Seed Orchard Design

Outline

- **Mating Design**
  - Maximize representation of all lines
- **Seed Orchard Design**
  - Test parents (as known)
  - Maximize crossing of unrelated lines
- **Genotype x Environmental Interactions**
- **Ideal vs. Reality**
  - No single answer

Purpose of a Seed Orchard

- **Progeny Testing** 3
  - Determine parental quality
- **Genetic gain**
  - Trait improvement
- **Seed Production** 2
- **Capture diversity** 1

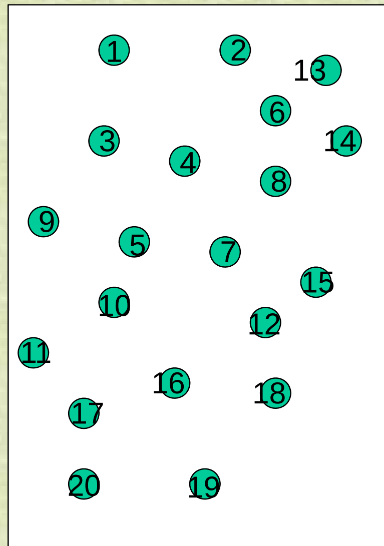
- **Purpose will drive mating design**

Mating Designs

- **Incomplete Pedigree**
  - Open-pollinated mating
  - Polycross (pollen mix)
- **Complete Pedigree Deigns**
  - Nested, Factorial, Single-Pair, Full Diallel, Half Diallel, Partial Diallel

Incomplete Pedigree

- **Pollen parent is unknown**
  - Open-pollinated mating
  - Polycross (pollen mix)
    - Almost controlled, but apply a mix of pollens to each flower/tree.
    - Useful if not all trees in one location.



Complete Pedigree

- **Single Pair Mating**
  - Provide good information for full-sib family performance
  - Provide estimates of some genetic parameters
  - Maximum unrelatedness but not optimum for selection
  - Low cost

1 x 2
3 x 4
5 x 6
7 x 8
… x …

Complete Pedigree

- **Nested Mating**
  - Provide information for parents and full-sib families
  - Provide estimates of both additive and dominance effects
  - Not efficient for selection
  - Low cost for controlled mating

♂

♀	1	2	3
A	X		
B	X		
C	X		
D		X	
E		X	
F		X	
G			X
H			X
I			X

Complete Pedigree

- **Factorial Mating**
  - Provide good information for parents and full-sib families
  - Provide estimates of both additive and dominance effects
  - Limited selection intensity
  - High cost

♂

♀	1	2	3	4	5
A	X	X	X	X	X
B	X	X	X	X	X
C	X	X	X	X	X
D	X	X	X	X	X
E	X	X	X	X	X

Complete Pedigree

- **Full Diallel**
- **Half Diallel**
  - Provide good evaluation of parents and full-sib families
  - Provide estimates of both additive and dominance effects
  - High cost

♀ \ ♂	1	2	3	4	5
1	X	X	X	X	X
2	X	X	X	X	X
3	X	X	X	X	X
4	X	X	X	X	X
5	X	X	X	X	X

♀ \ ♂	1	2	3	4	5
1	X	X	X	X	X
2		X	X	X	X
3			X	X	X
4				X	X
5					X

Open Pollination vs. Controlled Pollination

- **To capture 95% of allelic diversity:**

$(2^n - 2)^a / (2^n)^a$

which reduces to $(1 - 2^{1-n})^a$

9 progeny from each straight BC3

vs

10 progeny from each cross of two straight BC3s (?)

Open Pollination – Capturing Allelic Diversity

Number of selected Progeny	Percentage Captured (%)	How much to add? (%)
1	50.00	50.00
2	62.50	37.50
3	71.88	28.13
4	78.91	21.09
5	84.18	15.82
6	88.13	11.87
7	91.10	8.90
8	93.33	6.67
9	94.99	5.01

Open Pollination vs. Controlled Pollination

- **To capture 95% of allelic diversity:**

  $(2^n - 2)^a / (2^n)^a$

  which reduces to $(1 - 2^{1-n})^a$

9 progeny from each straight BC3

vs

10 progeny from each cross of two straight BC3s (?)

Controlled Pollination – Capturing Allelic Diversity

			Number of selected Progeny	Percentage Captured (%)	How much to add? (%)
1	x	2	1	50.00	50.00
1	x	2	2	75.00	25.00
1	x	2	3	87.50	12.50
1	x	2	4	93.75	6.25
1	x	2	5	96.88	3.13

RrRrRr x RrRrRr

- 64 possible combinations
- 1 with all Dominant alleles

	RRR	RRr	rRR	RrR	Rrr	rrR	rRr	rrr
RRR	RRRRRR	RRRRRr	RrRRRR	RRRrRR	RRRrRr	RrRrRR	RrRRRr	RrRrRr
RRr	RRRRrR	RRRRrr	RrRRrR	RRRrrR	RRRrrr	RrRrrR	RrRRrr	RrRrrr
rRR	rRRRRR	rRRRRr	rrRRRR	rRRrRR	rRRrRr	rrRrRR	rrRRRr	rrRrRr
RrR	RRrRRR	RRrRRr	RrrRRR	RRrrRR	RRrrRr	RrrrRR	RrrRRr	RrrrRr
Rrr	RRrRrR	RRrRrr	RrrRrR	RRrrrR	RRrrrr	RrrrrR	RrrRrr	Rrrrrr
rrR	rRrRRR	rRrRRr	rrrRRR	rRrrRR	rRrrRr	rrrrRR	rrrRRr	rrrrRr
rRr	rRRRrR	rRRRrr	rrRRrR	rRRrrR	rRRrrr	rrRrrR	rrRRrr	rrRrrr
rrr	rRrRrR	rRrRrr	rrrRrR	rRrrrR	rRrrrr	rrrrrR	rrrRrr	rrrrrr

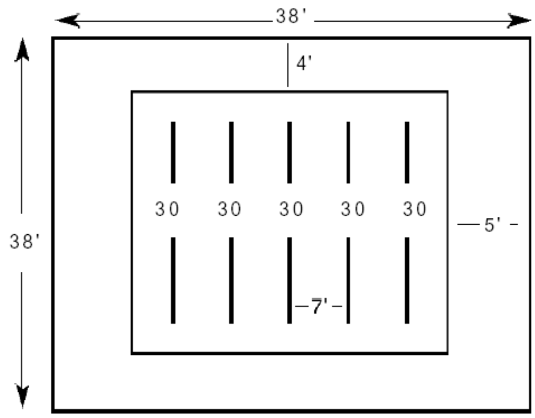
How Many Trees to Plant

- **$P = 0.99 = 1 - \sum_{m=0}^{3} 74 \ (0.125^{m})(08.75)^{74-m}$**
- **OPEN POLLINATION**
  - 64 trees * 2 chances = 128 trees
  - 85% survival = plant 150 seed
  - 150 trees * 20 lines = 3000 seed
    - 3000 trees / block
  - 3000 x 9 replicates = 27,000

How Many Trees to Plant

- **CONTROLLED POLLINATION**
  - 64 trees * 2 chances = 128 trees
  - 85% survival = plant 150 seed
  - 150 trees * 10 lines = 1500 seed
    - 1500 trees / block
  - 1500 x 10 replicates = 15000

Plots



The Plot: the smallest unit

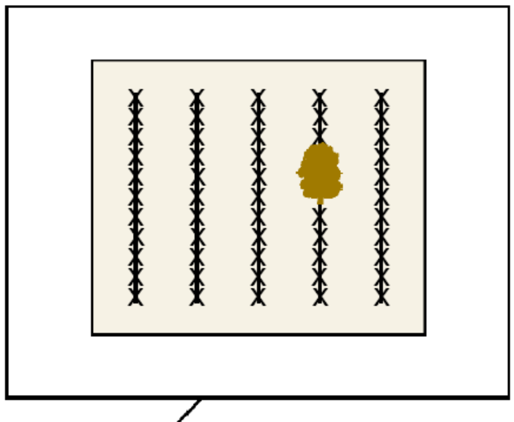
One hundred and fifty B_3-F_2 nuts, all progeny of the same B_3 tree, are planted in one plot in five rows of thirty nuts each. The rows are seven feet apart and the nuts are one foot apart within each row. A four to five foot border is maintained around the seedlings.

- **150 seed from same genetic line per plot**
- **20 plots (lines) within a block**

Blocks

- **There are 20 plots within a block.**
  - Each plot represents a different genetic line, defined by it's BC3 heritage.
- **There are nine replications of these plots.**

Selection within Plots



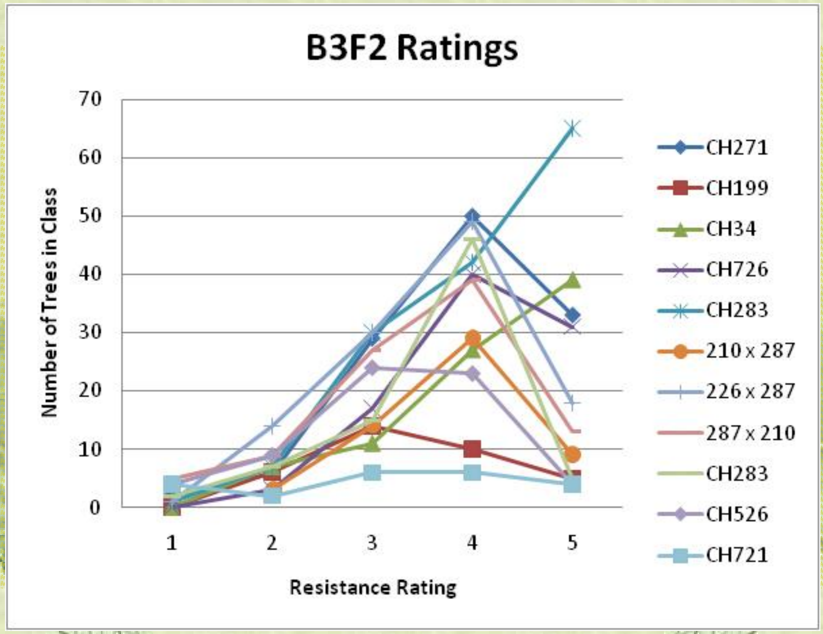
Selection occurs in each plot

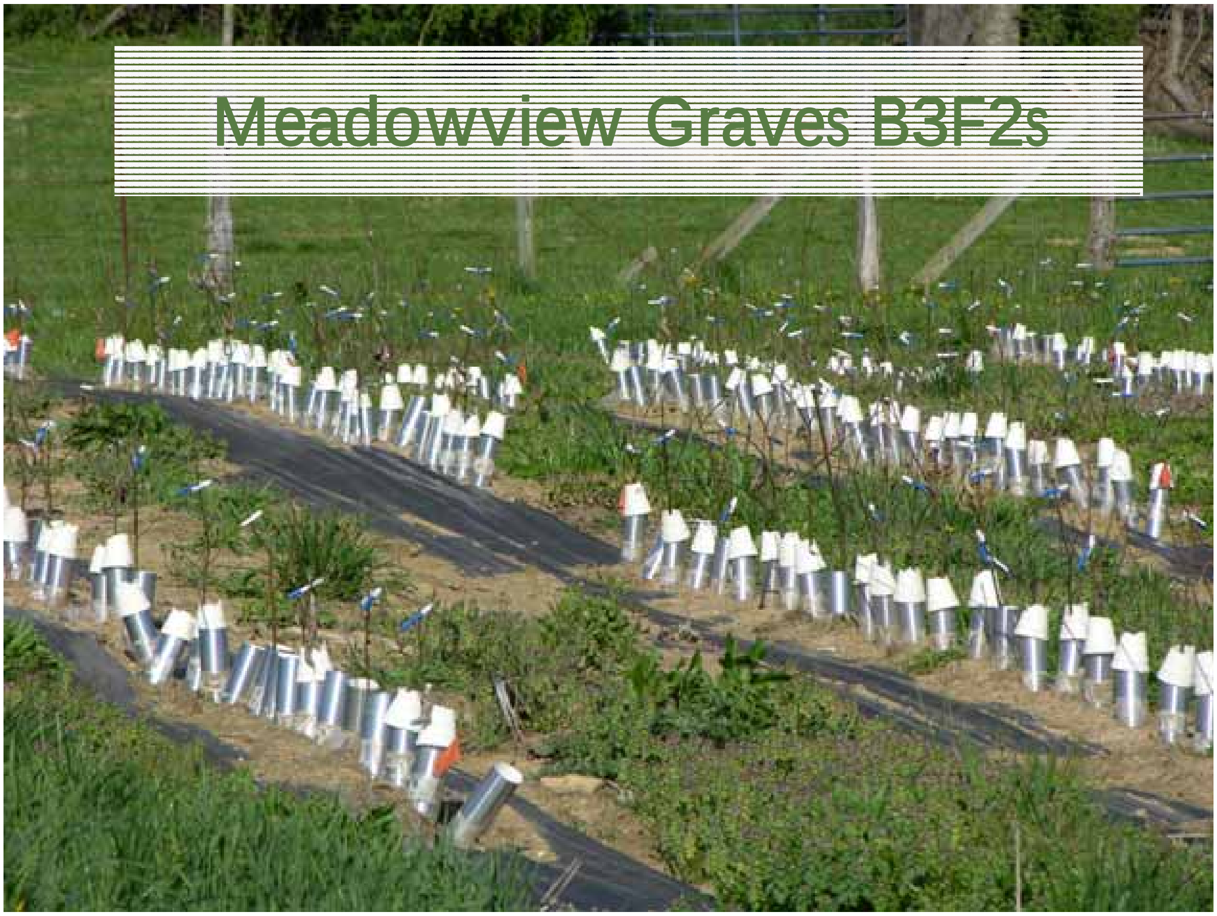
At two years of age the seedlings are inoculated with the blight fungus. The trees are rogued over period of years, with the most blight-susceptible rogued first. Only one seedling, the most blight resistant, is ultimately chosen to remain as part of the seed orchard.

- **Inoculation occurs when trees within a plot average 1 cm in stem diameter at 1 foot above the ground.**
  - This may be within the 2nd or 3rd growing season
  - Maybe even 3rd or 4th
  - Now going toward a staggered inoculation design

Inoculations

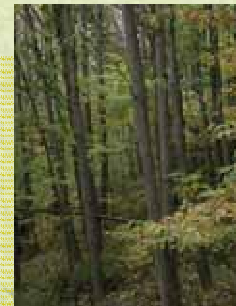
Skewed Distribution



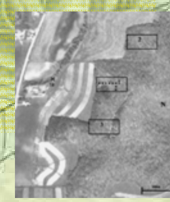

Meadowview Graves B3F2s


Meadowview Clapper B3F2

What if We Don't Get Good Resistance?



- **Recurrent Selection?**
- **What's the effect?**
- **Inbreeding concerns?**
  – Limited Reintroduction Does Not Always Lead to Rapid Loss of Genetic Diversity: An Example from the American Chestnut (Castanea dentata; Fagaceae)

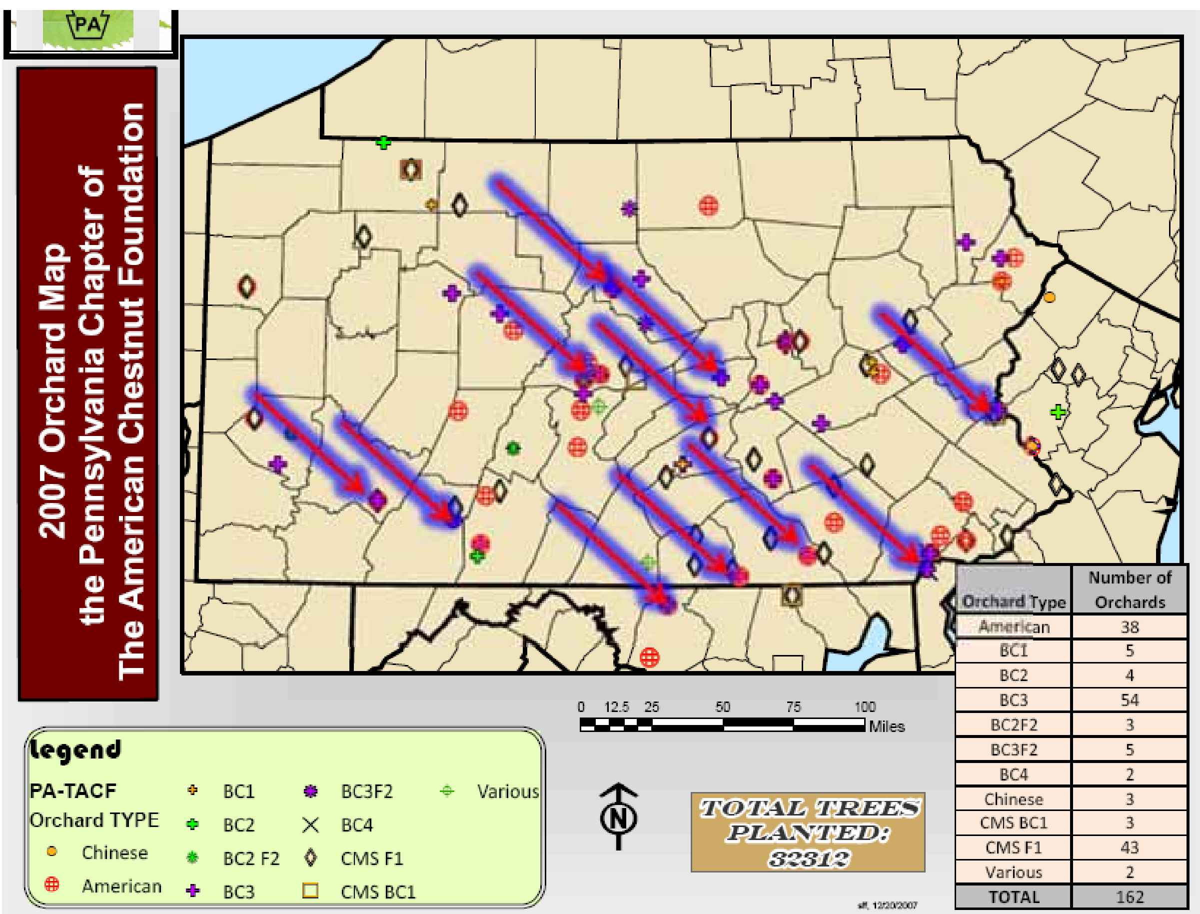


Open Pollination

- **Very straightforward**
- **Start with 20 lines**
- **Each has an (assumed) equal opportunity to be pollinated by each of another parent.**
- **Replicate 9 times and have a possible 9 combinations of parents**

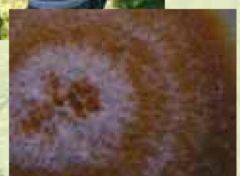
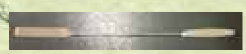
Controlled Pollination?

- **Lines**
  - Staggered coming out
  - Some can open-pollinate, some can't
- **Cross each line to each other?**
- **Or just cross one line to one or a couple of other lines?**
- **Combination of Open and Controlled?**
- **Pollen Mix?**



Orchard Type	Number of Orchards
American	38
BC1	5
BC2	4
BC3	54
BC2F2	3
BC3F2	5
BC4	2
Chinese	3
CMS BC1	3
CMS F1	43
Various	2
TOTAL	162




64

Brogue: Ort x CL287

Br96-66, rated 2.5, 18ft tall,4.5"dbh (2006)

Reels Corner: JoSc x GR210

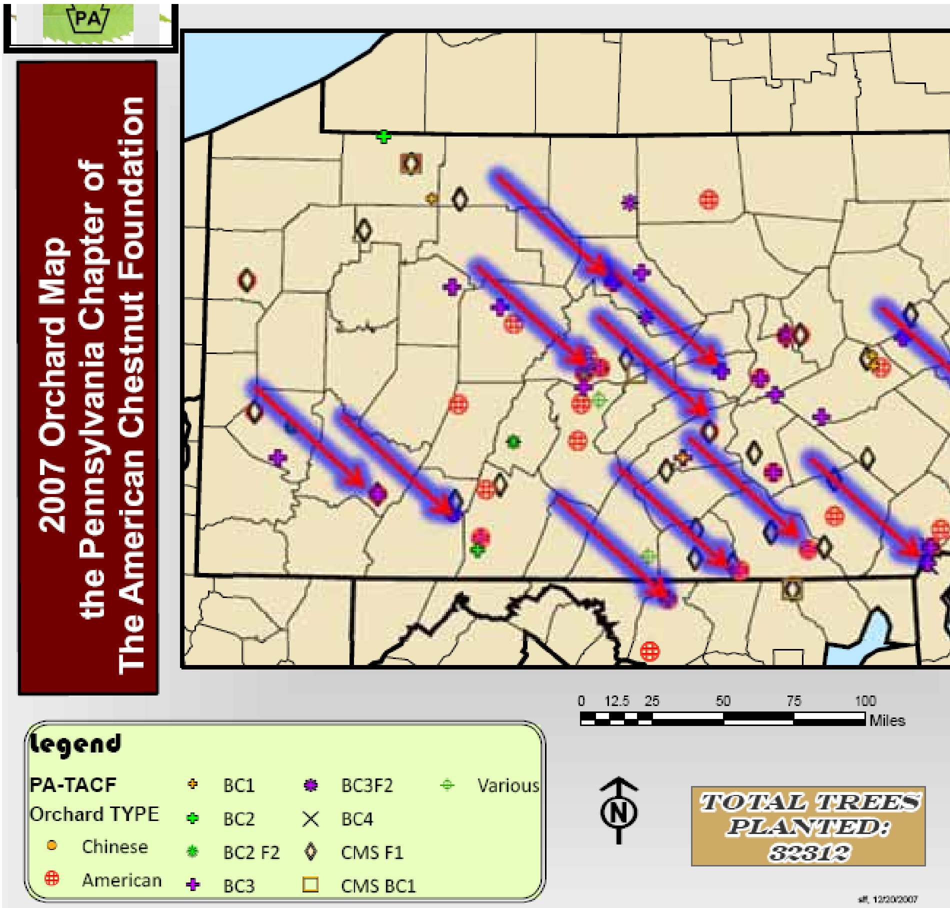
Rc96-96, rated 3, 10 ft tall (2001)

3.15 dbh
Planted 2000
Rated 2
(prelim. 2007)

Red Clay: LhWe-m x GR97

Environmental Effects?

- **Ober**
  - Same lines replicated elsewhere
  - Worst performing
  - Huge initial lesions
  - Large final canker size



Orchard Type	Number of Orchards
American	38
BC1	5
BC2	4
BC3	54
BC2F2	3
BC3F2	5
BC4	2
Chinese	3
CMS BC1	3
CMS F1	43
Various	2
TOTAL	162

Space and Time

- **Open-pollination vs. controlled**
- **Combination?!**

Farm	Year Inoculated	Number of Lines
Beech Creek	2007	3
Brogue	2000	2
Brogue	2001	4
Brogue	2006	3
Codorus	2006	2
Dornsife	2000	1
Dornsife	2001	2
Hummelstown	2004	2*
Kuhns	2006	7**
Kuhns	2008	7**
Ober	2004	4
Ober	2008	3
Red Clay	2004	3
Red Clay	2007	9
Reels Corner	2002	1
Riegelsville	2005	2
Thorpewood	2006	3

Please.

- **Only use one, maybe two trees as a representative for any given line.**
- **Use THE BEST TREES!**
- **That DOES NOT mean to destroy them.**
  - Save just in case.

LINE	Female	Male	Planted
CL287 x GR210	br96-012	br97-123	150
	br96-026	br97-199	6
	br96-066	br97-105	28
	br96-066	br97-184	122
	br96-106	br97-109	53
	br96-106	br97-199	92
	br96-115	br97-199	184
	br97-001	br96A-016	53
	br97-090	br96A-016	29
	br97-105	br96-012	28
	br97-109	br96-115	4
	br97-109	br96-087	23
	br97-109	br96-026	100
	br97-111	br96A-016	15
	br97-123	br96-106	1
	br97-184	br96-106	15
	br97-199	br96-106	20
	br97-199	br96-115	31
	br97-199	br96-066	190
	do96-030	do97-130	9
	do97-053	br96-106	37
	do97-053	br96-012	3
	do97-130	do96-030	20
	do97-171	do97-130	3
	rc97-031	br96-087	80
	rc97-075	br96-066	74
	rc97-076	br96-106	27
	rc97-107	br96-066	4
TOTAL			1401

Controlled Pollination?

- **Lines**
  - Staggered coming out
  - Some can open-pollinate, some can't
- **Cross each line to each other?**
- **Or just cross one line to one or a couple of other lines?**
- **Combination of Open and Controlled?**
- **Pollen Mix?**

Crossing Schemes

- **Purpose?**
  - Capture allelic diversity
    - Capture all of which is contained within a certain line
      - Which is represented by a single BC3 tree
- **So all you _really_ need, probably, is just one cross replicated 9 times**
  - But . . .

Open Pollination

- **Capture each by having 9 representatives**
  - Potential of having 9 different crosses
  - But maybe only one combination of mother and pollen parent occurs
    - Not likely
    - But won't know

Controlled Pollination

- **Parents are known.**
- **How many crosses?**
  - Minimum of 1 cross replicated 9 (or 10?) times.
  - Ultimate possibility would be Half-diallel
- **COMPROMISE!!**

Depends on Timing, Location, and Resources Available

- **Look at Handouts**

Farm	Year Inoculated	Number of Lines
Beech Creek	2007	3
Brogue	2000	2
Brogue	2001	4
Brogue	2006	3
Codorus	2006	2
Dornsife	2000	1
Dornsife	2001	2
Hummelstown	2004	2*
Kuhns	2006	7**
Kuhns	2008	7**
Ober	2004	4
Ober	2008	3
Red Clay	2004	3
Red Clay	2007	9
Reels Corner	2002	1
Riegelsville	2005	2
Thorpewood	2006	3

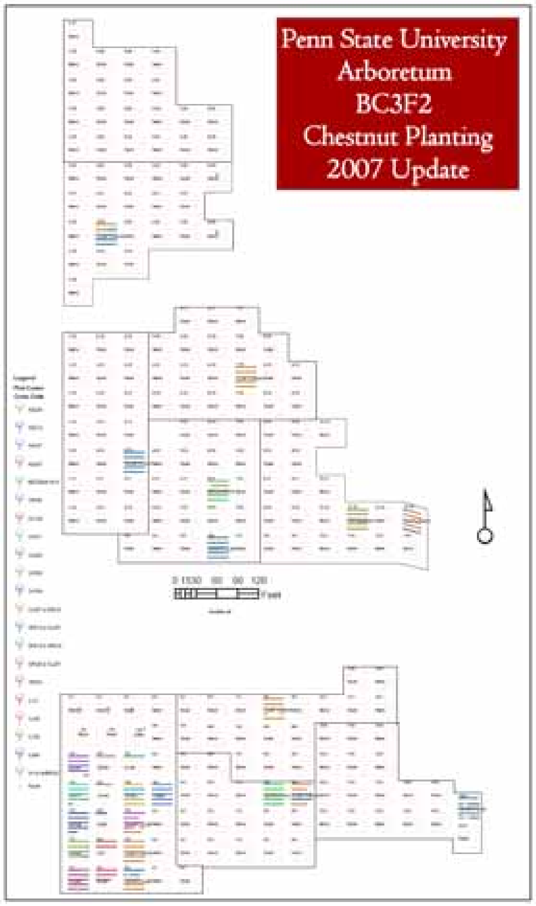
* At Hummelstown, one Graves and one Clapper line were inoculated

** The Kuhns orchard was planted on 5' centers. Trees need to be rogued via inoculation and selection to make room for rest of trees to grow.

Farm_Name	Farm_code	Seed_Type	Fem_Code	Mal_Code	MaxOfObs_Date	No_Inoculated	Seed
Beech Creek	PA-bc	BC3	GiYo	AB185	5/18/2007	9	HY804
Ober	PA-ob	BC3	PeNo	AB185	6/6/2004	11	HY805
Red Clay	PA-re	BC3	LuPe	AB185	6/15/2007	2	HY806
Brogue	PA-br	BC3	GsSc-1	AB393	6/7/2006	15	HY943
PSU - Kuhns	PA-ku	BC3	GsSc-1	AB393	6/23/2006	16	HY943
Brogue	PA-br	BC3	FaYo	AB427	6/7/2006	6	HY980
Brogue	PA-br	BC3	TaYo	AB427	6/7/2006	1	HY942
Codorus State Park	PA-cs	BC3	KeCu	AB427	6/7/2006	7	HY920
Codorus State Park	PA-cs	BC3	TRTCm	AB427	6/7/2006	47	HY920*
Merle Thorpe	PA-mt	BC3	KeCu	AB427	6/7/2006	17	HY920
PSU - Kuhns	PA-ku	BC3	KeCu	AB427	6/23/2006	25	HY920
PSU - Kuhns	PA-ku	BC3	TRTCm	AB427	6/23/2006	14	HY920*
Red Clay	PA-re	BC3	KeCu	AB427	6/15/2007	11	HY920
Merle Thorpe	PA-mt	BC3	AB329	Ar403	6/7/2006	25	HY803*
Beech Creek	PA-bc	BC3	DhCe	BE325	5/18/2007	1	HY928*
Merle Thorpe	PA-mt	BC3	DrCn-2	BE325	6/7/2006	30	HY930*
PSU - Kuhns	PA-ku	BC3	DhCe	BE325	6/23/2006	5	HY928*
PSU - Kuhns	PA-ku	BC3	DrCn-2	BE325	6/23/2006	24	HY930*
PSU - Kuhns	PA-ku	BC3	DrCn-m	BE325	6/23/2006	14	HY911*
PSU - Kuhns	PA-ku	BC3	CaYo	BE325	6/23/2006	17	HY942
Riegelsville	PA-ri	BC3	DrCn-2	BE325	6/4/2005	48	HY930*
Beech Creek	PA-bc	BC3	SuWe-m	BE400	5/18/2007	4	HY807*
Beech Creek	PA-bc	BC3	GL42-m	BE400	5/18/2007	20	HY808
Ober	PA-ob	BC3	SuWe-m	BE400	6/6/2004	17	HY807*
Red Clay	PA-re	BC3	GL42-m	BE400	6/15/2007	33	HY808
Brogue	PA-br	BC3	TA7	CL234	5/29/2000	10	HY802

Number of Trees Inocul

Seed_Type	Male	Female	Farm_Name	Total
BC3	AB185	GiYo	Beech Creek	9
		LuPe	Red Clay	2
		PeNo	Ober	11
	AB393	GsSc-1	Brogue	15
			PSU - Kuhns	16
	AB427	FaYo	Brogue	6
		KeCu	Codorus State Park	7
			Merle Thorpe	37
			PSU - Kuhns	25
			Red Clay	11
		TaYo	Brogue	1
		TRTCm	Codorus State Park	47
			PSU - Kuhns	14
	BE325	CaYo	PSU - Kuhns	17
		DhCe	Beech Creek	1
			PSU - Kuhns	5
		DrCn-2	Merle Thorpe	30
			PSU - Kuhns	24
			Riegelsville	48
		DrCn-m	PSU - Kuhns	14
	BE400	GL42-m	Beech Creek	20
			Red Clay	33
		SuWe-m	Beech Creek	4
			Ober	37
	CL234	TA7	Brogue	10
	CL287	CrYo	Brogue	17
		OrYo	Brogue	89
			Dornsife	54
	CL53	DeNo1	Dornsife	4
		DeNo2	Brogue	28
			Dornsife	30
		RcSo-13	Red Clay	35
		[illegible]	PSU - Kuhns	14



Orchard Layout

- **PSU Arboretum**
  - 10 blocks
    - 9 for PA material
    - 1 for Meadowview

10 acre Arboretum

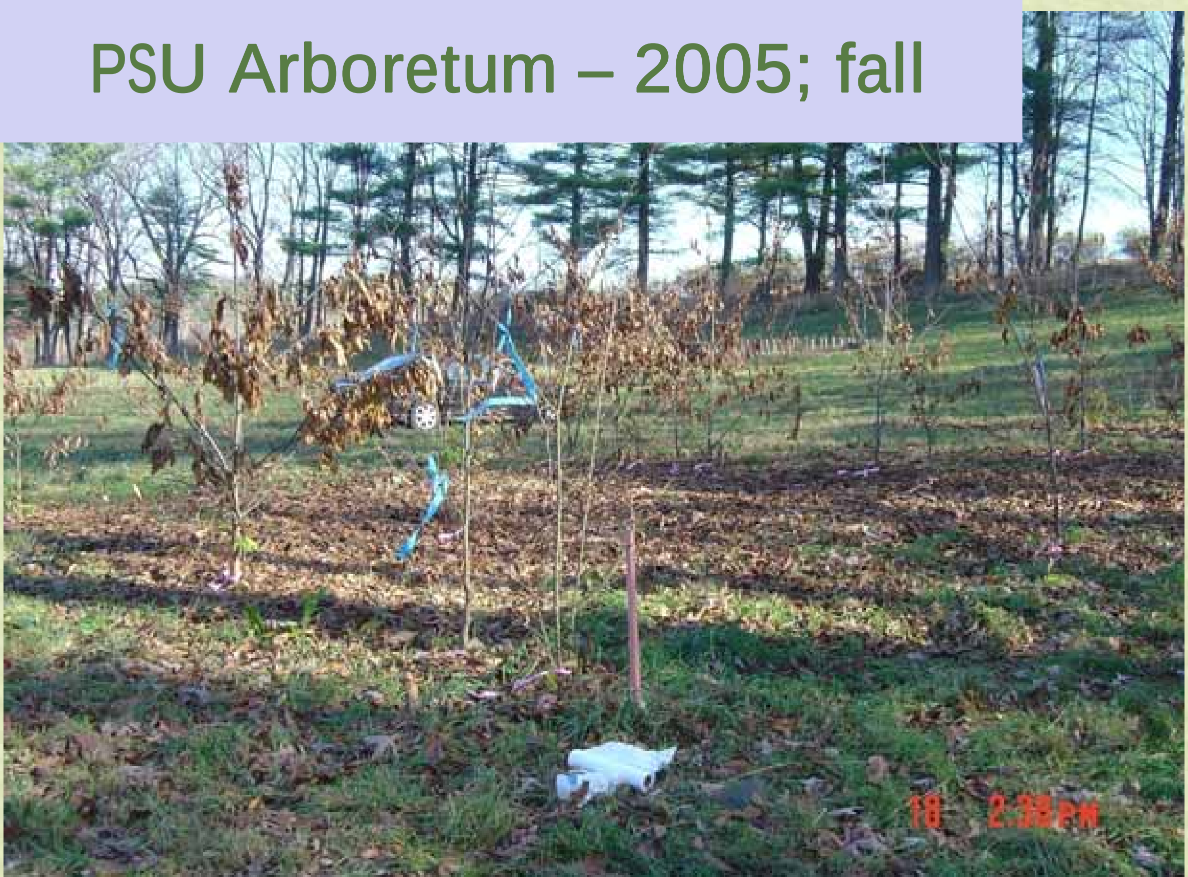
http://www.arboretum.psu.edu/research/orchard.html

PSU Arboretum - 2002

PSU Arboretum – 2004; winter

PSU Arboretum – 2005; summer

PSU Arboretum – 2005; fall



PSU Arboretum – 2006; summer

Delegate Responsibility

- **Split the blocks up as necessary**
- **Recommend**
  - At least one larger orchard
    - 4 – 5 blocks
    - Single cooperator
      - One with an established infrastructure
- **Makes overall maintenance easier and cheaper**
- **Better guarantee long-term success**
- **Potential for designed research on trees.**

Single blocks

- **A volunteer grower can maintain a single block,**
- **But the time commitment, especially, may be too much for a volunteer**
  - Time Commitment: 25-30 years

Carbaugh B3F2s – 1st planting

Carbaugh block

- Planted 2005
- Ready to inoculate 2008

Carbaugh B3F2s – 2nd planting

Requirements

- **Each block**
  = approximately 1 acre

- **Maintenance per year:**
  - Planting
    - A group of 5 volunteers can plant over 300 seedlings in three hours which includes laying ground cloth, digging, planting, and watering. This is decreased somewhat when planting by seed (which is preferred).
  - Weed Control
    - Herbicide vs. Landscape Fabric and hand-weeding
    - Mowing
  - Irrigation
  - Fertilization

Yearly Labor Inputs

- **Varies**
  - Depends on type of maintenance
- **Main need: One person to guarantee oversight**

Our seed orchards in Meadowview, for 10 acres:

- 2-3 days for a crew of 5-7 people for a few thousand seed.
- It takes about 2-3 mornings to spray them with herbicide, which we do twice a year for a total of 5-6 mornings.
- It takes 1-2 days to mow them, and we do that about three times a year, about 6 days.
- Finally, it takes 2 afternoons to fertilize them by broadcasting granular fertilizer, which also could be done with a truck operated by the fertilizer-store folks.

Other Inputs

- Deer Protection (fence)
- Equipment
  - Tractor / Mower / WeedEater

- **Seed Protection**
  - May have in storage (plastic tubes) or get donated (aluminum flashing)
- **Fertilizer**
  - We can often get great deals
- **Herbicide/Landscape Fabric**
  - Latter is more expensive, but can sometimes have it donated.

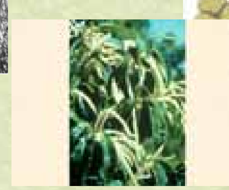
Funding

- **Funding for our current Clapper BC3F2 Orchard comes from:**
  - PA Chapter
  - TACF
  - Robertson Endowment (5% interest on $50,000)
    - Pays for maintenance; wage payroll at University
  - Hardwood Forestry Fund - fencing
  - National Tree Trust - fencing
  - Private donations for PSU Arboretum Establishment

Partnerships

- **The Key!**
  - Utilize established infrastructure, if possible.
  - Also helps get grant funding
    - USFS seed orchard locations?
    - Colleges/Universities
    - Partner with other non-profits
      - Land trusts

Questions?



More Information

- **Further Information Available in:**
  - Volume 16, Issue 1 of TACF Journal which can be downloaded at (*see also Volume 8, Issue 1*)
    - http://ecosystems.psu.edu/research/chestnut/information/journal
- **Contact Information**

Sara Fern Fitzsimmons
Penn State University
206 Forest Resources Lab
University Park, PA 16802

Phone: 814-863-7192 (W)
Phone: 814-404-6013 (C)

E-Mail: sff3@psu.edu

Pennsylvania Chapter of the American Chestnut Foundation (PA-TACF)

Attn: Dave Armstrong
691 Pumping Station Road
Hanover, PA 17331-8608

Phone: 717-632-8669

E-mail: darm@blazenet.net