. Hebard, Philip A. Rutter

Growing chestnut from seed is easy. If you can germinate one avocado in twenty tries, you will be able to germinate at least fifteen chestnuts. The nuts are relatively big, and seedlings grow fast, so most survive the rigors of their first winter. The six essentials are proper storage before planting, an appropriate potting mix for planting, adequate protection from rodents and deer, no weeds, good fertilization, and watering.

Storing Nuts

Chestnuts may be planted in the fall, as soon as they drop, or stored over winter in a refrigerator.

If freshly husked nuts are wet on the outside, they should be dried at room temperature for no more than 2 to 3 days after harvest. Then they should be placed in moist, but not wet, peat moss in plastic bags or buckets. Moisten dried, milled peat moss with 2 to 3 cups of water to 1 gallon of moss. Numerous holes should be placed in the plastic bag with a toothpick so the nuts can breathe. Each nut should be completely surrounded by peat moss, not touching other nuts or the sides of the bag. The moss keeps the nuts moist and retards spoilage. The bagged nuts can be stored at 320 to M^{0} F (near the top of the refrigerator) for 2 to 3 years. They require 2-3 months of cold before they will germinate.

Planting Nuts Indoors

Planting chestnuts indoors in a warm (700 to 800 F), sunny window or green-house ensures the highest rate of seedling establishment. Do not let the nighttime temperature drop below 600 to 650 F. Nuts should be planted in the potting mix described in Table 1. Commercial potting mixes generally do not provide enough aeration, so nuts and seedlings frequently rot in them, especially with too much watering.

The best pots for planting are 1- or 2-quart cardboard milk cartons with the bottoms removed. This ensures air-pruning of deep roots and adequate root aeration. The cartons can be placed on window screening on top of hardware cloth (rat wire) in milk crates to prevent the potting mix from falling out the bottom. Moistening the mix before putting it in the cartons also helps prevent it from falling out the bottom.

Nuts should be planted on their sides ~_ to 1 inch deep in the mix. The easiest method of fertilization is daily watering with MirAcid or a similar soluble azalea-rhododendron food that contains micro-nutrients. Follow the package instructions for amounts to use.

Nuts can be sown in late March for transplanting in mid-May. Earlier sowing can lead to overcrowding of seedlings in milk crates, requiring more space than one window can provide. Seedlings should be hardened off by placing them outdoors in shade (under a tree for instance) for several days before transplanting. To avoid sunscald, cover the leaves with a moderate or light cover of hay for two weeks after transplanting and remove the hay gradually over the next two weeks. if the hay gets wet or mold sets in, pull it back immediately. Do not transplant before the last frost-free date for your area. This is mid-May in southwestern Virginia. Additional information follows.

Planting Nuts Outdoors

Like potted seed, nuts should be planted outdoors 1/1 to 1 inch deep, on their sides. They must be protected from rodents.

Seedbeds. As mentioned above, nuts may be planted outdoors in the fall, right after harvest. The main danger of this method is freeze-killing of nuts during cold winter weather. Chestnuts die at temperatures much below freezing. Fall seed-beds should be covered with 6 to 18 inches of mulch to keep them warm. The covering mulch should be removed in April.

Moist peat moss (see "Storing Nuts" above) or compost are good seedbed soils. You can economize on their use by planting chestnuts in rows spaced 3 to 6 feet apart. Line the bottoms of trenches with an

inch or two of moss or compost, lay the nuts 6 to 12 inches apart and cover with a half inch of moss or compost. Cover the moss or compost with soil and pack it down to prevent it from being blown away by wind or heaved by frost. You want the top of the trench to be level with the surrounding soil. To ensure proper aeration, it is necessary to place the seed-bed in well drained soil or to raise it. However raised seedbeds can be vulnerable to freezing, especially along the edges.

Seedbeds can be protected from rodents by fencing them with quarter-inch mesh hardware cloth, sunk 6 to 12 inches into the soil. Hardware cloth also can be laid on top of the beds, but it must be removed before germlings become immeshed in it. Enterprising rodents may still tunnel underneath. If birds raid a seedbed, it must be covered with hard-ware cloth or netting.

Seedlings are generally transplanted from seedbeds after one or two years.

Direct Seeding. At the farm in southern Virginia, we sow most nuts directly at orchard spacing. We will plant more than 2000 nuts next year, and other methods are not feasible with our limited resources, especially if we have to water transplants during pollinating season.

In order to avoid deep winter freezes, we start planting in mid-February. In our climate, there can be significant root development beginning in the latter half of March. Because of those deep roots, seedlings of February-planted nuts that emerge in mid-May can tolerate drought well. We try to plant before March, before nuts germinate, as ungerminated nuts seem to develop better seedlings. But nuts can be planted successfully until mid June.

At Meadowview we plow, disk and fertilize orchards before planting. More details follow.

Rodent and Weed Control

We protect directly sown nuts from rodents by placing a 3-inch diameter cylinder of 1~inch aluminum flashing around the nuts. We make the top of the cylinder slightly smaller than the bottom (about _ inch) to reduce internal reflection of sunlight onto the leaves; they are slightly cone shaped. The cylinder is sunk in the ground 1 to 2 inches. We mound soil around the cylinder to keep it from blowing away in heavy winds. The cylinder is covered with a paper or Styrofoam cup until May to exclude rain and to keep the nuts at soil temperature. Holes are placed in the cup for aeration. We try to remove the cups on a cloudy day or in the evening so the leaves are not suddenly exposed to full sunlight.

Before planting, a plug of soil is removed with a handled bulb planter and the hole filled with moist peat moss (see "Storing Nuts" above). This keeps the nuts moist and free of mold and prevents weed growth inside the cylinder.

Transplants can be protected from rodents with a cylinder of quarter-inch hardware cloth. The cylinders should be sunk 2A inches into the ground and extend 18 inches above ground. For nuts, we use aluminum flashing instead of hardware cloth to keep our winter-planted nuts warm, and to protect them from wind. Such cylinders are also less expensive.

In areas where deer are a problem, 4-or 5-foot tree shelters, such as those sold by Tubex (75 Bidwell St., Suite 105, St. Paul, MN 55107), should be used for transplants and directly sown nuts. Alter-natively, trees can be protected with 4- or 5-foot-tall cylinders of hardware cloth. Either tree shelters or hardware cloth must be tied securely to strong stakes to prevent deer from tearing them out to reach the nuts and seedlings. Nuts sown directly in tree shelters or hardware cloth cylinders should be planted in peat-moss-filled holes larger than the shelter, to prevent weed growth inside it. It may be best to wait until mid-April to sow nuts in tree shelters, as the warm shelter can lead to early germination, with consequent risk of frost damage.

Weeds and grass must be kept at least 2 feet away from seedlings, and clear out to the ends of branches in larger trees. This can be done manually, with mulches or with herbicides such as Round-Up plus Princep. Be sure to protect the trunk with an open stove pipe or similar device when applying herbicides; plants in tree shelters are already protected. Mulching not only controls weeds, but also helps retain soil moisture.

Either plastic or organic mulches can be used. We use plastic mulches at the farm because of our large number of plants. However, plastic mulch does impede fertilization of trees and can harbor rodents, so there are drawbacks to its use. Mulches should be cleared 6 to 12 inches from the base of trees in fall and winter to reduce rodent damage.

Fertilization

Chestnut prefers well drained, acid soils. It has the same soil preferences as blueberries and azaleas. At soil pHs above 6.5, nitrogen frequently is limiting.

Most transplants should not be fertilized their first year, but if any leaves start turning yellow before early August, immediately apply 1/4 LB of 10% nitrogen. A foliar spray of soluble fertilizer such as MirAcid or fish emulsion will restore leaf color quickly while the ground-applied fertilizer is being absorbed.

Seedlings from nuts planted outdoors can be fertilized heavily the first year. Fertilize transplants that have been in the ground for a year and seedlings with a 10-10-10 or 2~2~20 N-P-K ratio at lower soil pHs. At higher soil pHs, a 20-6A or 30-1~ 10 ratio is frequently satisfactory. This often can be found premixed with instructions as azalea and rhododendron food; either granular or liquid formulations (such as MirAcid) can be used. Organic gardeners can use fish emulsion as a nitrogen source.

As a general rule, apply 1 LB of granular fertilizer per inch of trunk diameter per year, half in the spring and half at the beginning of summer. Apply the fertilizer uniformly over the root zone, 6 inches from the base of the trunk to the ends of the branches. Follow the package instructions for fish emulsion and liquid fertilizers. These frequently are applied every 2 weeks until early August.

Watering and Transplanting

It is best to dig a hole at least twice as wide as the root system of the transplant, and to mix 1 part of compost or peat moss with 3 to 4 parts of soil. This promotes healthy roots and reduces watering needs.

Water transplants immediately after setting them. During dry periods, soak the soil around the tree once a week. It is best to keep watering for four years after transplanting. Watering needs vary, of course, with the weather. Soil conditions also influence watering needs.

A 10 by 15 foot spacing of transplants is suitable if space is tight. Wider spacing, such as 10 by 20 or 20 by 20 feet can facilitate artificial pollination when trees mature. Old orchards of Chinese chestnut probably should be spaced at 40 by 40 feet. Frequently, growers will start such orchards at 20 by 20 foot spacing, then thin later to 40 by 40 feet. Doing so increases yields during the early years.

Table 1

Chestnut Potting Mix

Ingredients: peat moss, vermiculite **and perlite.** Lime to pH 6.

12 quarts perlite
12 quarts vermiculite
12 quarts ground peat moss
2 ounces (114 cup) lime

Mix **well in a** wheelbarrow with a shovel. Sprinkle the lime over the mix to insure even distribution, **and** mix **well.** Moisten with water if dust is a problem during mixing.
It is important that pots have good drainage.

SCHOLARSHIP

Effect of Phosphorous on the Number of Pistillate and Staminate flowers, Diameter of Shoots and Yield of Chinese Chestnuts

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Abstract

The objective of this study was to determine the effect of phosphorus on pistillate and staminate flower number, diameter of shoot, and yield by Chinese chestnut (*Castanea mollissima* Blume).

Eight-year-old Chinese chestnut trees, 'Thong Chi Li' cultivar, were grown in sandy soils in Luo Tian County, Hubei Province, PRC. Phosphorus treatments ranged from I kg to 4 kg of triple super-phosphate in the soil per tree. Other treatments were 100 kg decomposed manure per tree and the control trees with no treatment.

Soil applications of phosphorus to Chinese chestnut trees significantly increased the number of pistillate shoots per staminate shoot, number of burs per pistillate shoot and nut yield.

Introduction

Chinese chestnut is a monoecious plant. Mostly there are large numbers of staminate flowers and few pistillate flowers on the same tree. According to our previous research on the relation between flower differentiation and nutrition of the chestnut tree (Zhang et al. 1985), the initiation and formation of pistillate flowers require higher phosphorus levels in the tree than for staminate flower or catkin formation. Generally the soils of Hubei Province are deficient in phosphorus, particularly in the leading chestnut production area of Dabei Mountain in Hubei Province, PRC. These soils are sandy and eroded. Therefore, soil mineral nutrition is poor.

Also we found that the differentiation and formation of pistillate flowers occur from April to June, which is too late to secure sufficient phosphorus from stored nutrients in the tree. Staminate flower formation and tree growth depleted the residual phosphorus in the tree prior to pistillate flower formation *(Chestnut 1979).* Soil test and leaf diagnosis indicate that less phosphorus in soils or in leaves results in fewer pistillate flowers on the trees and lower yields of nuts. In order to secure high yields, an increase in phosphorus content in the tree is needed. This research focused on increasing phosphorus content of the tree by removing male catkin shoots and weak vegetative shoots, and by applying phosphorus to the soil to increase the pistillate flowers per tree and secure high nut yields.

Methods and Materials

The experiment was conducted on a sandy soil in Luo Tian County, Hubei Province PRC. A randomized block design with three replications of three trees per plot was utilized for the experiment. The treatments were as follows:

1) Low phosphorus: 1 kg triple super-phosphate in soil per tree,
2) Medium phosphorus: 2 kg triple superphosphate in soil per tree,
3) High phosphorus: 4 kg triple superphosphate in soil per tree,
4) Medium phosphorus + pruning (pruning off male catkin shoots, weak vegetative shoots, crossed shoots etc.),

5) Decomposed manure: 100 kg in soil per tree,

6) Phosphorus solution: 2 kg triple superphosphate were dissolved in 50 kg water and applied in the root system areas of each tree, and

7) Control: no treatment.

All experimental trees were eight-year-old Chinese chestnuts. The variety was Thong Chi Li. In September, 1986, phosphorus was applied by digging a circular ditch 30 cm deep around each tree. Pruning was conducted in December, 1986. Prior to initiation of the experiment, the soil of each experimental block was tested (Table 1). During the experiment, N and P diagnoses were made in pistillate shoots, staminate shoots, previous mix shoots, vegetative shoots, and leaves. The bearing characteristics, burs per pistillate shoot, the rate of pistillate shoots per staminate shoot, and yield of each treatment were recorded.

Results and Discussion

The number of pistillate shoots was increased with applications of phosphorus (Table 2). When compared with the control (treatment 7), the pistillate shoots were increased by 1.8 times with application of 2 kg of triple phosphate in the soil per tree and pruning. In addition to raising the phosphorus concentration in the tree, this treatment also reduced the amount of phosphorus consumed by differentiation and development of staminate flowers in staminate shoots, and by other pruned non-bearing shoots. The other phosphorus treatments (Zhang et al. 1985, Jaynes 1979, *Chestnut* 1979) also increased the number of pistillate shoots per tree. The correlation between the phosphorus concentration of previous mix shoots and the rate of pistil late shoots per male catkin shoot illustrates the important role of phosphorus in differentiation and development of female flowers (Table 3). Since the differentiation and development of female flowers require higher levels of phosphorus than that for staminate flowers, higher phosphorus stored in the previous mix shoots produced more pistillate shoots.

Phosphorus treatment increased the number of burs per pistillate shoot (Table 4). The larger pistillate shoots preduced more burs than the smaller shoots. Among the phosphorus treatments, the average diameter of the pistillate shoots for one bur was 0A5 cm (0.42-0.48 cm), for two burs was 0.59 cm (0.560.62 cm), and for four to five burs was 0.66 cm (0.60-0.71) (Table 4). For the control treatment, the average diameter of pistillate shoot for one bur was 0.57 cm and for two burs 0.64 cm. Only one and two burs were found on pistillate shoots of the control. Bur numbers were increased on similar diameter pistillate shoots by the application of phosphorus, particularly by medium phosphorus treatment and pruning. The average number of burs per pistillate shoot in the medium phosphorus treatment and pruning was 0.33 more than that of the control (Table 5), offering an opportunity for increasing yields of chestnut.

Nut yield was increased with the addition of phosphorus to the soil (Table 5). Nut yields from phosphorus treatments were more than two times as high as the control. The medium phosphorus and pruning treatment increased the nut *yield* more than three times that of control. As indicated above, the number of pistillate shoots and burs per pistillate shoot were increased by phosphorus treatment. It is logical that higher yields among the phosphorus treatments when compared with the control (Table 5) were due to the larger number of pistillate shoots and burs per pistillate shoot in the phosphorus treatments. Differences of nut yields between soil phosphorus treatments and the Check are significant at the 1 % level, which demonstrates that phosphorus is a main factor for increasing yields in the phosphorus-deficient hill orchards.

No significant difference was found among Treatment 5 (decomposed manure), Treatment 6 (phosphorus solution) and the control, even when they raised the number of pistillate shoots per staminate shoot and number of burs per pistillate shoot. The lack of significance in these two treatments was due to low availability of phosphorus when the burs were developing in late season (2). In regard to significant differences among treatments 1, 3, 5, and 6, future research will be required to determine the most desirable level of phosphorus in the soil for maximum yield. Soil analysis indicated that 14.5 ppm of soluble phosphorus in the soil is essential for normal yields in hill soils of Hubei Province, PRC.

Summary

Soil applications of phosphorus to chestnut trees significantly increased the number of pistillate shoots per staminate, number of burs per pistillate shoot and nut yields. There was a 5% significant

positive correlation between phosphorus content of previous mix shoots and the ratio of pistillate shoots/male catkin *shoots.* Since the analysis of the shoots showed that the phosphorus content of the pistillate shoots was higher than that of staminate shoots, the differentiation and development of female flowers possibly require a higher level of phosphorus in the chestnut tree. This indicates that the main chestnut production area of Dabei Mountain region is seriously deficient in phosphorus (Table 1). Soil analysis showed that 14.5 ppm soluble phosphorus in the soil is essential for normal yields in Hubei Province (Chestnut 1979). These tests demonstrated that soil applications of phosphorus were essential for maximum yields of chestnuts on hill soils in Hubei Province, PRC.

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Table 1
Soil mineral nutrient content of experimental blocks
in Luo Tian County, Hubel Province.

	Block No.		
Mineral content	**I**	**II**	**III**
Total N%	0.02793	0.03430	0.03187
Total P2Os%	0.1683	0.1453	0.1681
Total K20%	1.7206	1.7289	1.9298
Total Oa%	1.13	1.44	1.72
Total Mg	0.88	0.96	1.52
Total B (ppm)	5.70	7.6	5.7
Soluble N (ppm)	49.39	45.59	44.os
Soluble P (ppm)	2.62	2.22	4.54
Soluble K (ppm)	38.37	32.08	38.36
Soluble B (ppm)	0.08	0.10	0.09
Soluble Ca%	0.08	0.10	0.16
Soluble Mg (ppm)	200.60	285.20	217.90

Table 2
Effect of treatments on number of
pistil late and male catkin shoots.

Treatment (No.)	Pistillate Shoots (No.)	Male catkin shoots[1] (No.)	Comparative Pistillate shoots (%)	male catkin shoots(%)
1	121	138	87.72%	149%
2	119	151	78.74	134
3	129	188	68.49	116
4	138	83	166.67	283
5	121	115	105.26	179
6	137	117	74.07	126
7 (control)	57	97	58.82	100

[1]Counting from two scaffolds per tree by randomized sampling.

Table 3
Correlation between phosphorus content of previous
mix shoots (PMS) and the rate of pistiliate shoots
(PS)/male catkin shoots (MCS) among treatments

Treatment No.	1	2	3	4	5	6	7	Correlation Coefficient
Pcontents of PMS	0.9299	0.9235	0.8793	0.9643	0.9505	0.9132	0.8602	r=0.8269
Rate of PS MCS	0.8772	0.7874	0.6849	1.6667	1.0528	7.7407	0.588	

The value is signficantly different at the 0 OS level

Table 4
Effect of phosphorus treatment on
diameter of pistil late shoots (PS) and number
of burs per pistillate shoot

Diameter of PS/treatment Number	1	2	3	4	5	6	7 (control)
of burs	cm	cm	cm	cm	om	cm	cm
1 bur	0.46	0.46	0.47	0.48	0.43	0.42	0.57
2 burs	0.53	0.53	0.52	0.53	0.53	0.56	0.64
3 burs	0.58	0.56	0.62	0.58	0.60		
4-5 burs	0.71	0.60					
Av. burs/PS	1.67	1.70	1.69	1.88	1.62	1.58	1.50

Table 5

Effect of phosphorus treatments on nut yields. Significance

Blocks		I	II	III	Mean	
Treatments	kg	kg	kg	kg	5X	lx
4	12.00	10.50	8.25	10.25	a[2]	ay
2	8.15	7.50	8.75	8.15	ab	a
	7.50	8.25	7.35	7.70	b	ab
3	12.25	7.00	7.75	7.00	bc	ab
5	8.10	4.75	5.75	4.85	cd	bc
6	4.00	5.25	4.30	4.50	d	bc
7	4.40	3.95	0.90	3.10	d	c

2 Mean separation in columns by Duncan's multiple range test, 5X level. Mean separation in columns by Duncan's multiple range test, 1% level

SCHOLARSHIP

Preliminary Evidence that a Single, Dominant Gene Determines Hairiness on Leaves and Twigs of Chinese Chestnut

FrederickV. Hebard

The leaves and twigs of American chestnut trees are relatively hairless cornpared to Chinese chestnut. This difference may be useful in backcrossing the blight resistance of the Chinese chestnut tree into American chestnut. Blight resistance is the only desired characteristic from the Chinese parent. Selecting blight-resistant progeny for American chestnut characteristics may accelerate the elimination of undesirable characteristics from Chinese chestnut, such as poor forest competitive ness. This would be possible if hairiness is not tightly linked with blight resistance. If it is linked with blight resistance, then it might facilitate selection of blight-resistant progeny.

Leaf and twig hairiness would be most useful in selecting for American chestnut characteristics if it were inherited from Chinese chestnut as a single dominant gene, because the trait would be possessed by about one-half of the progeny from backcrosses to the American chestnut. This study was undertaken to examine the inheritance of leaf and twig hairiness in first hybrids of Chinese and American chestnut and in progeny of backcrosses to both parents.

Material and Methods

Leaves growing in full sunlight were collected from 22 progeny of crosses between Chinese and American chestnut trees (Fl first hybrids), and 41 progeny of backcrosses of Chinese x American first hybrids to American chestnut (B1). In the case of seedlings in their first year of growth, leaves were collected in October from the youngest growth on the plant. In the field, I examined leaves and twigs of 183 progeny of a backcross of a Chinese x American first hybrid to a Chinese chestnut (Chinese B1). During the year in which leaves were collected or examined, all plants had only been growing in the field.

The collected leaves were examined at lOx magnification for the presence of hairs (simple and stellate non-glabrous trichomes) on interveinal portions of the larnina. American chestnut leaves do have simple hairs on their mid ribs and secondary veins, but very few simple or stellate hairs on interveinal portions of the lamina. Leaves were considered hairy if some interveinal portions had more than 10 hairs per square centimeter. When it was difficult to distinguish simple and stellate hairs, the leaves were examined at up to loOx magnification.

Twigs of American chestnut possess simple hairs, but they are sparse and short and not visible to the naked eye. Twigs of Chinese chestnut are hirsute. Twigs were examined for the presence of hairs at the time of leaf collection, without the aid of magnification.

Results and Discussion

Six out of nine Chinese chestnut trees only had first-hybrid progeny with hairy leaves (Table 1). Furthermore, hairy leaves occurred on all 183 progeny of a backcross of one of these first-hybrids to Chinese chestnut. These data indicate that the hairiness trait in those six Chinese parents is dominant. One Chinese chestnut parent (cv Nanking, Table 1) had both hairy and hairless progeny, and two parents (PI 7273 and LF, Table 1) had only hairless progeny. The hairless progeny were either in their first year of growth (cv Nanking and PI 7273) or were less than 30 cm tall (LF), suggesting that leaf hairiness is not always expressed in juvenile leaves when plants are heterozygous for the trait.

All of the older hybrids had both simple and stellate leaf hairs. Some of the hairy 0-year-old progeny had only simple leaf hairs whereas others had both simple and stellate leaf hairs. Juvenile leaves of Chinese chestnut also may possess only simple hairs, although all leaves of Chinese chestnut seedlings growing in full sunlight are hairy. This indicates that juvenile leaves develop simple hairs more readily than stellate hairs.

All Fl progeny with hairy twigs also had hairy leaves, but some 0-year-old progeny with hairy leaves

had hairless twigs. Chinese chestnut seedlings always have hairy twigs, as did all 183 first back-crosses to Chinese chestnut. This suggests that juvenile leaves of heterozygous plants develop hairs more readily than juvenile twigs, but that additional, epistatic factors modify expression of hairiness in juvenile twigs. The expression of simple leaf hairs, stellate leaf hairs, and twig hairs appears to be controlled by a common mechanism. It will be interesting to follow the development of leaf and twig hairs on the 0-year-old progeny in coming years.

Hairs occurred on the leaves of some backcross progeny from all four Chinese chestnut grandparents (Table 2). About one half of the backcross progeny had hairless leaves, while one half had hairy leaves. The ratio of hairy-leaved to hair-less-leaved progeny fit the 1:1 ratio expected if one dominant gene in Chinese chestnut controls the hairiness trait ($p > .5$). The 3:1 ratio expected if two genes control the trait did not fit ($p < .005$). However, there is evidence of heterogeneity in expression of the trait among the progeny of different Chinese chestnut grand parents (Table 2). We also do not know whether any hairless 0-year-old progeny will become hairy in subsequent years. More time and more progeny will confirm whether leaf and twig hairiness in Chinese chestnut is indeed controlled by a single dominant gene, as these results suggest.

SCHOLARSHIP

A Rapid Method of Assessing Whether a Chestnut Tree is Surviving Blight Due to Resistance or Hypovirulence

Frederick V Hebard

The American chestnut tree remains common throughout its range, but most trees are less than 10 cm in diameter and do not flower. In Virginia and West Virginia, flowering American chestnut trees exceeding 40 cm in diameter are found at densities of 1 to 7 per county. Some of these are surviving blight due to genetic resistance, at least in part, while others appear to be surviving solely due to hypovirulence in the blight fungus (Griffin et al., 1983). The latter trees would be good mother trees for backcrossing the blight resistance of Oriental chestnut trees into American chestnut. Partially blight resistant American chestnut trees would not be useful in such a backcross breeding pro-gram, because their resistance could impede selection for the Oriental sources of resistance. It may be possible however to use them in a program of recurrent selection for blight resistance.

Unfortunately, it takes several months or years to determine whether a large American chestnut tree has been surviving blight due to hypovirulence or resistance. The most rapid method is to inoculate each tree with virulent strains of the blight fungus and to measure canker growth over periods of 4 to 12 months; but such *in situ* resistance tests are not conclusive proof. This paper presents a method for quickly assessing whether a chestnut tree is surviving blight primarily due to resistance or to hypovirulence in the blight fungus.

Materials and Methods

Chestnut blight cankers can be classified as lethal or non-lethal. Lethal cankers are slightly sunken, with abundant sporulation, and reach the vascular cambium (are not superficial). When they encircle a stem, distal portions of the stem die.

Non-lethal cankers are characterized by: superficial canker development (cankers do not reach the vascular cambium), sometimes extending for several meters up the bole; swelling of host tissues under and around superficial cankers (swelling is less pronounced in highly blight-resistant trees); little sporulation; and survival of distal portions of encircled stems. In some nonlethal cankers, wood exposed by a canker which would have been lethal becomes surrounded by 2 to 8 years of non-necrotic callus formation (callus ridges); this may be an indication of hypovirulence (Griffin, et al., 1982). Intermediate forms exist between these two broad categories, and are discussed by Roane, et al. (1986).

Any chestnut tree surviving blight by definition has non-lethal cankers located on its bole. Additionally, if it is surviving blight due to resistance, the remainder of the cankers also should be non-lethal. In contrast, one might expect lethal cankers to occur in the crowns of chestnut trees surviving blight due to hypovirulence. This is because hypovirulence viruses may not have had time to infect virulent strains of the fungus causing new cankers in the crown. Virulent inoculum is always present: hypovirulent strains of the fungus emit spores lacking the hypovirulence virus (Russin and Shain, 1985).

The crowns of chestnut trees surviving blight due to resistance and hypovirulence were examined for the occurrence of lethal and nonlethal cankers (Table 1).

Results and Discussion

Blight-susceptible American chestnut trees located in Michigan (10 trees), Connecticut (10 trees) and Virginia (2 trees) that were surviving blight due to hypovirulence had nonlethal cankers on their boles, but several lethal cankers occurred in their crowns (Table 1). In contrast, ten blight-resistant Chinese chestnut trees and four of their moderately resistant hybrids with American chestnut had only nonlethal cankers in both their crowns and boles. Likewise four large, surviving American chestnut trees which tested as slightly or moderately resistant to blight (Griffin, et al., 1983) had only nonlethal cankers in their crowns and boles.

Therefore, the combination of nonlethal bole cankers with nonlethal crown cankers is a preliminary indication of survival due to resistance, whereas the combination of nonlethal bole cankers with lethal

crown cankers is a preliminary indication of survival due to hypovirulence.

At the VPI & SU airport, 10 third-generation irradiated chestnut trees with nonlethal bole cankers had lethal cankers in their crowns. Similarly, irradiated trees at Stronghold, Maryland, had lethal cankers in their crowns and nonlethal cankers on their boles. Hence those irradiated trees may be surviving blight primarily due to hypovirulence.

In areas clearcut 10 to 15 years previously, it is common to find a few American chestnut sprouts with nonlethal *Endothia* cankers. Most of the remaining sprouts in old clearcuts are dead due to blight. Some sprouts with nonlethal cankers have been observed to survive blight for many years. Lethal cankers were observed in the crowns of seven sprouts with nonlethal bole cankers located in three clearcuts. Multiple isolations were made from nonlethal cankers on the seven trees and tested for pathogenicity. Preliminary data suggest reduced pathogenicity in some isolates. Proportionally fewer isolates with reduced pathogenicity were obtained from nearby control trees with lethal cankers. No blight resistance has been detected during pathogenicity tests on more than 700 chestnut sprouts in clearcuts (Griffin, et al., 1983, and Hebard, unpublished data). Thus the nonlethal cankers on sprouts in old clearcuts may be due solely to hypovirulence in *Endothta parasitica*

This method suffers some limitations. The crowns of small trees may not contain enough cankers to have lethal crown cankers. Furthermore, cankers of the nonlethal type may kill a stem on a blight-resistant chestnut tree, especially if it is stressed by drought, shading, low fertility or winter injury. Frequently however, such cankers will not be sunken, with abundant sporulation. The occurrence of sunken, abundantly sporulating cankers in the crown of chestnut trees with non-lethal bole cankers appears to be a good preliminary indication of survival due to hypovirulence.

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