Conifer Translational Genomics Network
Coordinated Agricultural Project

Genomics in Tree Breeding and Forest Ecosystem Management

Module 12 – Marker Informed Breeding (MIB) – Program Management

Nicholas Wheeler– Oregon State University

Marker applications

- Quality control
- Introgression and hybrid breeding
- Parentage analysis
  - *Pollen contamination*
- Enhanced breeding designs

Quality control in a clonal seed orchard

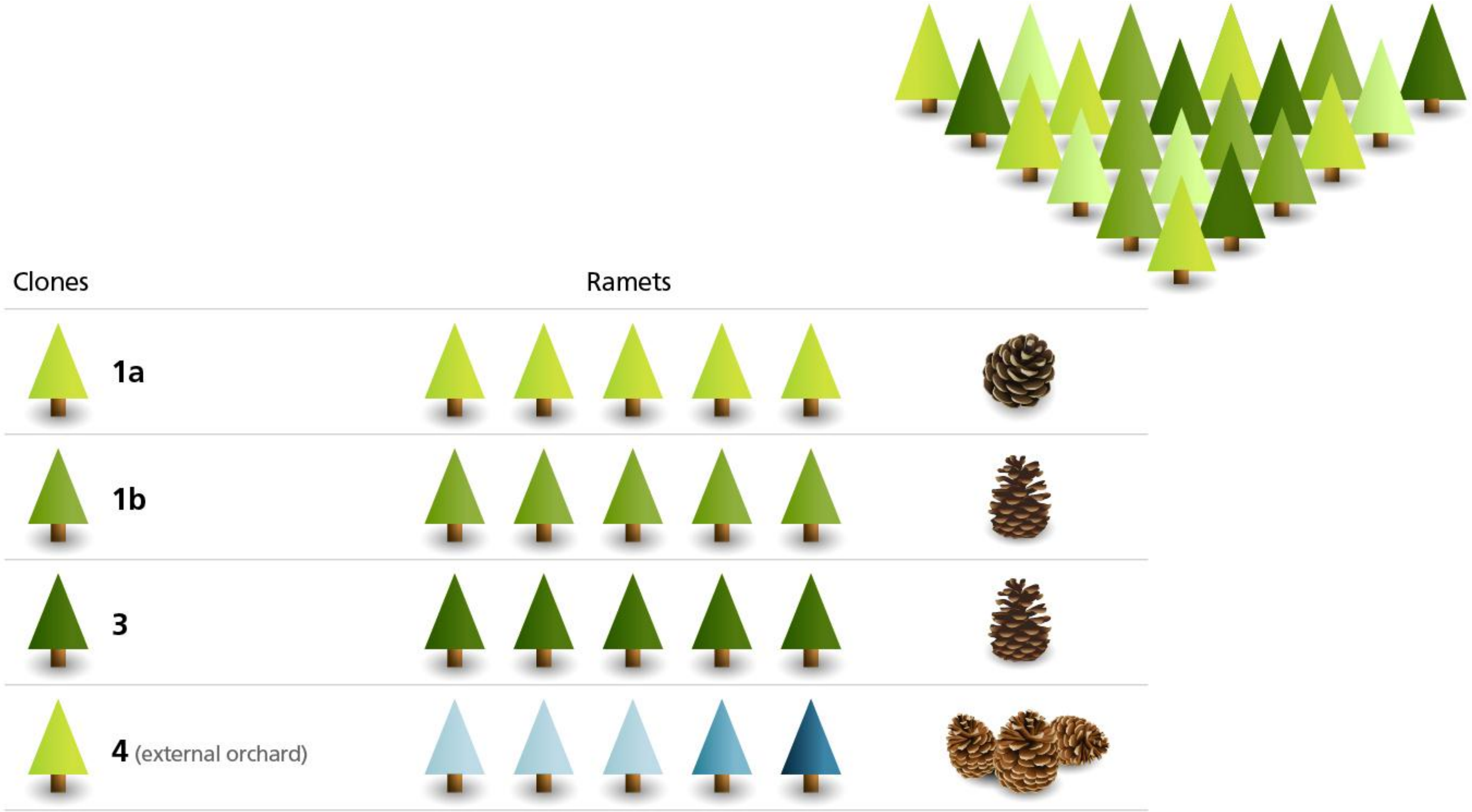

Figure Credit: Nicholas Wheeler, Oregon State University

Opportunity cost of orchard errors

Assume

- Progeny of wrong orchard clone have 5% less genetic gain (volume) than progeny of desired clone
- 1% gain = $10 present value/acre planted ($60 future value at rotation)
- Annual planting requirement = 10,000 acres, of which 10% is planted with the desired clone (1000 acres)
- 40% of trees in desired clone are mislabeled; thus, 400 acres per year are planted with trees that are performing 5% below expected
- 400 acres * 5% * $10/acre/percent = $20,000 present value ($120K future value). If unchecked for 15 years, = $300K present value

Marker assisted backcrossing or introgression

- This is easily the single greatest application of markers to animal and plant breeding in the world
- Used predominantly for simply inherited traits such as disease or insect resistance
- Commonly used to introduce an important allele from an unimproved individual / species into a highly selected individual / line / variety

Backcross breeding approaches

- Marker-assisted “foreground” selection
  - *Select for markers that identify favorable alleles from the donor population*
  - *Markers must be in very tight linkage with desired trait*

- Marker-assisted “background” selection
  - *Select for marker alleles that identify the host (recurrent) parent genome*
  - *Must have many alleles with complete genome coverage*

- Typically, both approaches are used together

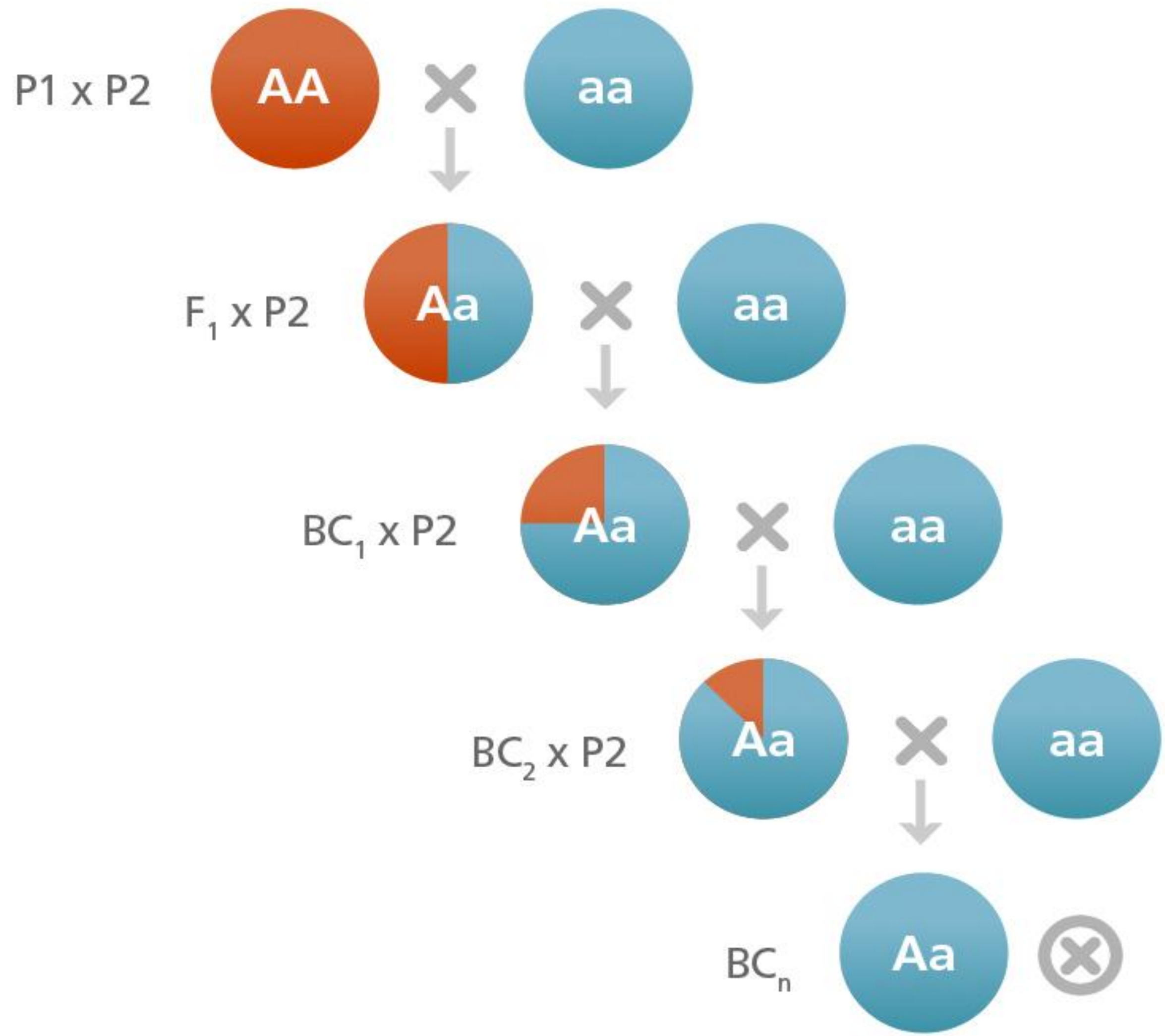

Figure Credit: Modified from Welz and Geiger, 2000

Distribution of genotypes in simulated BC_1 population: recovery of RP genome

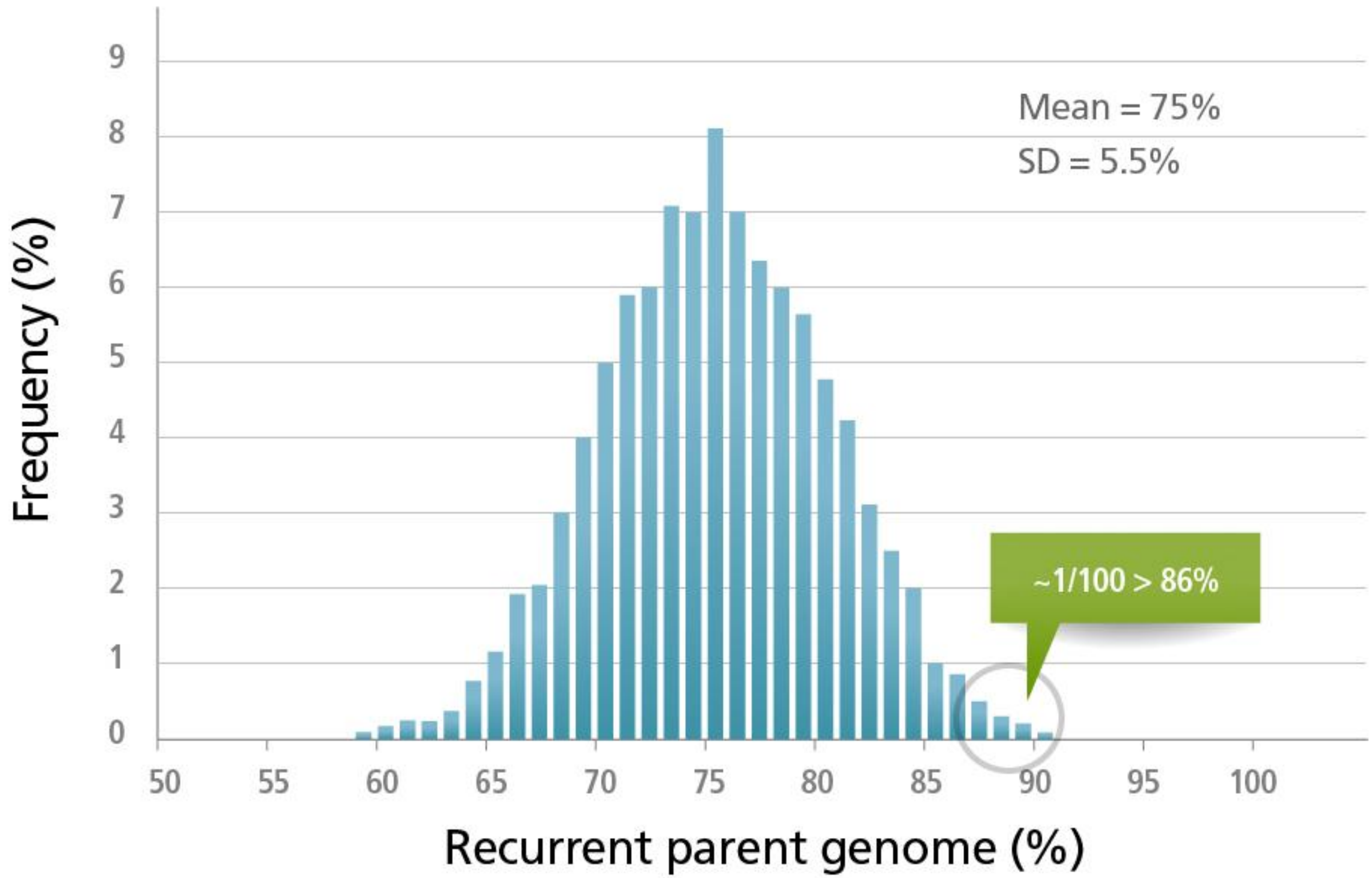

Figure Credit: Modified from Welz and Geiger, 2000

American chestnut

- Early in the 20th century, the Appalachian Mountains were full of giant chestnut trees
- Chestnuts were abundant, accounting for 25% of all Appalachian hardwoods
- Tree diameters of 8 to 10 feet were often reported. One in Francis Cove, North Carolina, was 17 feet in diameter
- Chestnut grew tall (up to 120 feet) and straight -- often clear of branches up to 50 feet, making them ideal lumber

Image Credit: John Carlson, The Pennsylvania State University and The American Chustnut Foundation.

American chestnut

Image Credits: John Carlson, The Pennsylvania State University and The American Chestnut Foundation

American chestnut

- Chestnut was an important commodity for the early European settlers of the Appalachians

- Uses included
  - *Nuts for food, mast, and cash*
  - *Tanning hides*
  - *Building materials*

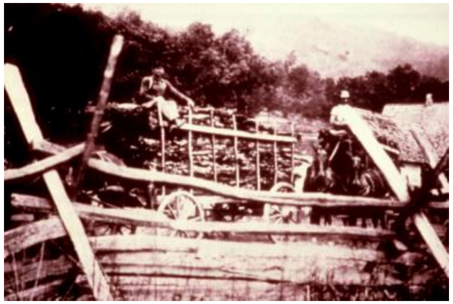

Image Credits: John Carlson, The Pennsylvania State University and The American Chestnut Foundation

The chestnut blight

- By the 1950s, chestnut was virtually eliminated as a dominant forest tree





Image Credits: John Carlson, The Pennsylvania State University and The American Chestnut Foundation

QTL studies reveal three sources of major resistance

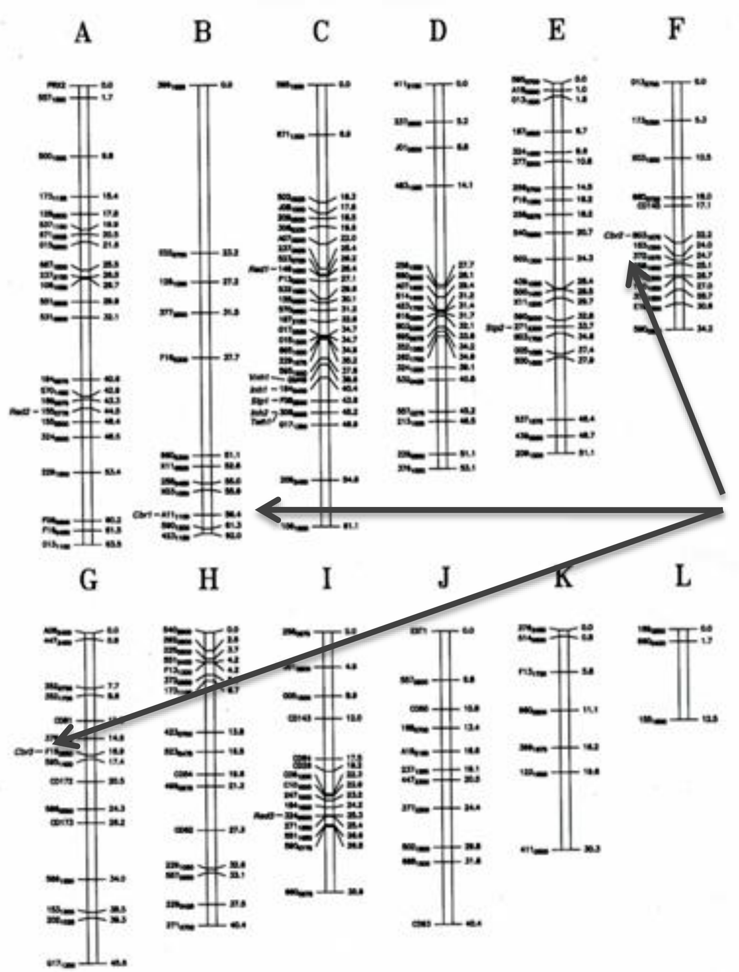

Figure Credit: Modified from Kubisiak et al., 1997

Marker applications in chestnut restoration

- Quality control in breed orchards
- Genetic mapping
- QTL mapping
- Marker informed introgression
- Locating resistance genes

Image Credits: Brad Smith and Fred Heberd, The American Chestnut Foundation

Propagation population characterization

- Varietal protection (quality control)
- Parentage analysis / orchard efficiency
  - *Pollen contamination*
  - *Parental contribution to the gene pool*
  - *Pollen competition*
  - *Supplemental mass pollination success*
  - *Mating systems*

Pollen contamination

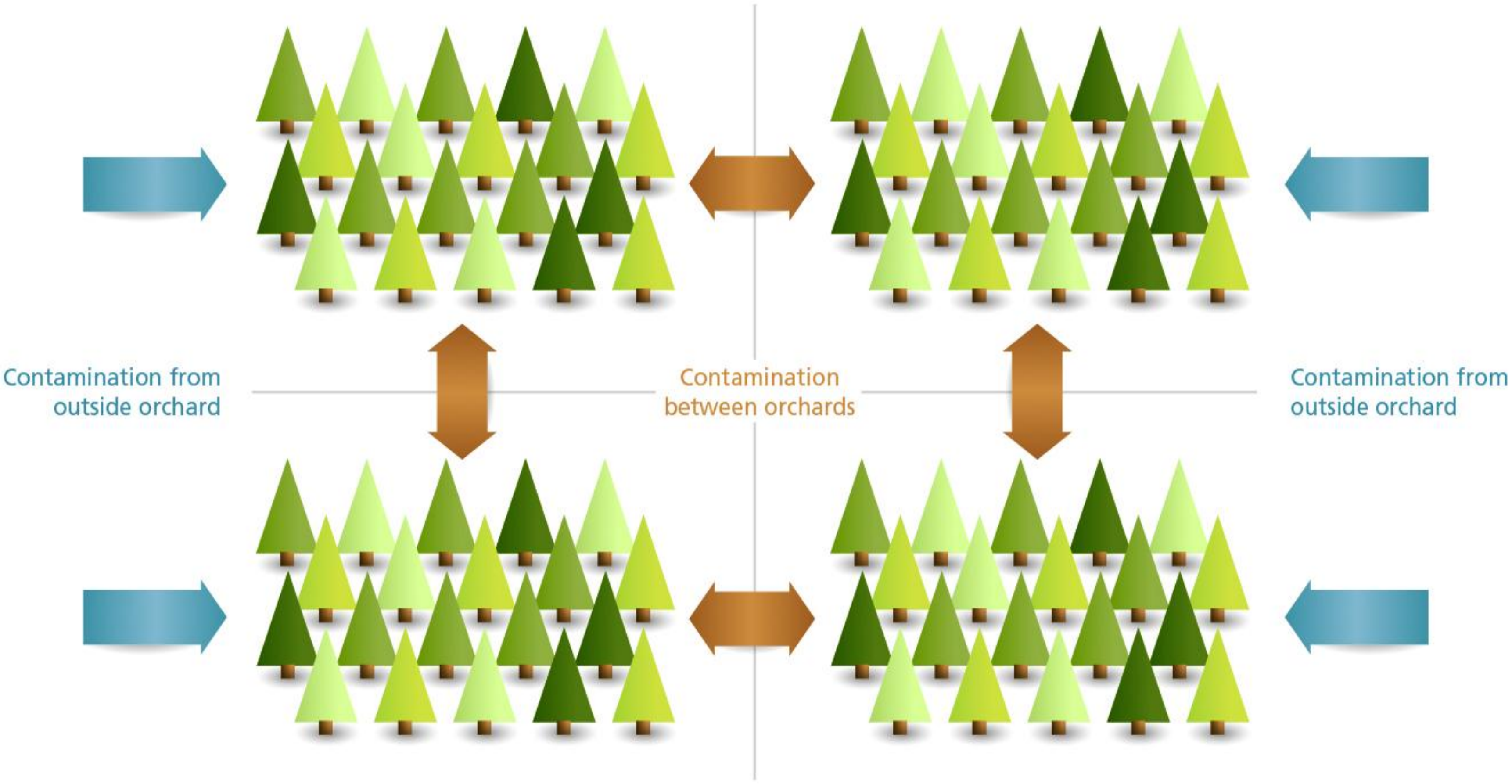

Figure Credit: Nicholas Wheeler, Oregon State University

Pollen contamination in DF orchards

Year	Type of seed collection	# of seeds analyzed	Observed seed contamination (%)	Pollen contamination (%) ± SE
1999	Bulk	192 (190[b])	1.0	31.0 ± 3.5
2000	Bulk	192 (102[b])	46.9	36.8 ± 5.2
2000	Individual-ramet	240	0	32.0 ± 3.2
2003	Individual-ramet	336	0	41.3 ± 2.8
Mean				**35.3 ± 2.4**

Slavov et al., 2005; Figure Credit: Glenn Howe, Oregon State University

PC is higher in parents that flower early

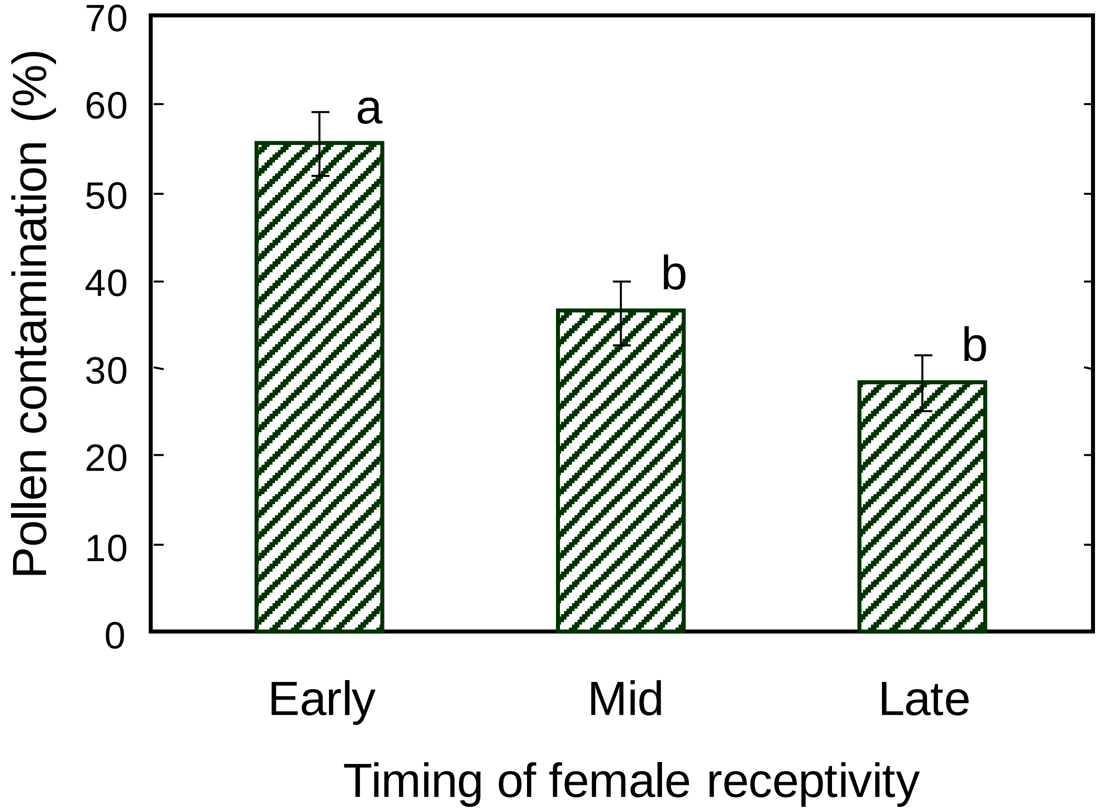

Figure Credit: Glenn Howe, Oregon State University

Differential paternal success

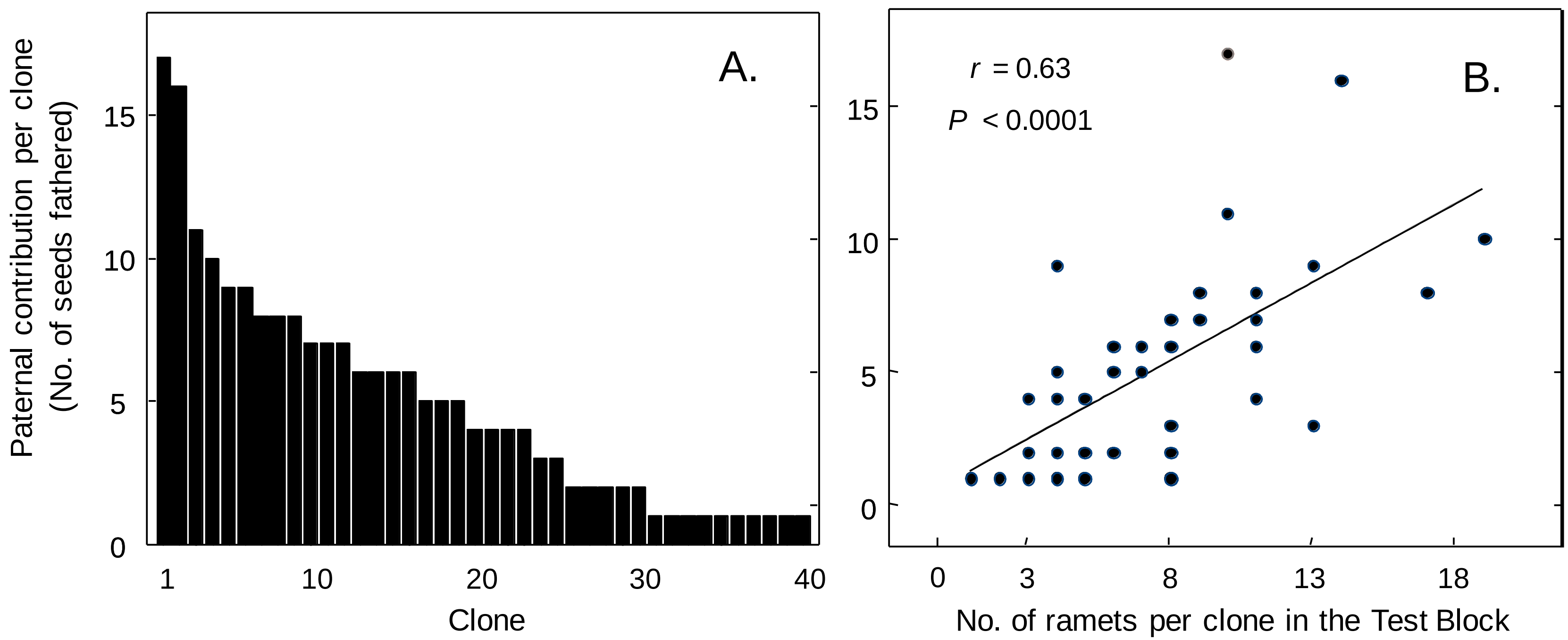

Figure Credit: Glenn Howe, Oregon State University

Parentage reconstruction

Table 5. Parent tree identification using SSR genetic markers. The SSR genotypes of two putative open-pollinated (OP) field selections were compared to the genotypes of known progeny growing in genetic test plantations (n = 8-10). If the putative parents in the field are correct, then all of their progeny must have at least one of the two parental alleles. Cases in which a progeny allele matches one of the alleles in the putative parent are shown in white. Allele numbers (e.g., 209 or 216) represent relative lengths of alternative SSR alleles.

Putative OP parent (i.e., tree in the field)			Progeny number (in progeny test plantation)											
ID	SSR genotype (marker name)	Progeny allele	1	2	3	4	5	6	7	8	9	10	Inferred genotype of real OP parent	Conclusion: putative parent is:
1	209, 216	Shorter =	205	209	202	216	209	216	216	209	209	216	209, 216	Correct
	(OSU_3F1)	Longer =	216	209	209	216	219	227	223	216	211	218		
1	184, 191	Shorter =	172	188	184	184	176	172	191	191	186	184	184, 191	Correct
	(OSU_3B9)	Longer =	191	191	184	184	184	191	191	191	191	191		
1	254, 256	Shorter =	254	254	228	256	226	226	254	254	226	254	254, 256	Correct
	(OSU_4A7)	Longer =	254	284	256	284	256	254	254	254	254	254		
2	204, 222	Shorter =	210	210	192	210	210	214	190	214			210, 214	Incorrect
	(OSU_3F1)	Longer =	224	214	210	214	226	214	210	220				
2	130, 190	Shorter =	130	188	130	130	188	130	130	130			130, 188	Incorrect
	(OSU_3B9)	Longer =	208	194	210	196	200	130	188	190				
2	226, 226	Shorter =	226	226	226	226	226	268	228	282			226, 284	Incorrect
	(OSU_4A7)	Longer =	226	228	244	248	284	284	284	284				

Figure Credit: Glenn Howe, Oregon State University

Number of loci needed for paternity in DF

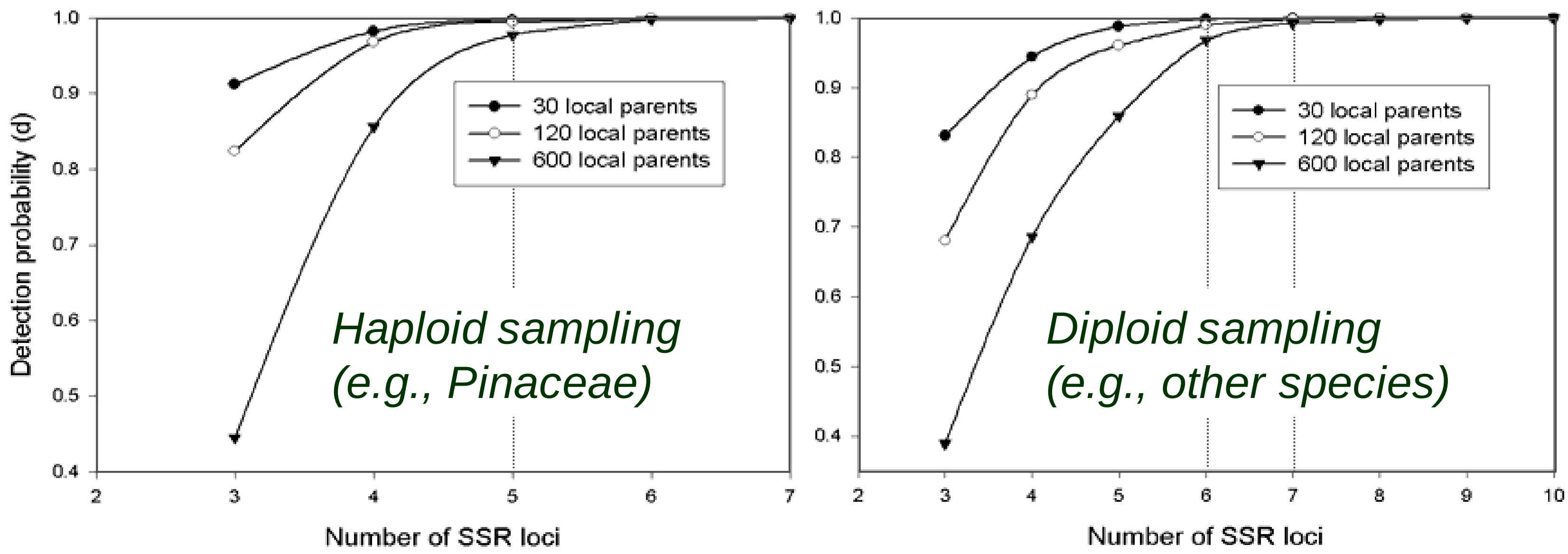

Figure Credit: Glenn Howe, Oregon State University

Enhanced breeding designs: Informed choices for full-sib matings

- Complementary breeding
  - *Identify individuals that differ (genotypes) from one another at a key locus (i.e. R genes for disease resistance)*
  - *Mate to distribute R genes into other backgrounds*
- Pyramiding genes
  - *An extension of complementary breeding where you attempt to accumulate desirable alleles at two or more loci into one progeny cohort (see example later)*
- Diversity index breeding

Enhanced breeding designs: Using markers for paternity analysis

Polymix breeding with paternity analysis (PMX/WPA)*

- Replace multiple breeding designs (PMX for BV estimation + Full-sib design for advanced generation selection) with just a single PMX test
- Fingerprint the top individuals in the PMX test to determine paternity (maintains pedigree control and manages inbreeding)
- Saves time and cost of full-sib breeding while actually increasing gain potential

* Lambeth, et al., 2001

Tree improvement flow diagram

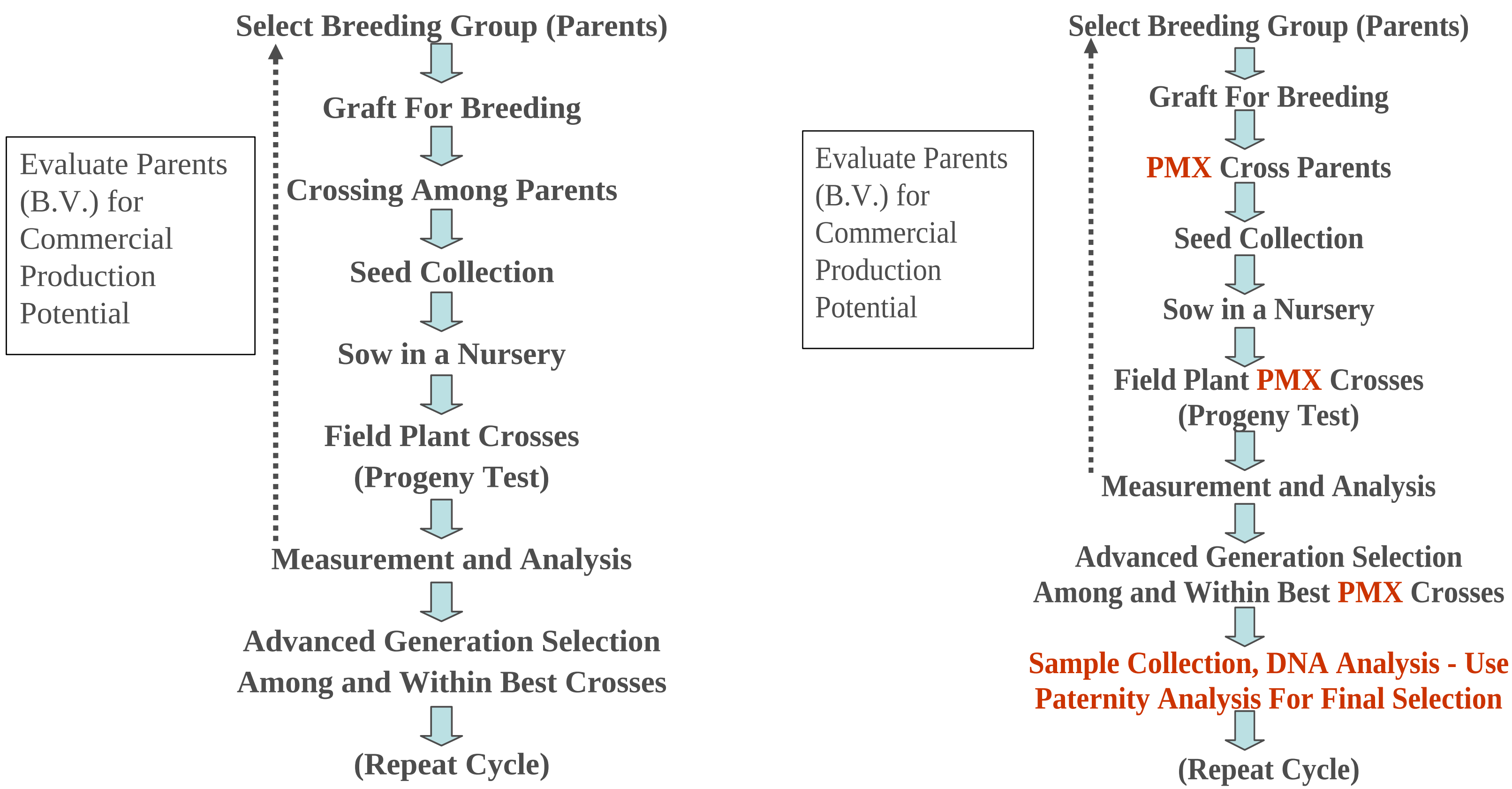

Figure Credit: Clem Lambeth

FEMALE PARENT \ MALE PARENT	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
1		X	X	X	X	X																								
2			X	X	X	X																								
3				X	X	X																								
4					X	X																								
5						X																								
6																														
7								X	X	X	X	X																		
8									X	X	X	X																		
9										X	X	X																		
10											X	X																		
11												X																		
12																														
13														X	X	X	X	X												
14															X	X	X	X												
15																X	X	X												
16																	X	X												
17																		X												
18																														
19																				X	X	X	X	X						
20																					X	X	X	X						
21																						X	X	X						
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23																								X						
24																														
25																										X	X	X	X	X
26																											X	X	X	X
27																												X	X	X
28																													X	X
29																														X
30																														

DISC. HALF-DIALLELS
75 crosses
5 crosses per parent
7 of top 28 crosses made

PARENTAL B.V. ESTIMATION	_	**O.K.**
SELECTION FOR FUTURE	_	**GOOD TO WEAK**
PEDIGREE (INBR.) CONTROL	_	**GOOD**
BREEDING & TESTING	_	**MOD. DIFF.**

Figure Credit: Clem Lambeth

		M	A	L	E		P	A	R	E	N	T																				
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	PMX
F	1	3	4	9	2	4	5	6	3	3	7	2	4	4	4	5	4	0	8	1	6	6	4	3	4	3	3	5	5	1	2	X
E	2	2	7	1	5	3	3	5	7	5	3	3	3	3	5	3	6	4	3	5	5	5	2	4	5	4	5	4	4	5	1	X
M	3	0	2	7	3	5	3	3	6	5	2	3	2	1	6	4	3	2	5	7	2	6	6	8	5	2	3	8	6	2	3	X
A	4	0	4	7	2	3	8	2	3	9	5	6	3	6	7	3	3	4	4	4	1	3	3	3	8	1	6	5	3	2	2	X
L	5	1	3	2	8	4	10	5	5	3	1	2	6	3	2	3	3	4	7	5	3	5	6	5	2	4	4	3	5	3	3	X
E	6	1	4	4	3	3	3	4	4	5	3	2	1	4	5	2	8	2	7	3	5	3	10	5	7	5	6	5	4	2	0	X
	7	2	4	4	3	4	2	6	3	8	2	1	4	1	7	3	2	3	4	2	6	3	4	7	7	6	4	5	6	4	3	X
P	8	2	4	4	2	4	5	1	7	2	6	2	6	4	9	5	7	4	7	2	5	3	6	2	2	3	2	3	4	5	2	X
A	9	4	6	3	5	4	7	3	3	3	3	8	3	2	3	3	4	5	6	3	5	3	2	3	3	4	5	5	3	8	1	X
R	10	2	7	5	1	9	3	4	5	4	3	2	2	4	2	3	5	7	4	4	3	2	4	8	4	5	2	3	3	8	2	X
E	11	2	4	5	1	3	3	4	8	4	5	6	4	3	5	4	4	4	3	4	6	1	3	4	5	5	4	7	3	4	2	X
N	12	2	2	3	5	3	4	9	2	1	4	2	6	6	6	6	5	5	6	3	3	2	7	5	4	2	6	4	2	4	1	X
T	13	4	1	6	7	2	3	5	6	1	4	1	2	5	1	6	4	6	2	9	4	2	3	5	8	1	3	9	5	3	2	X
	14	2	4	7	4	0	7	3	2	4	4	3	2	5	4	5	2	4	2	5	4	5	3	5	2	5	2	4	7	10	4	X
	15	0	5	3	3	6	6	3	3	6	7	5	3	4	2	2	2	1	3	4	3	5	12	8	3	3	5	7	2	3	1	X
	16	0	5	5	5	7	2	5	4	5	4	4	3	4	4	4	1	7	7	3	2	3	6	2	4	3	4	6	4	3	4	X
	17	2	6	3	5	5	2	5	4	6	0	2	3	9	8	3	7	3	1	3	4	5	1	2	7	4	5	4	3	7	1	X
	18	1	5	4	3	3	2	5	6	5	6	4	1	7	2	5	5	6	3	4	2	5	1	4	4	6	1	5	8	5	2	X
	19	2	6	7	4	5	1	4	1	8	2	2	0	4	1	3	2	2	6	10	2	6	6	4	2	8	5	4	1	10	2	X
	20	2	8	5	4	2	6	4	2	3	1	2	3	2	5	2	9	5	5	5	4	11	4	1	6	5	4	2	4	3	1	X
	21	1	8	3	3	4	1	4	6	1	7	4	5	6	2	6	2	6	2	6	6	4	7	4	4	6	3	4	4	0	1	X
	22	4	1	6	4	1	5	5	0	5	2	5	8	5	5	2	1	2	1	5	10	5	5	3	2	6	6	3	5	4	4	X
	23	1	3	1	2	2	3	6	3	6	5	4	4	5	2	7	5	4	3	7	2	7	1	3	7	9	5	4	5	3	1	X
	24	1	5	2	4	5	3	5	3	4	7	3	5	1	7	5	4	4	5	5	3	5	5	3	3	3	7	5	4	3	1	X
	25	5	4	2	3	4	3	7	2	5	5	3	4	7	4	2	1	5	4	1	4	4	5	4	3	5	8	5	5	2	4	X
	26	3	2	2	7	10	6	1	1	3	3	7	4	2	4	2	4	2	4	2	5	5	6	5	7	4	4	2	6	5	2	X
	27	3	2	2	3	10	0	7	3	3	6	9	4	3	3	7	4	2	1	7	2	4	3	3	5	4	2	3	4	9	2	X
	28	2	5	6	3	5	4	4	1	6	2	2	4	2	3	4	4	7	5	3	3	7	3	1	5	8	5	6	5	3	2	X
	29	1	2	4	8	2	4	1	7	5	4	4	4	6	5	1	3	8	5	6	4	7	2	3	3	4	6	5	4	1	1	X
	30	2	3	5	4	2	1	2	2	4	2	2	2	7	8	6	4	5	4	3	2	8	4	3	3	4	5	2	7	9	5	X

All 435 crosses
30 crosses per parent
All 28 best crosses

PARENTAL B.V. ESTIMATION	_	**EXCELLENT**
SELECTION FOR FUTURE	_	**EXCELLENT**
PEDIGREE (INBR.) CONTROL	_	**EXCELLENT**
BREEDING & TESTING	_	**SIMPLE**
PATERNAL ANALYSIS	_	**DIFFICULT**

Figure Credit: Clem Lambeth

Retrospective selection strategy of elite parents based on paternity testing of progeny individuals displaying superior performance using microsatellite markers.

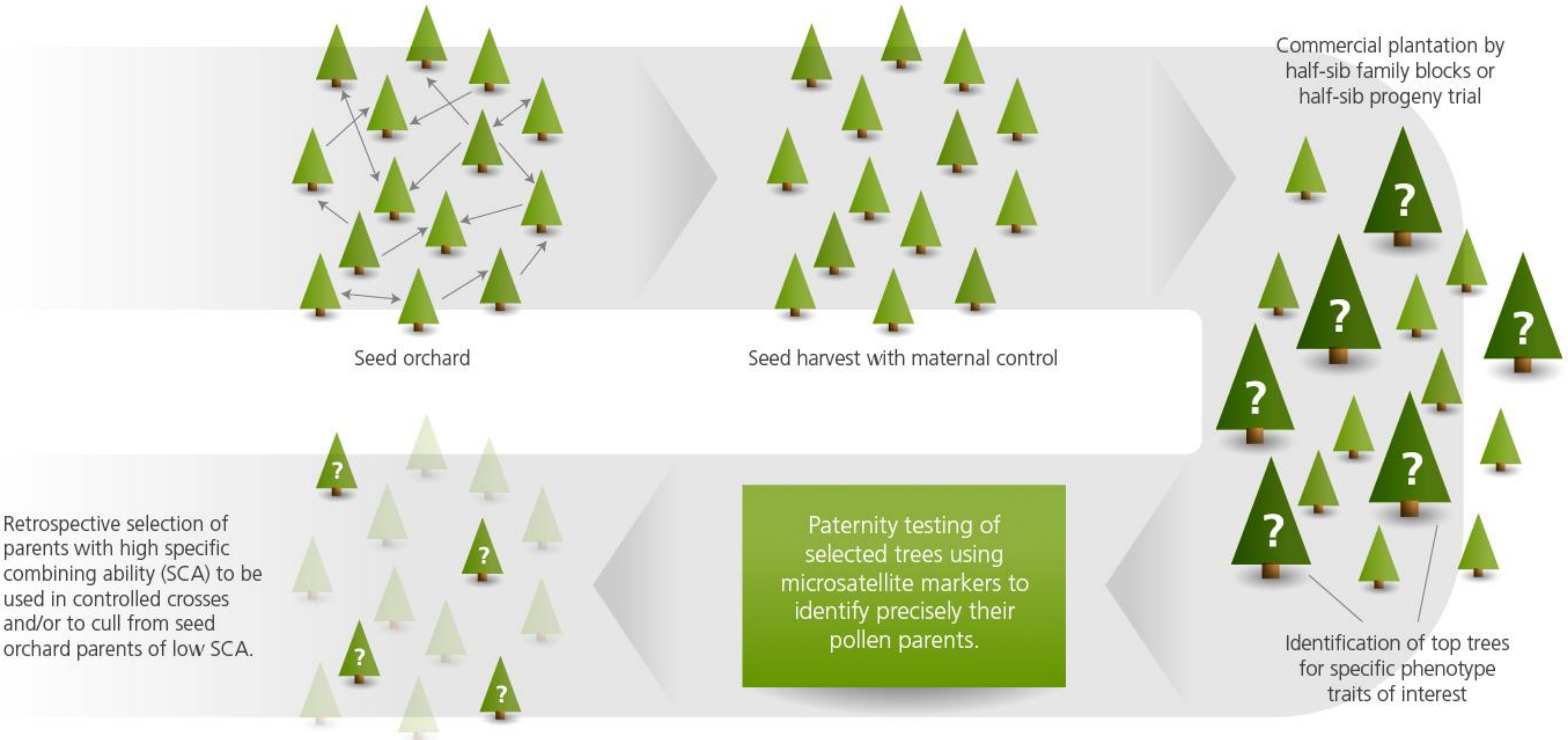


Figure Credit: Modified from Grattapaglia, 2007

Implementation strategies for marker informed breeding in tree improvement

- “All forms of MAS can be applied separately or in conjunction with classical methods of selection (mass, family, within-family, combined and index selection) and can be utilized to make selections for selected, breeding and/or production populations”
  - *In White, Adams and Neale, Forest Genetics, Chapter 19, Page 554*

Approaches to MAS: (Classified by mapping precision)

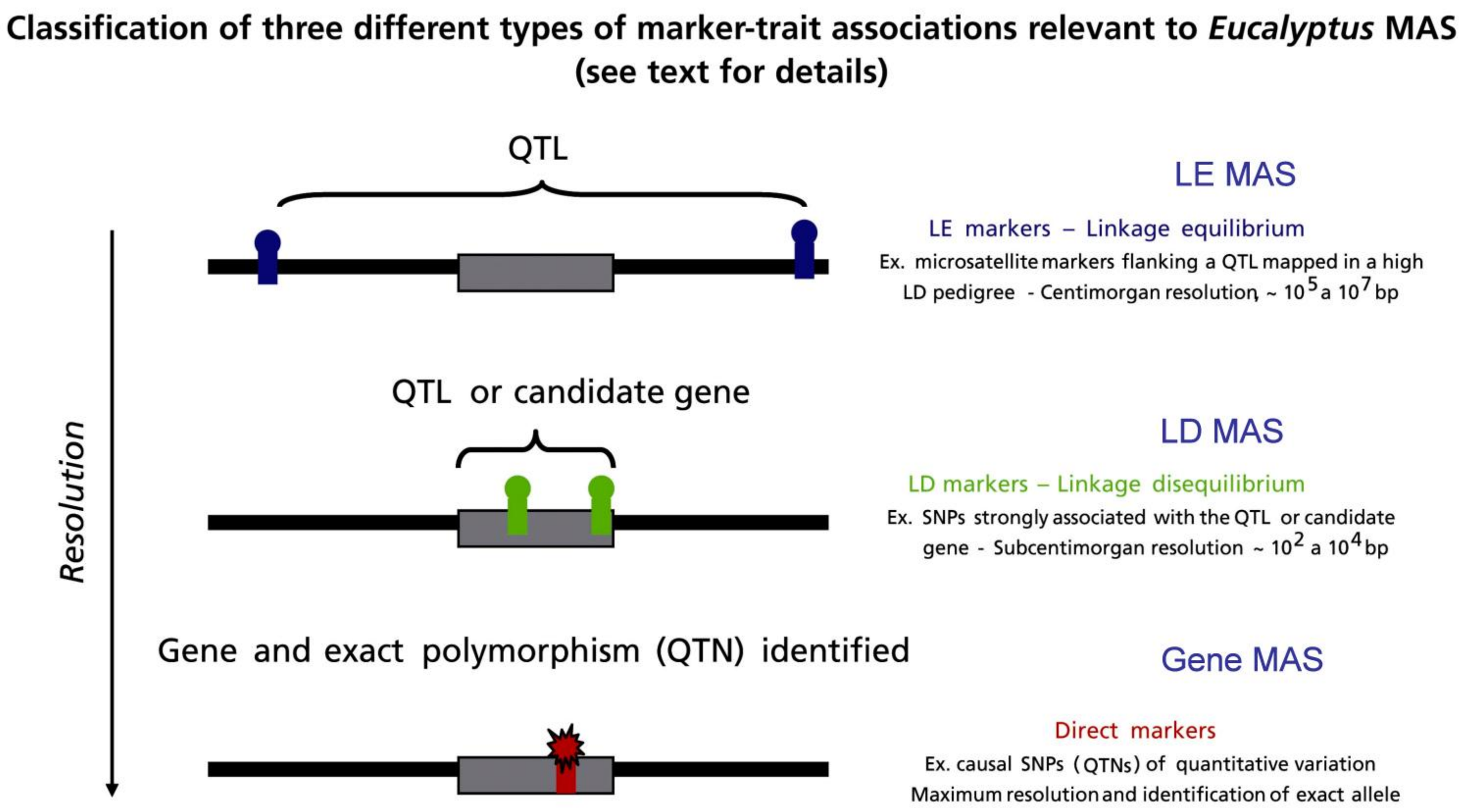
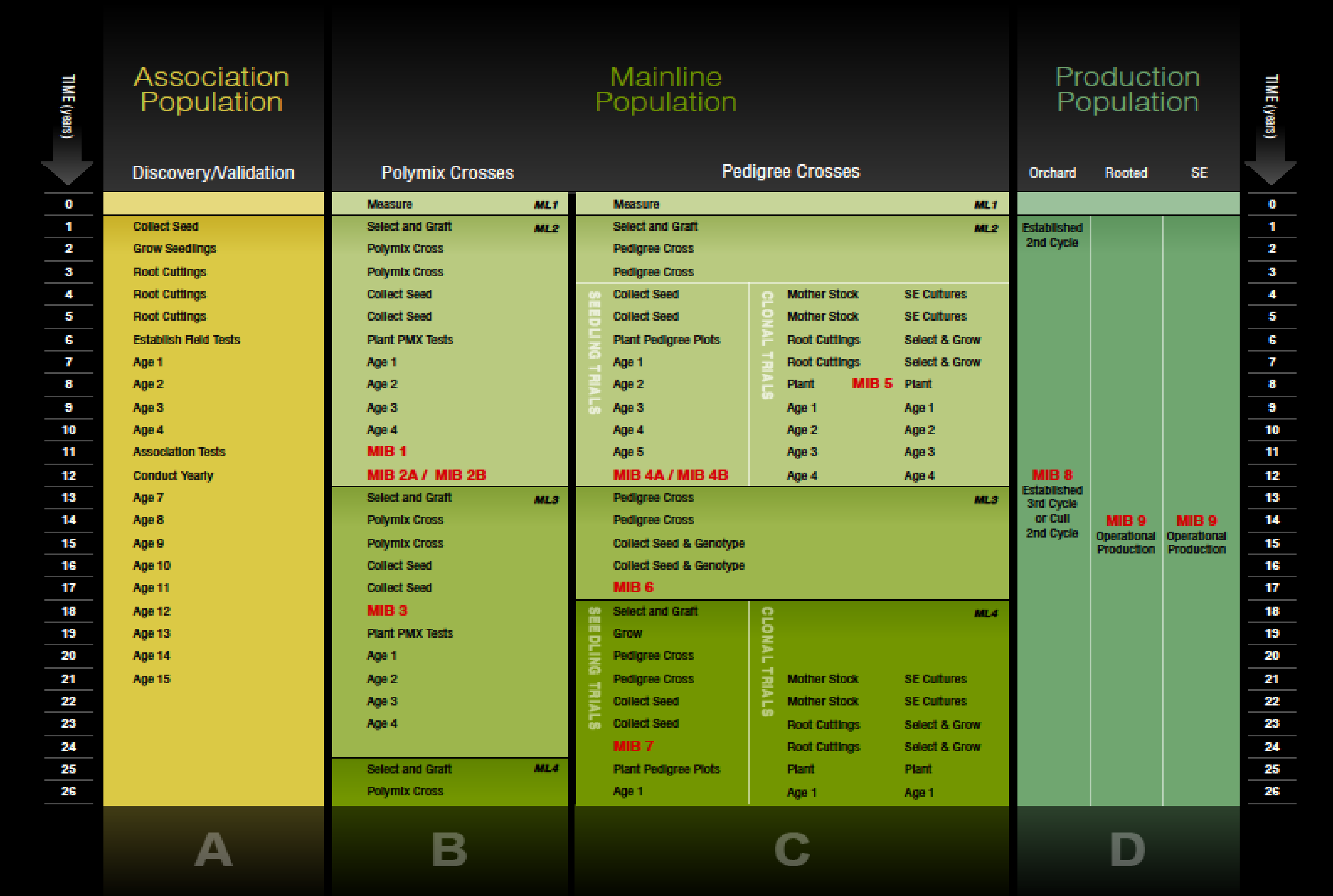


Figure Credit: Modified from Grattapaglia, 2007



www.pinegenome.org/ctgn

Figure Credit: Nicholas Wheeler, Oregon State University.

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External link

- Forest health initiative. [Online]. Available at: http://foresthealthinitiative.org (verified 2 June 2011).

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National Institute
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