Carya: The Next Generation

L. J. Grauke
Research Horticulturist
Curator, National Collection of Genetic Resources-Carya
USDA ARS Pecan Breeding & Genetics

Agricultural Research Service

Guiding Concept: Boundaries

- Teams
  - Multi-disciplinary, multi-institutional, multi-national
- Trees
  - Organization of Genus by Section, Species
  - Reproductive isolation
  - Geographic and genetic distributions
- Tests
  - Living collections, integrated effects, verified identities
- Targets
  - Markets
  - Clientele
- Time
  - Windows of opportunity
  - Looking backward, walking forward

Teams:

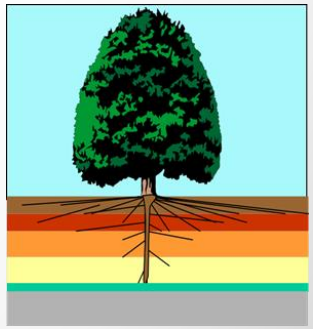
The National Plant Germplasm System

- The U.S. National Plant Germplasm System (NPGS) is a collaborative effort to safeguard the genetic diversity of agriculturally important plants.
- The NPGS is managed by USDA-ARS.
- Funding for the NPGS comes mainly via appropriations from the U.S. Congress.
- NPGS is a partnership between the public and private sectors, with land-grant universities prominently involved.
- USDA ARS designated the USDA ARS Pecan Breeding Program at Brownwood, Texas as the site of the National Clonal Germplasm Repository in 1978, based on the pecan cultivar collections developed by Louis Romberg, the first breeder, for use as parents in the Breeding Program.
- First meetings of Crop Germplasm Committee were in 1984, and elected a dynamic strategy of provenance collections and species collections in addition to cultivar collections.

USDA ARS Pecan Breeding & Genetics two interconnected projects:

- **The National Collection of Genetic Resources for Pecans and Hickories (collect, conserve)**
  - Collect worldwide sources of genetic diversity
  - Maintain living collections
  - Evaluate diversity to facilitate use
  - Document accessions
  - Distribute germplasm to appropriate domestic and international users

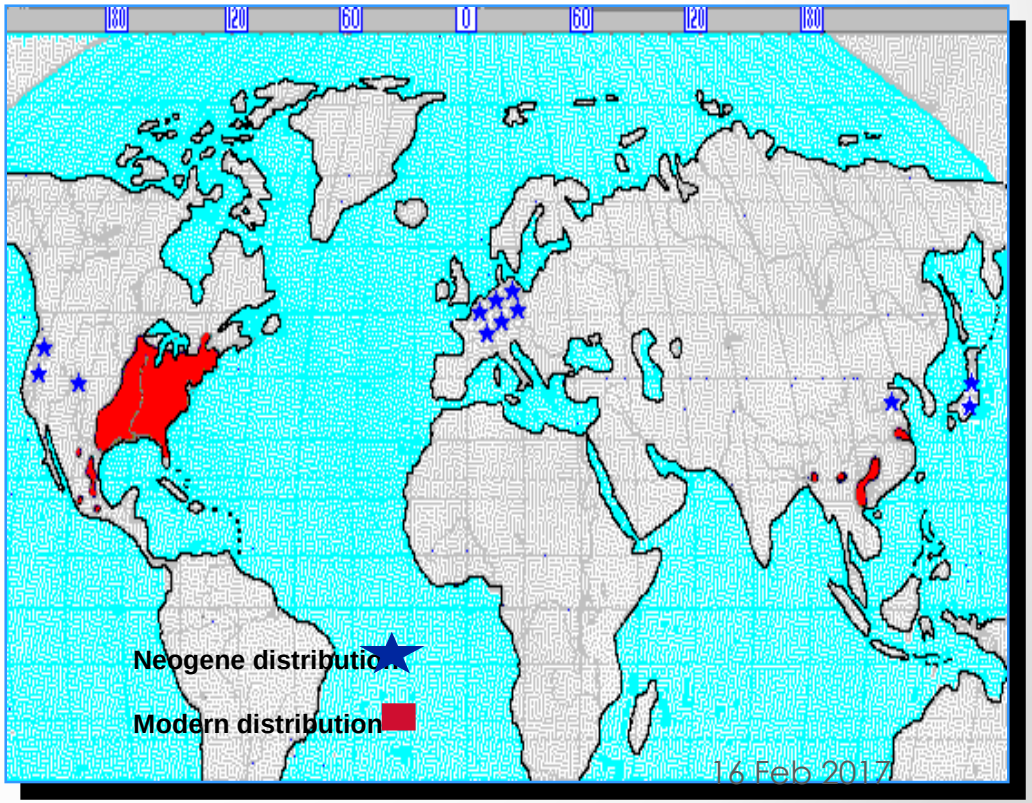
- **The Pecan Breeding Program (select, cull)**
  - Develop improved pecan scion cultivars
  - Develop improved pecan rootstocks



The National Collection of Genetic Resources for Pecans and Hickories

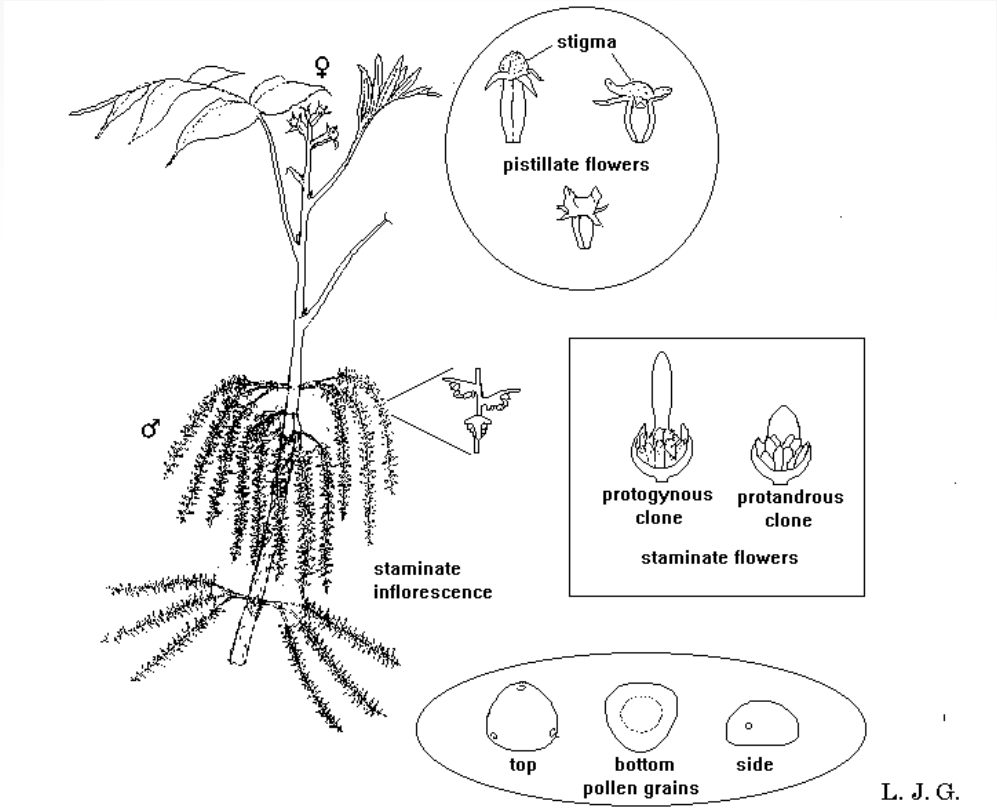
"Collection will be from worldwide sources of wild species and domestic cultivars to provide for maximum genetic diversity in each genus."

- There are 19 species of *Carya* worldwide.
  - 11 occur in the SE U.S.
  - 1 is endemic in Mexico.
  - 6 species are found only in Asia (where 1 other may be extinct).
- NCGR has 15 of the 19 species (10 U.S. , 1 Mexican, plus 4 Asian).



Taxa of the genus Carya, by sections

Section *species*	Common name	Distribution	Ploidy
Section Apocarya			
***C. aquatica*.**	**Water hickory**	**SE. U.S.**	**2n=2x=32**
C. cordiformis	**Bitternut hickory**	**E. U.S.**	**“**
C. illinoinensis	**Pecan**	**SW. to E. U.S. & Mex.**	**“**
C. palmeri	**Mexican hickory**	**Mex.**	**“**
Section Carya			
C. laciniosa	**Shellbark hickory**	**E. U.S.**	**2n=2x=32**
C. myristiciformis	**Nutmeg hickory**	**E. U.S. & Mex.**	**“**
C. ovata (+ C. car.-sep.)	**Shagbark hickory**	**E. U.S. & Mex.**	**“**
C. floridana	**Scrub hickory**	**Florida**	**2n=4x=64**
***C. glabra* (+ *C. ovalis*)**	**Pignut hickory**	**E. U.S.**	**“**
C. pallida	**Sand hickory**	**E. U.S.**	**“**
C. texana	**Black hickory**	**Central U.S.**	**“**
C. tomentosa	**Mockernut hickory**	**E. U.S.**	**“**
Section Sinocarya			
C. cathayensis	**Chinese hickory**	**E. China**	**2n=2x=32**
C. dabieshanensis	**Dabie Mt. hickory**	**E. China**	**“**
C. hunanensis	**Hunan hickory**	**C. China**	**“**
C. kwechowensis	**Guizhou hickory**	**C. China**	**“**
C. tonkinensis	**Vietnam hickory**	**China, India, Vietnam**	**“**
C. poilanei ?	**Poilane’s hickory**	**Laos & Vietnam**	**?**
Section Rhamphocarya (or genus Annamocarya)			
C. sinensis	**Beaked hickory**	**China & Vietnam**	



- Flowering is **heterodichogamous**
  - Male and female flowers on the same tree mature at different times
    - Protandrous trees mature male flowers first
    - Protogynous trees mature female flowers first
- **Cultivars** are **consistent** in the bloom pattern they show from year to year, but **seasons differ** in the inception of bloom and patterns of overlap between cultivars can vary.
- Thompson and Romberg (J. Hered. 76:456-458. 1985) described this as a simple dominant trait, with protogyny dominant over protandry. 'Mahan' is homozygous dominant, and all its progeny are protogynous.

Protandrous cultivar ‘Clark’ shedding pollen prior to receptivity 28 April 2003

Protogynous cultivar ‘Choctaw’ stigmas receptive prior to pollen shed 24 April 2003

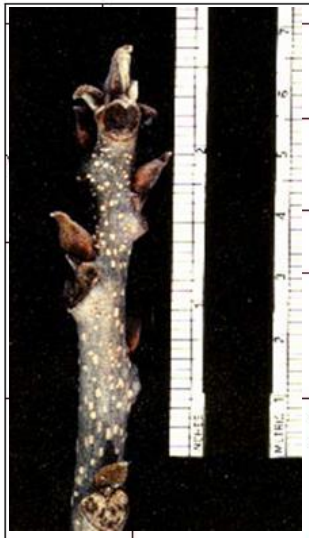
Flowering in Carya

- This flowering system encourages **cross pollination** and maintains **genetic diversity** in populations.
- Cross-pollinated nuts have higher percent kernel than self-pollinated nuts from the same tree.
- Cross-pollinated nuts produce more vigorous seedlings than self-pollinated nuts, giving those seedlings a selective advantage.

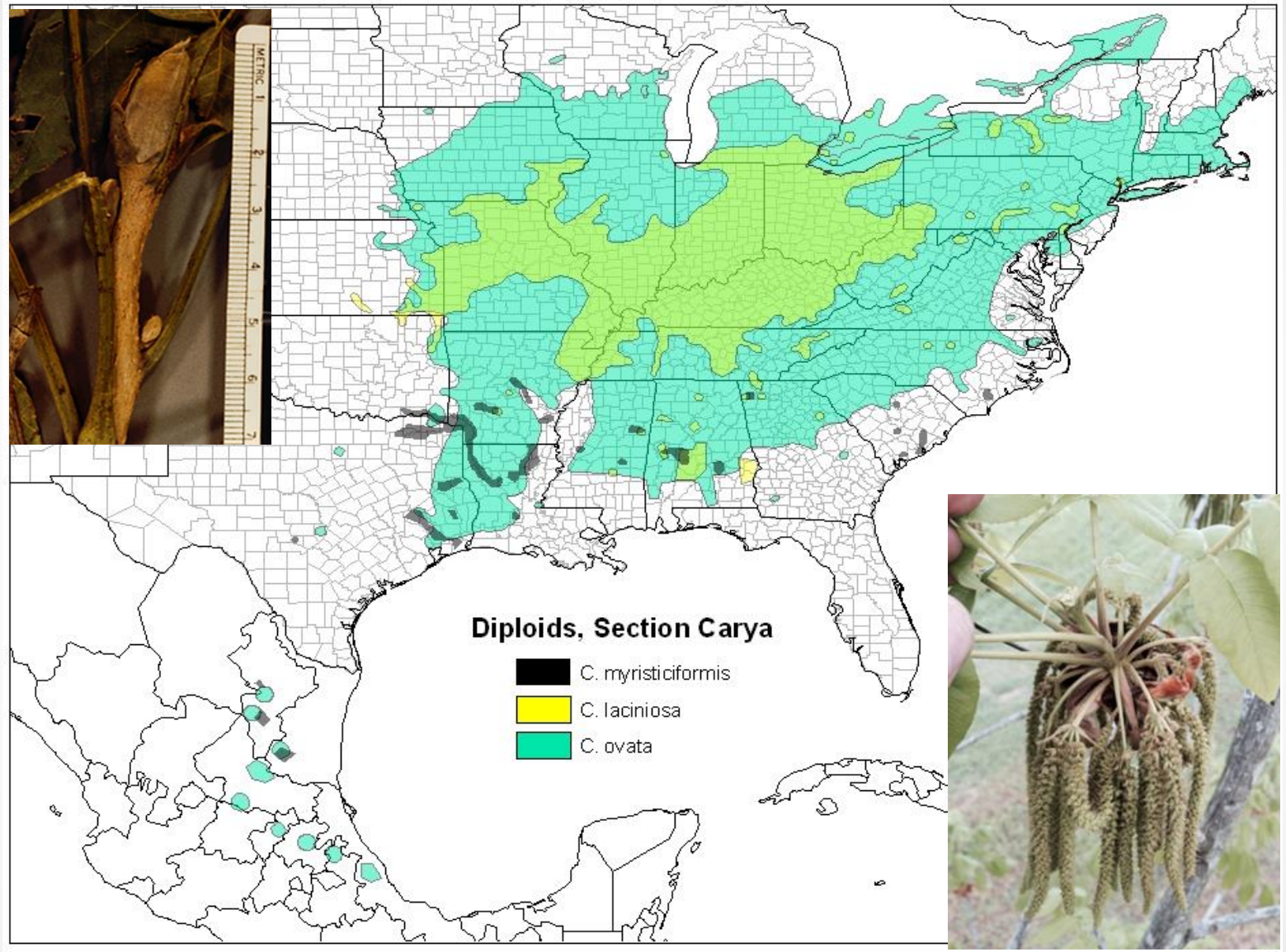
- This has implications for **forest diversity** at the broadest level, for the selection of cultivars to be used as **seed-stocks**, and for the cultivars to plant for **bloom overlap** in the orchard

Species maintain reproductive isolation by:

- Spatial separation due to site adaptation
  - Native pecan is riverine, primarily found on well-drained, bottomland sites.
  - *Carya aquatica* occurs on poorly drained, frequently flooded sites
  - Upland species (*C. tomentosa*, *C. texana*, *C. pallida*, *C. glabra*, *C. floridana*)inhabit xeric sites
- Temporal separation of bloom time
  - Inception of growth is typically early in upland species, late in more bottomland species, last in *C. aquatica*.
  - Inception of growth and bloom varies between geographic populations of the same species across provenances of origin, beginning in south.
- Incompatibility due to ploidy level
  - Sympatric species of the same ploidy level hybridize.



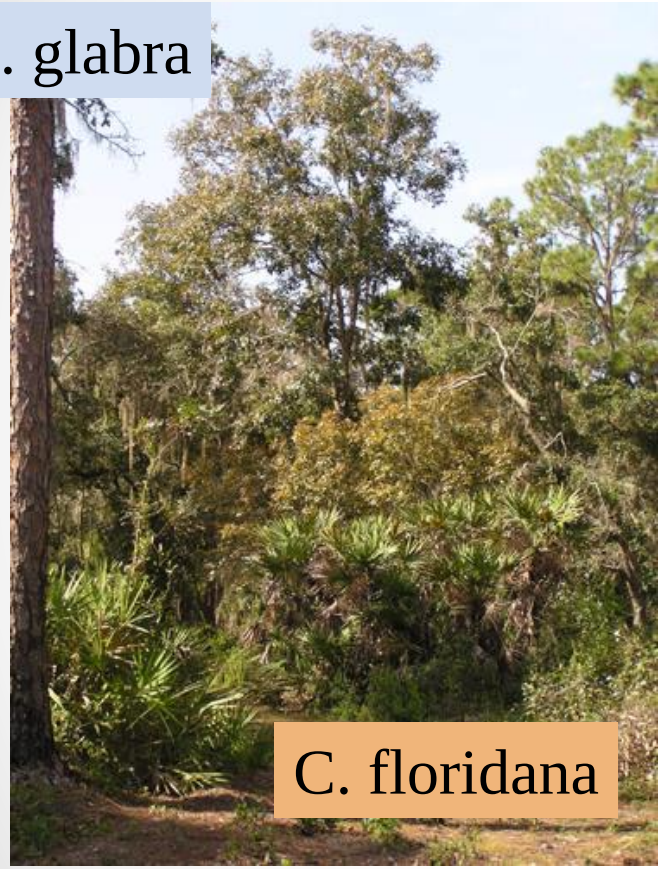
Diploids, Section Apocarya
Pecan samples
C. aquatica
Native Pecans
C. cordiformis
C. palmeri

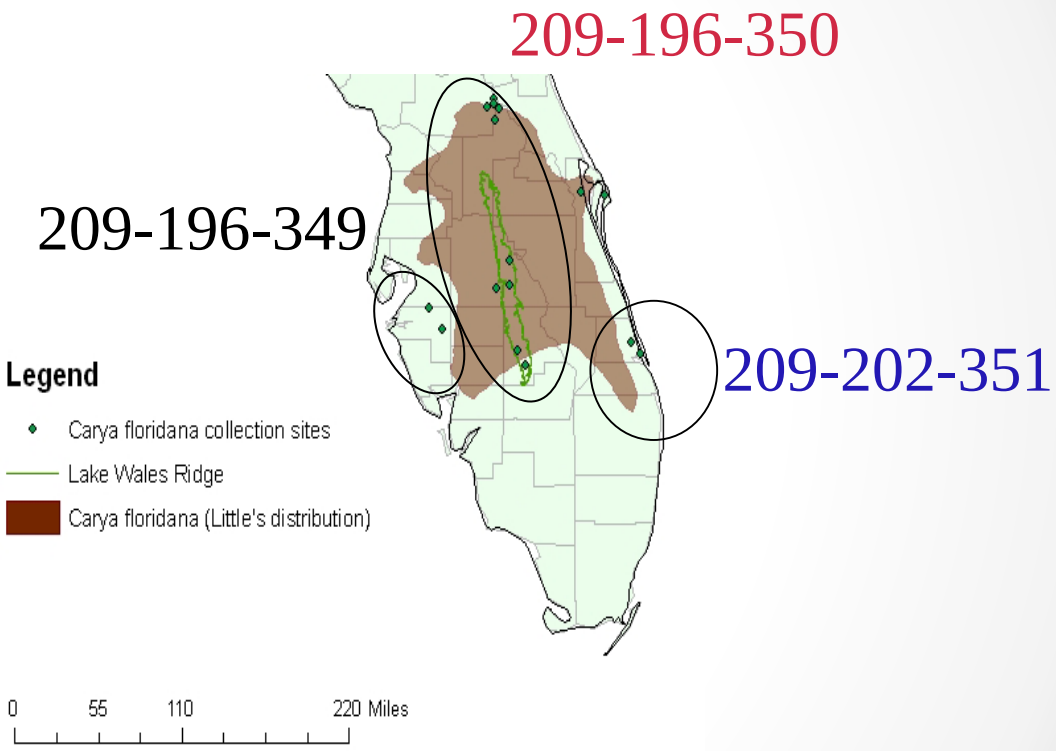

Diploids, Section Carya
C. myristiciformis
C. laciniosa
C. ovata

- All tetraploid Carya accumulate Rare Earth Elements.
- They seem to be adapted to more xeric sites.
- Tetraploidy may aid environmental stress adaptation.

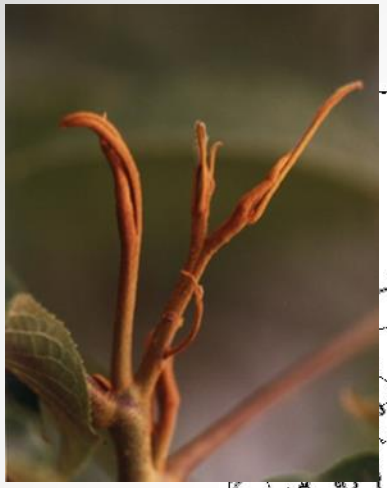
Wood and Grauke. J. Amer. Soc. Hort. Sci. 136(6):389–398. 2011.

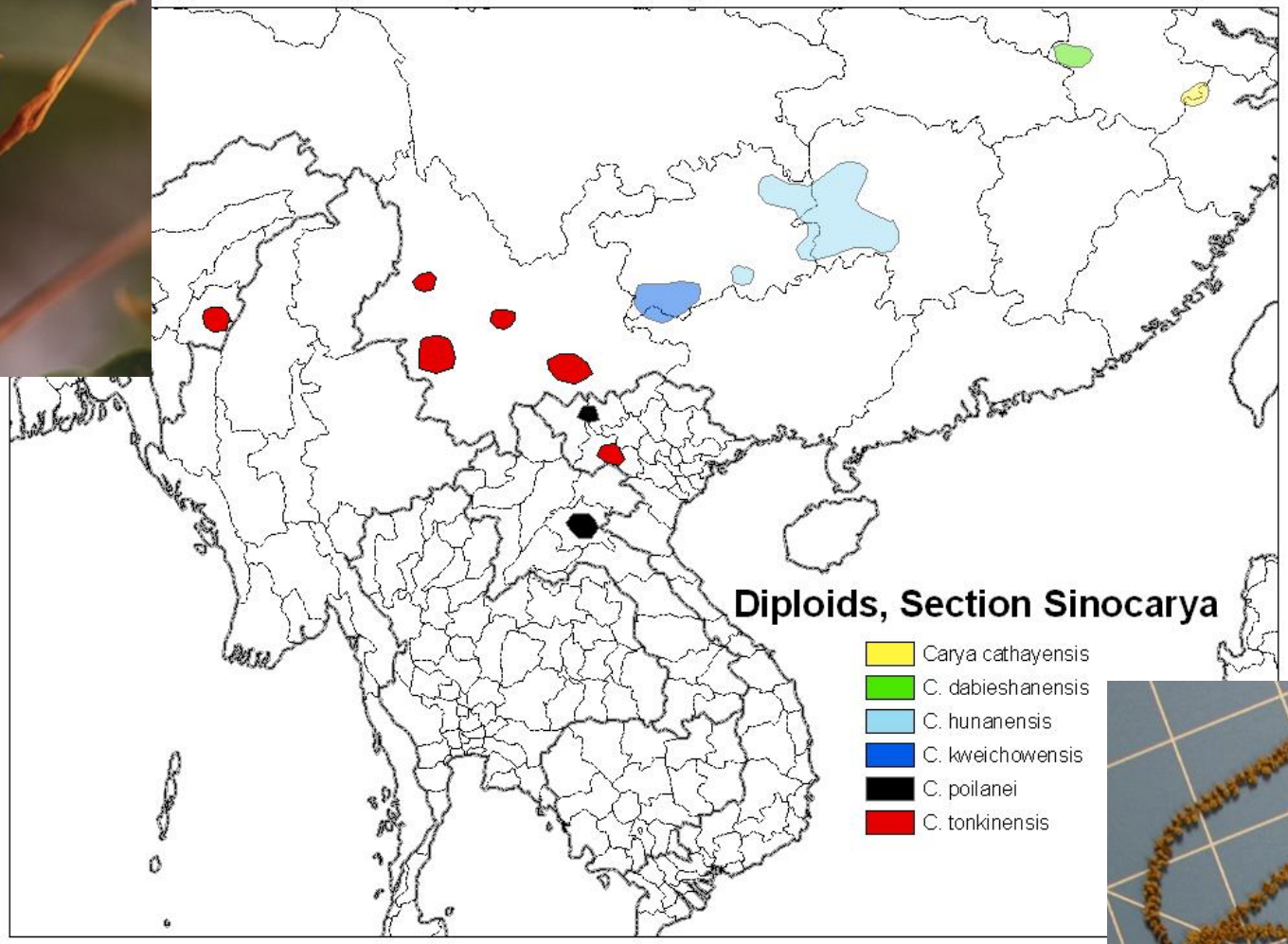
Tetraploid species of Carya show evidence of interspecific hybridization in sympatric populations

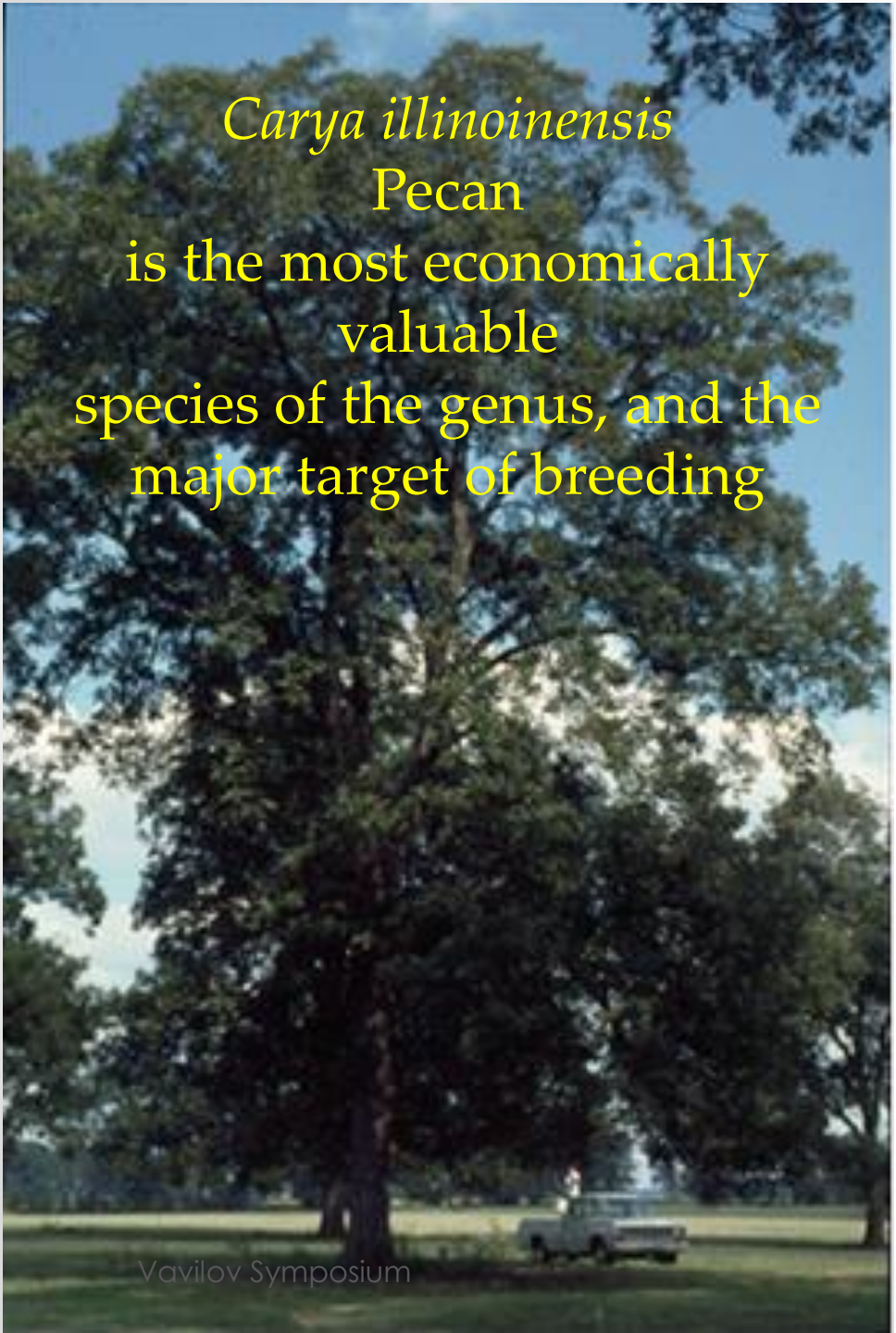




Maternally inherited plastid profiles differ between geographic populations, but are shared by sympatric *C. glabra* and *floridana*



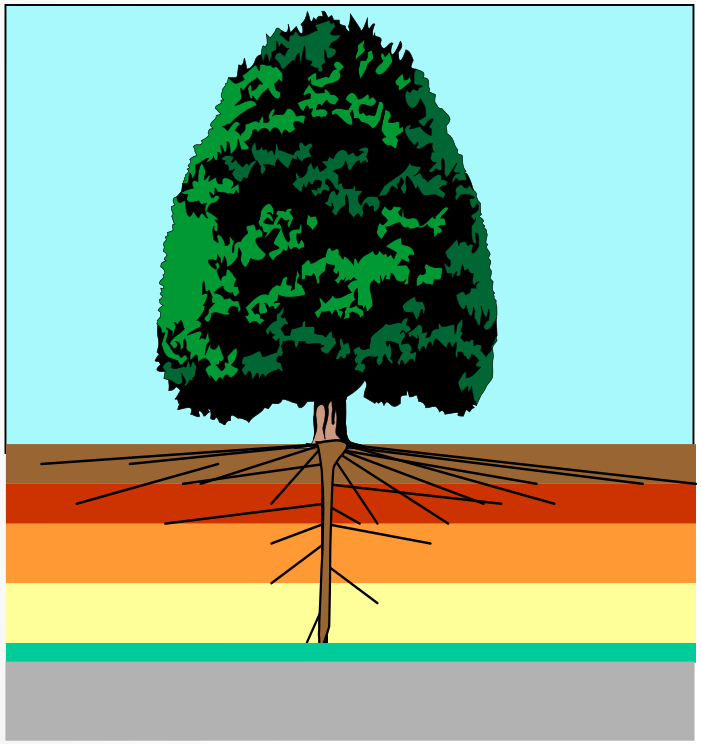

Diploids, Section Sinocarya
Carya cathayensis
C. dabieshanensis
C. hunanensis
C. kweichowensis
C. poilanei
C. tonkinensis



Challenges/Opportunities

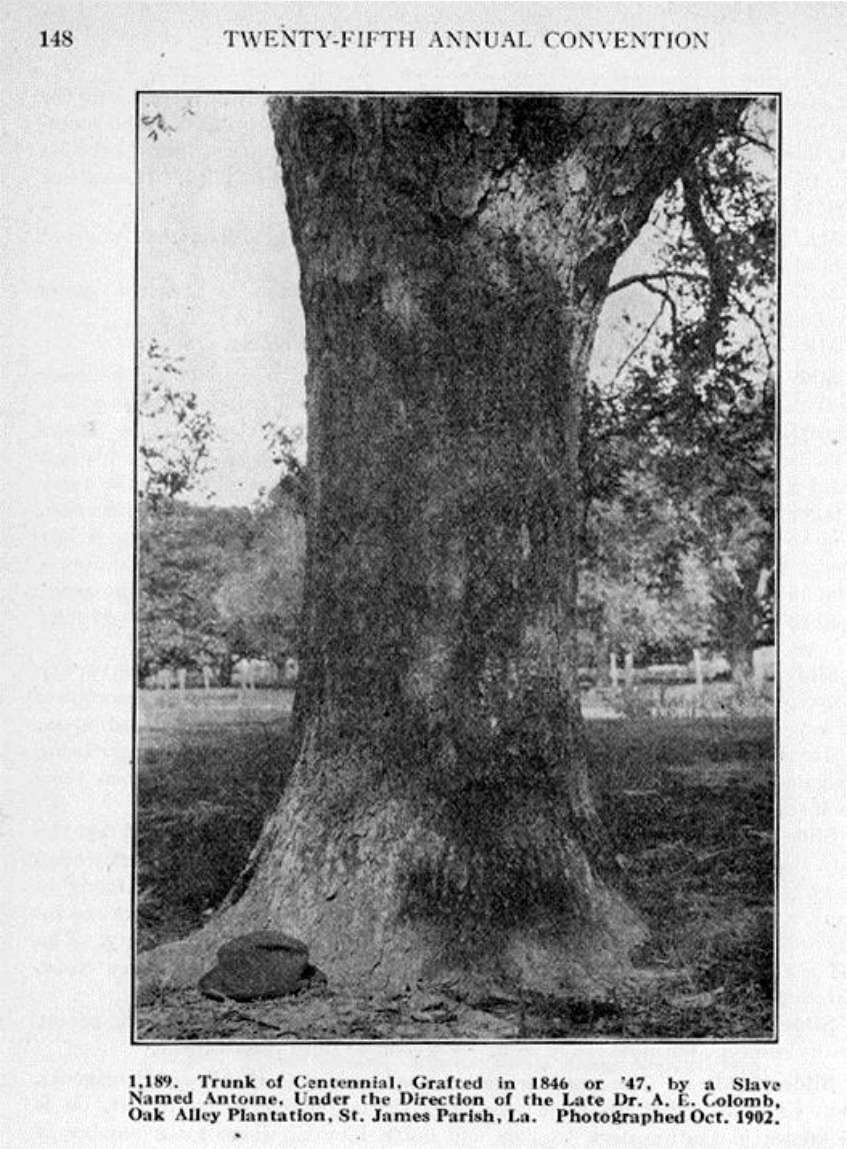
- Native species (co-evolved pests)
- Broadly distributed (17-42° N lat.)
- Regional adaptation
- Large tree (to 44 m)
- Long-lived (over 200 y)
- Slow to bear (5 to 12+ yr)
- Out-crossing (heterodichogamy)
- Wind pollinated
- In-breeding depression
- Graft propagation began 1846
- "Immortal" cultivars
- Site effects
- Rootstock effects
- Fragmented industry (by region, producers vs shellers)
- Fragmented research community
- Intellectual property issues isolate

Pecans are large, long-lived trees whose growth shows the integrated, cumulative effects of:



- **Genetics**
  - Rootstock
  - Scion
- **Environment**
  - Climate
  - Soils
  - Biotic factors
- **Culture**
  - Irrigation
  - Fertility
  - Pest Control

‘Centennial’


148 TWENTY-FIFTH ANNUAL CONVENTION

1,189. Trunk of Centennial, Grafted in 1846 or '47, by a Slave Named Antoine, Under the Direction of the Late Dr. A. E. Colomb, Oak Alley Plantation, St. James Parish, La. Photographed Oct. 1902.

By linking a molecular profile to a verified inventory, we facilitate future recognition.

Pecan Cultivar Collection

- Over 350 grafted pecan accessions are maintained at the two worksites.
- Most accessions have been verified using a combination of techniques: origination from trustworthy sources; morphological verification based on known nut size and shape; isozyme profiles that confirm known parentage, microsatellite profiles that confirm parentage or relation to controlled cross progeny.
- Inventories have been seasonally monitored for phenology of budbreak, flowering, nut maturation, nut quality and patterns of disease expression.

The Pecan Breeding Program

1931-the 21st Century

- Twenty-nine cultivars have been released by the USDA ARS Pecan Breeding Program.
- After the first, 'Barton', all were given the names of Native American tribes, in honor of the native origin of these important forest trees.
- The first releases were known for high nut quality and increased production.
- In 1984, 'Pawnee' inaugurated early harvest, and has altered management and marketing.
- 'Wichita' has shown the highest sustained yields
- 'Lakota' and 'Kanza' have excellent scab resistance.

‘Wichita’

'Halbert' X 'Mahan‘made in 1940 by L. D. Romberg, USDA-ARS, Brownwood, TX.

First fruited in 1947, tested as 40-9-193. Released in 1959.

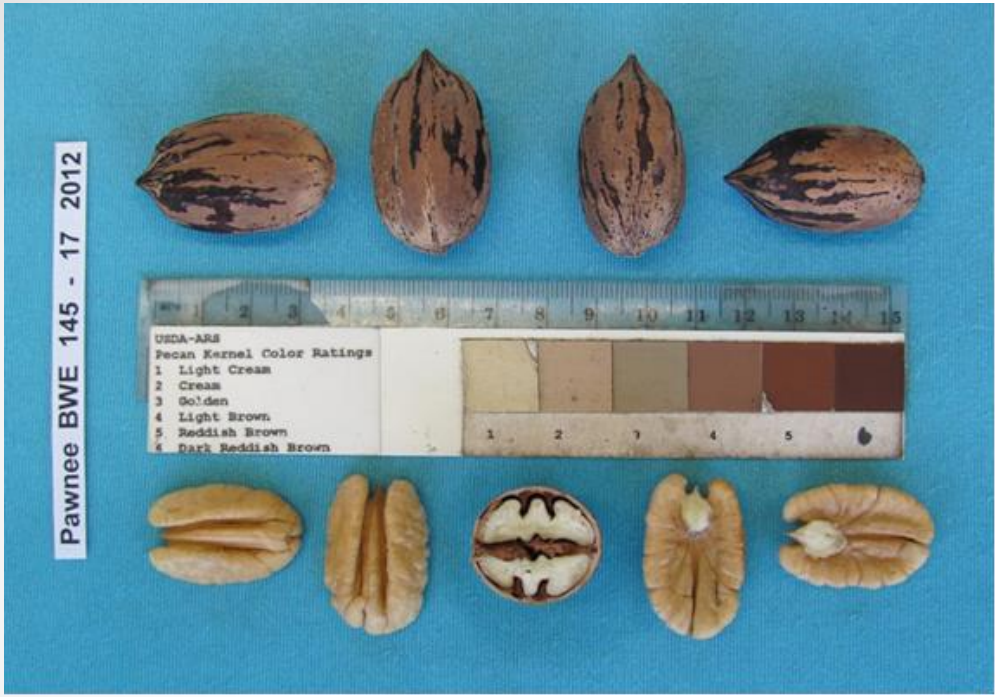
Precocious and prolific.

Very susceptible to pecan scab (caused by *Venturia effusa* = *Fusicladium effusum* Winter).

43 nuts/lb, 62% kernel

‘Wichita’ is a standard cultivar for use in the western US, working well as a protogynous cultivar with the regional favorite ‘Western’. In the arid west its scab susceptibility is not a limitation. ‘Wichita’ produces consistent high yields of high quality nuts.

‘Pawnee’



'Mohawk' X 'Starking Hardy Giant' made in 1963 by L. D. Romberg, USDA-ARS, Brownwood, TX.

Selected and tested by Tommy E. Thompson and R. E. Hunter as 63-16-125 and released in 1984.

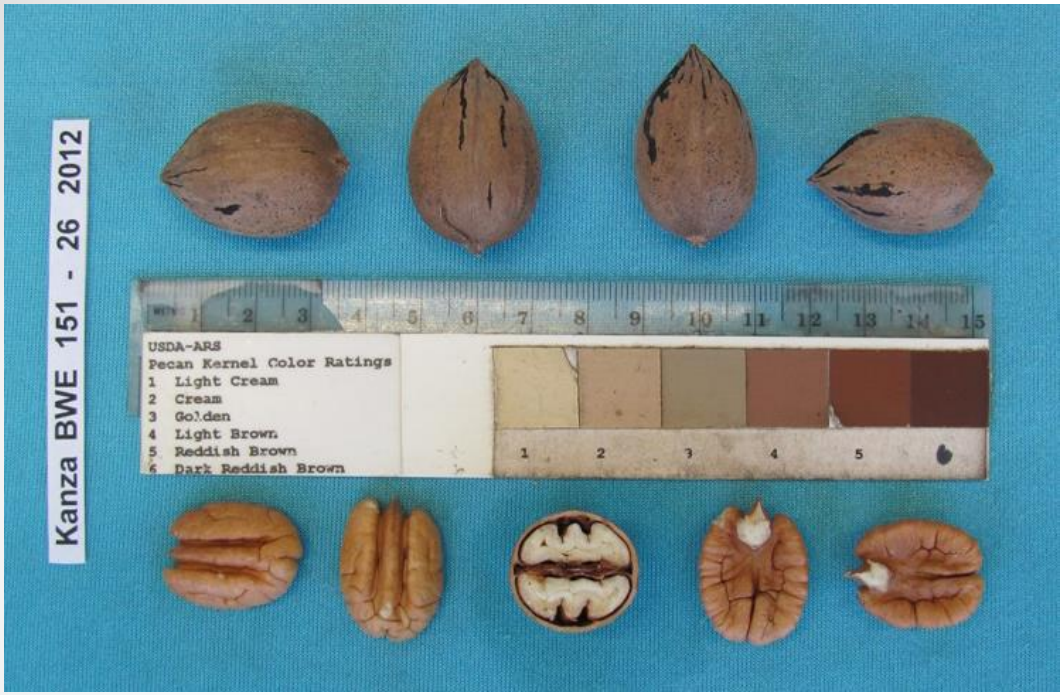
Nut matures early, mid Sept. in College Station, TX.

44 nuts/lb, 58% kernel

Although 'Pawnee' is susceptible to pecan scab, the disease is relatively easy to control using a prophylactic fungicide spray schedule. ‘Pawnee’ has outstanding resistance to yellow aphids.

‘Pawnee’ is overtaking ‘Desirable’ to become the most commonly planted cultivar in Georgia pecan orchards. (Wells, L. 2014. Pecan planting trends in Georgia. HortTechnology 24:475-479.)

‘Kanza’



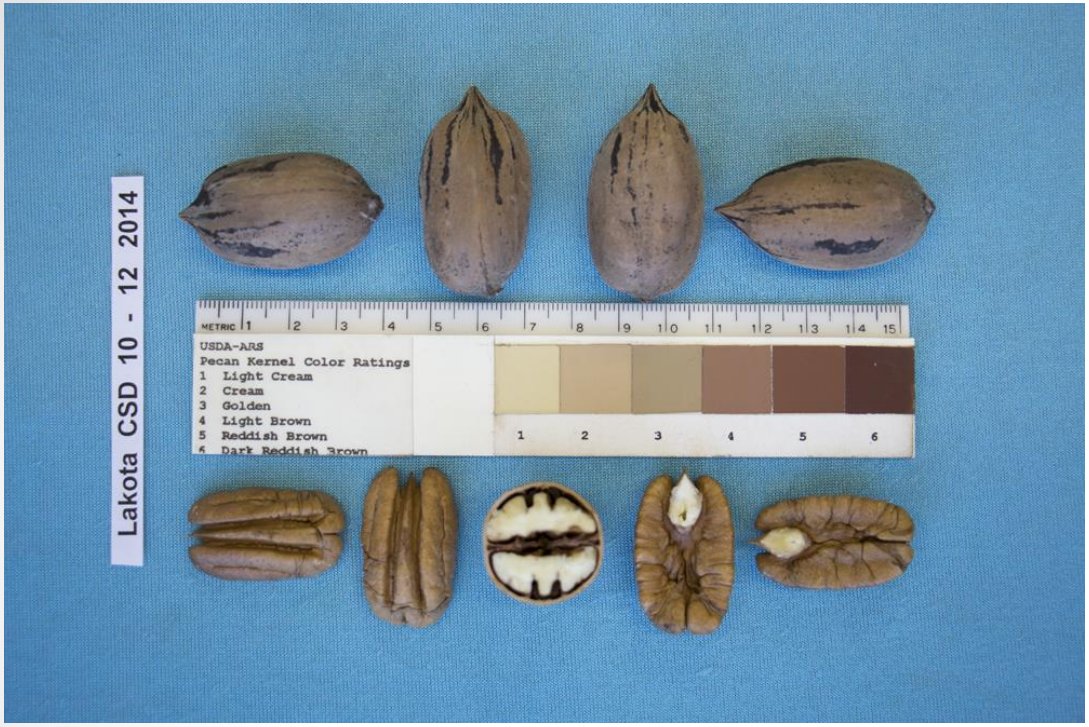
'Major' X 'Shoshoni' made in 1955 by L. D. Romberg, USDA ARS, Brownwood, TX.

Selected and distributed as 55-11-11 by G. M. Madden. Tested by T. E. Thompson, L. J. Grauke, Wm. Reid, M. W. Smith and S. R. Winter and released in 1996.

77 nuts/lb, 54% kernel

Released for its ability to produce good yields of high quality nuts in the northern pecan producing regions. ‘Kanza’ has excellent scab resistance and is suggested as a replacement for ‘Elliott’ (Wells, L. and P. J. Conner. 2015. Pecan varieties for Georgia orchards. UGA Extension Circular 898).

‘Lakota’



'Mahan' X 'Major' made in 1964 by L. D. Romberg, USDA-ARS Brownwood, Texas.

Selected by G. D. Madden. Tested as 64-6-502 by G.D. Madden, T. E. Thompson, L. J. Grauke and Wm Reid and released cooperatively by USDA-ARS and KSU in 2007.

59 nuts/lb, with 62% kernel.

‘Lakota’ performed well in tests in the northern pecan production areas of Kansas, Missouri, Illinois, Oklahoma and Texas.

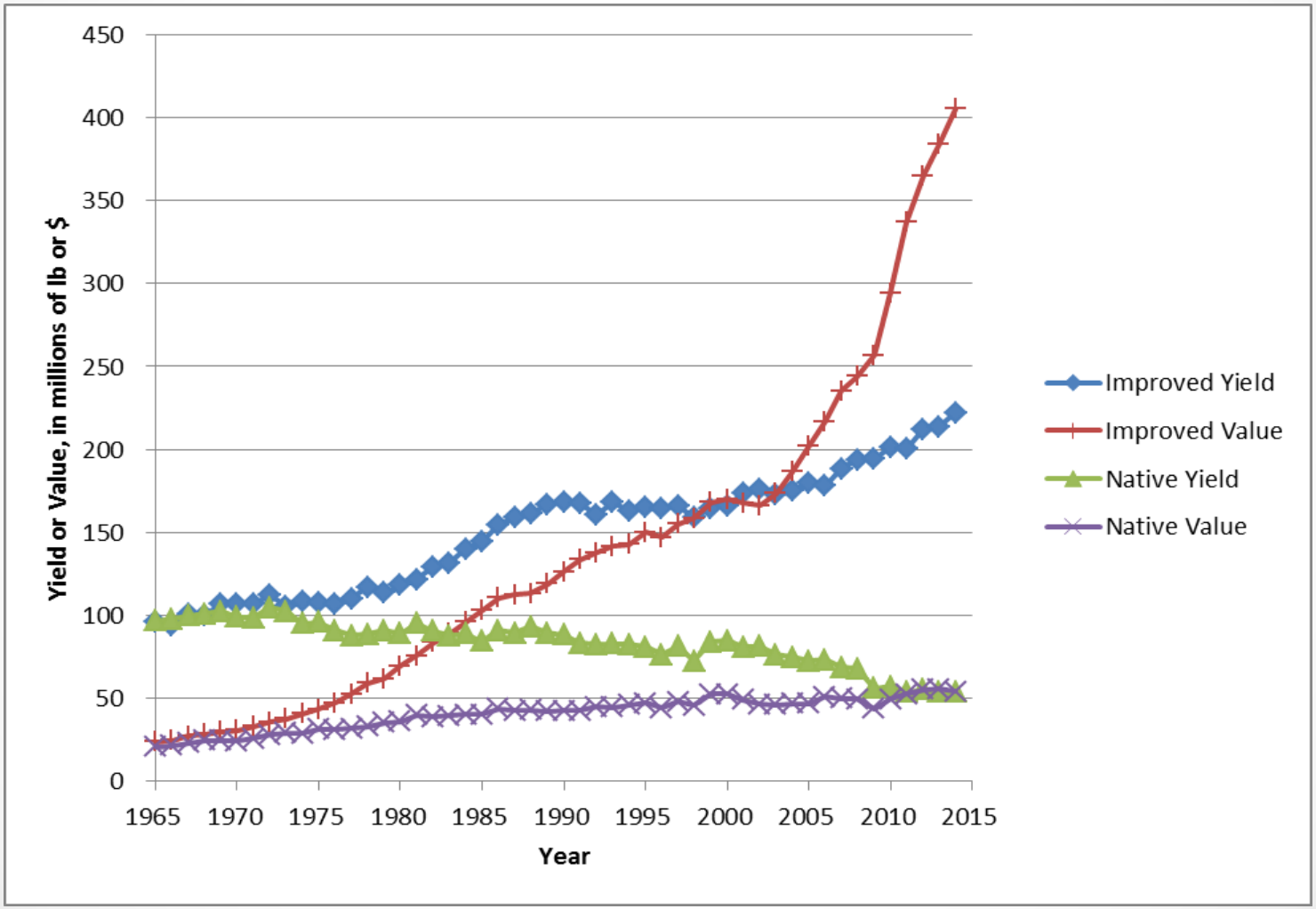
‘Lakota’ is very resistant to pecan scab disease. It is recommended for trial planting in Georgia (Wells, L. and P. J. Conner. 2015. Pecan varieties for Georgia orchards. UGA Extension Circular 898).

No.	CULTIVAR	FEMALE PARENT	MALE PARENT	CROSS #	RELEASED	Yrs to rel.	Breeder	Avg
1	Barton	Moore	Success	1937-03-0020	1953	16	Romberg	
2	Comanche	Burkett	Success	1938-07-0022	1955	17	Romberg	
3	Choctaw	Success	Mahan	1946-15-0276	1959	13	Romberg	
4	Wichita	Halbert	Mahan	1940-09-0193	1959	19	Romberg	
5	Apache	Burkett	Schley	1940-04-0017	1959	19	Romberg	
6	Sioux	Schley	Carmichael	1943-04-0006	1962	19	Romberg	
7	Mohawk	Success	Mahan	1946-15-0195	1965	19	Romberg	
8	Caddo	Brooks	Alley	Philema1175 (1922)	1968	46	Romberg	
9	Shawnee	Schley	Barton	1949-17-0166	1968	19	Romberg	20.8
10	Cheyenne	Clark	Odom	1942-13-0002	1970	28	Madden	
11	Cherokee	Schley	Evers	1948-22-0027	1971	23	Madden	
12	Chickasaw	Brooks	Evers	1944-04-0101	1972	28	Madden	
13	Shoshoni	Odom	Evers	1944-15-0059	1972	28	Madden	
14	Tejas	Mahan	Risien #1	1944-10-0293	1973	27	Madden	
15	Kiowa	Mahan	Desirable	1953-09-0191	1976	23	Madden	26.2
16	Pawnee	Mohawk	St H Giant	1963-16-0125	1984	21	Thompson	
17	Houma	Desirable	Curtis	1958-04-0061	1989	31	Thompson	
18	Osage	Major	Evers	1948-15-0003	1989	41	Thompson	
19	Oconee	Schley	Barton	1956-07-0072	1989	33	Thompson	
20	Navaho	Apalachee	Wichita	1974-01-0011	1994	20	Thompson	
21	Kanza	Major	Shoshoni	1955-11-0011	1996	41	Thompson	
22	Creek	Mohawk	Western	1961-06-0067	1996	35	Thompson	
23	Hopi	Schley	McCulley	1939-05-0050	1999	60	Thompson	
24	Nacono	Cheyenne	Sioux	1974-05-0055	2000	26	Thompson	
25	Waco	Cheyenne	Sioux	1975-05-0006	2005	30	Thompson	
26	Lakota	Mahan	Major	1964-06-0502	2007	43	Thompson	
27	Mandan	BW-1	Osage	1985-01-0002	2009	24	Thompson	
28	Apalachee	Moore	Schley	1948-13-0311	2009	61	Thompson	
29	Lipan	Cheyenne	Pawnee	1986-03-0624	2011	25	Thompson	35.1

‘Schley’ is the foundation parent for the improved pecan industry

- It is the most frequent female parent of released cultivars.
- It’s progeny ‘Mahan’ is also a frequent parent of released cultivars.
- Of the 29 cultivars released by USDA ARS, 20 trace back to ‘Schley’.
- Both ‘Pawnee’ and ‘Wichita’ descend from ‘Schley’, and they are the dominant parents in the nurseries of trees currently being evaluated.
- The University of Georgia has recently patented at least 4 progeny of ‘Wichita’ X ‘Pawnee.’

Fifty years of yield and value (10 yr. running avg.) Improved vs Native Pecans



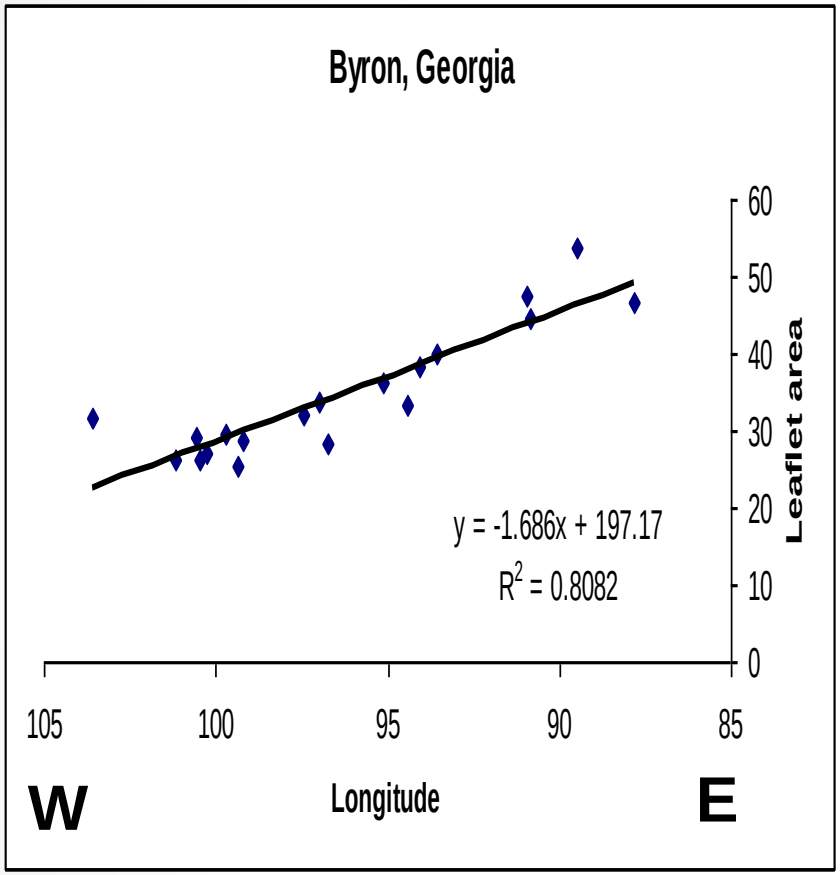
Pecan Provenance Tests

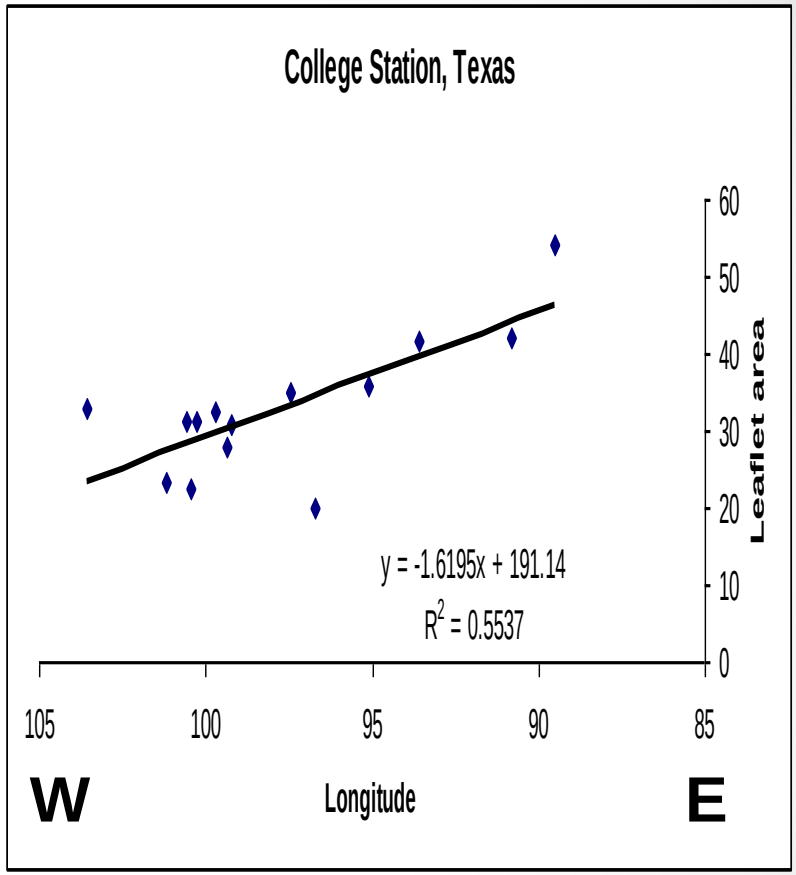
- At the first meetings of the Carya CGC, plans were made to collect from native pecan populations across the range of the species. Ruben Castro (CONAFRUT, MX) shared information on the distribution of Mexican native pecans, within Vavilov's Zone 7.
- Nut collections were made across the US in 1986 and across Mexico in 1987.
- Attempts were made to collect 100 nuts from each of 5 mother trees, separated by at least 200 m.
- Nuts were collected from 94 trees representing 19 native populations distributed from Missouri south to Oaxaca, Mexico.
- Replicated orchards of the full collection were planted in Byron, GA with partial plantings in Burleson and Brown County, Texas.

Patterns of leaf morphology vary by population of origin

- For two years, leaf samples were collected from each tree in the Byron and College Station Provenance orchards (mid leaflets of mid leaf in current season's growth).
- For each sample, ten leaflets were weighed, their area was determined, they were dried and reweighed.
- Leaf analysis was done by the Soil Test & Plant Analysis Lab, Louisiana Agri. Exp. Sta.

Leaflet area increases from west to east

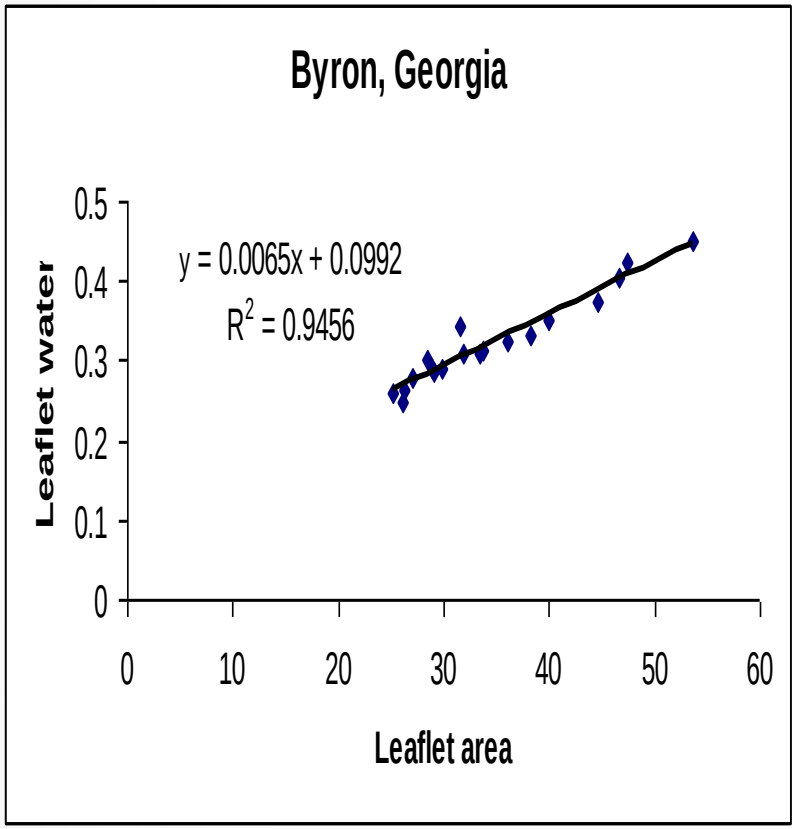




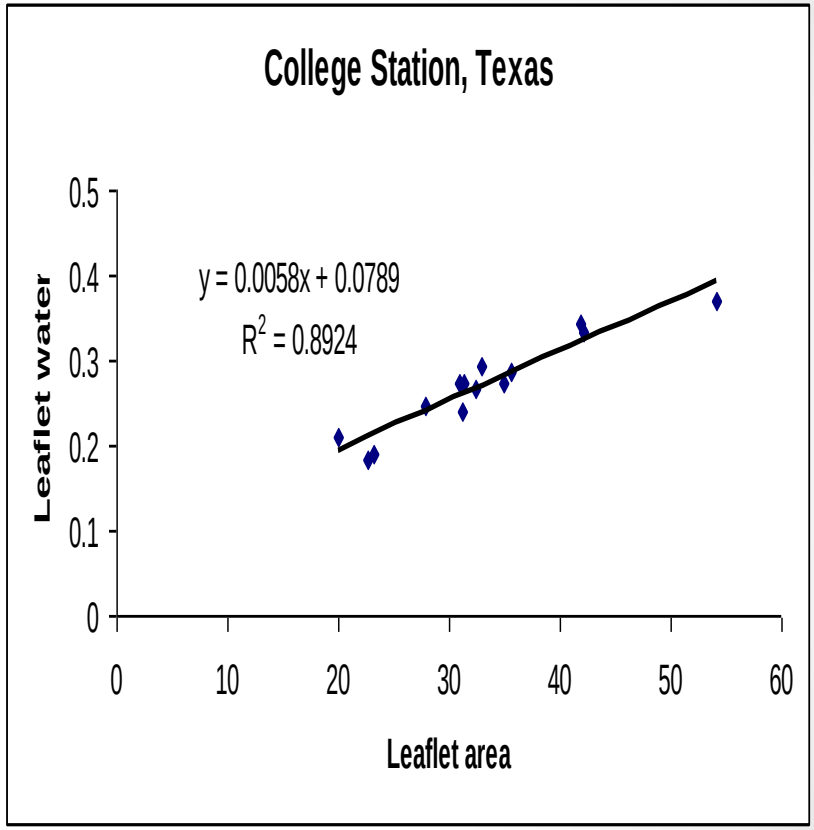
n = 19 populations, with 4-5 mother trees per population, each contributing seedlings in up to 11 blocks, with a total of over 900 trees observed in each of 2 years.

n= 14 populations, with 1-5 mother trees per population, each contributing seedlings in up to 8 blocks, with a about 200 trees observed in each of 2 years

Increased leaflet area is related to increased water in the leaflet.

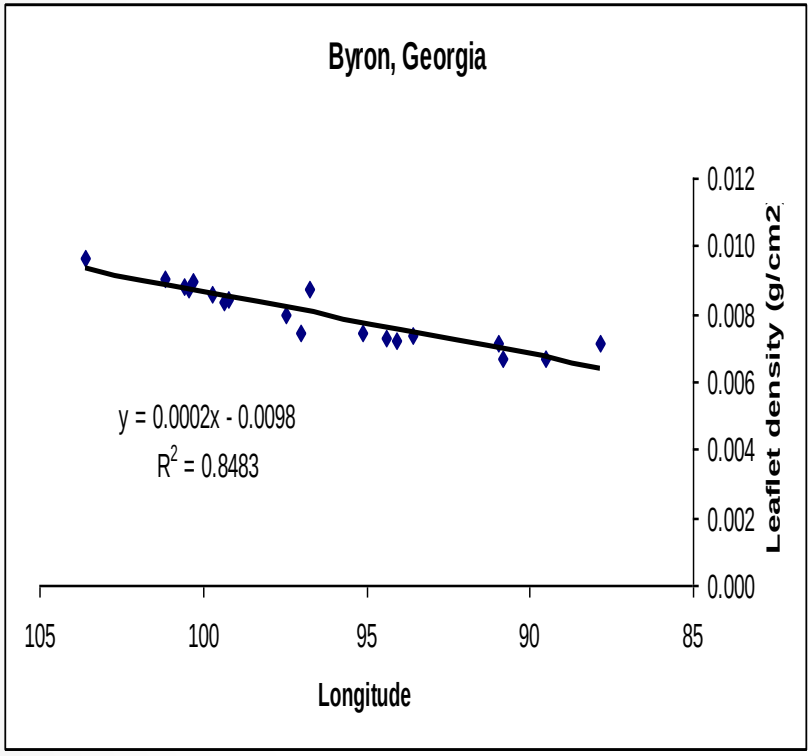


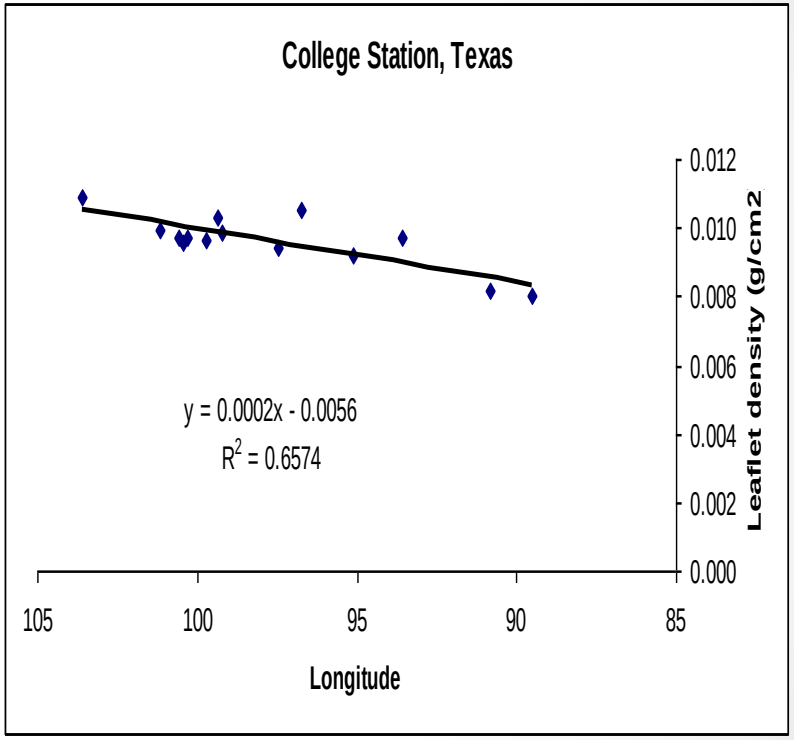
n = 19 populations, with 4-5 mother trees per population, each contributing seedlings in up to 11 blocks, with about 950 trees observed in each of 2 years.



n= 14 populations, with 1-5 mother trees per population, each contributing seedlings in up to 8 blocks, with a about 176 trees observed in each of 2 years

Leaflet density is greatest in seedlings originating from western trees, and is apparently greater in the College Station orchard than Byron.





n = 19 populations, with 4-5 mother trees per population, each contributing seedlings in up to 11 blocks, with a total of over 900 trees observed in each of 2 years.

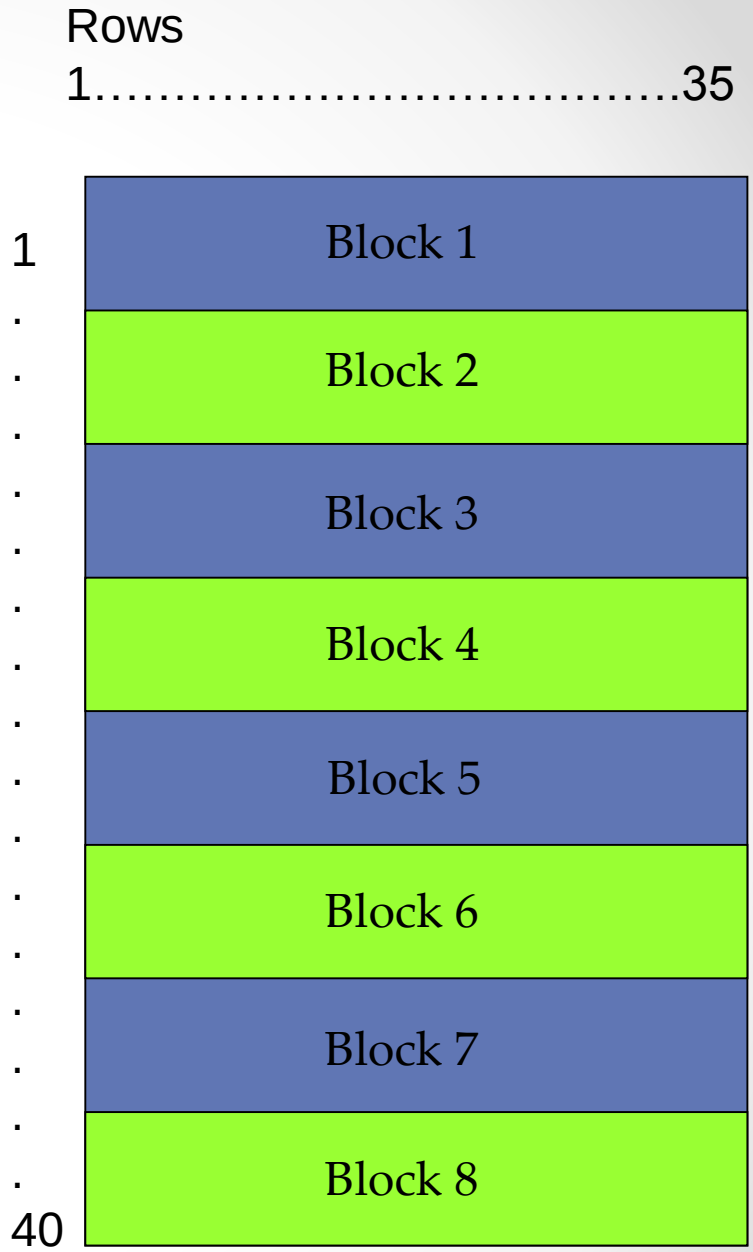
n= 14 populations, with 1-5 mother trees per population, each contributing seedlings in up to 8 blocks, with a about 200 trees observed in each of 2 years

Geographic patterns of disease resistance

- Recent research from the Byron, GA Provenance Orchard, showed differences in scab susceptibility between geographic populations, with the lowest levels (greatest resistance) from sources north of Texas.
- Leaf characteristics varied with provenance origin, but association with scab was not established.
- Bock, C.H., L.J. Grauke, P.J. Conner, S.L. Burrell, M.W. Hotchkiss, D.L. Boykin and B.W. Wood. 2016. Scab susceptibility of a provenance collection of pecan in three different seasons in the Southeastern USA. Plant Disease (Plant Disease 100:1937-1945.)

CSM Orchard

- Planted 1997
- 1400 trees
- 35 rows, 15 ft between rows,
- 40 trees per row, 18 ft between trees
- 8 blocks, each with 5 rows, 175 trees



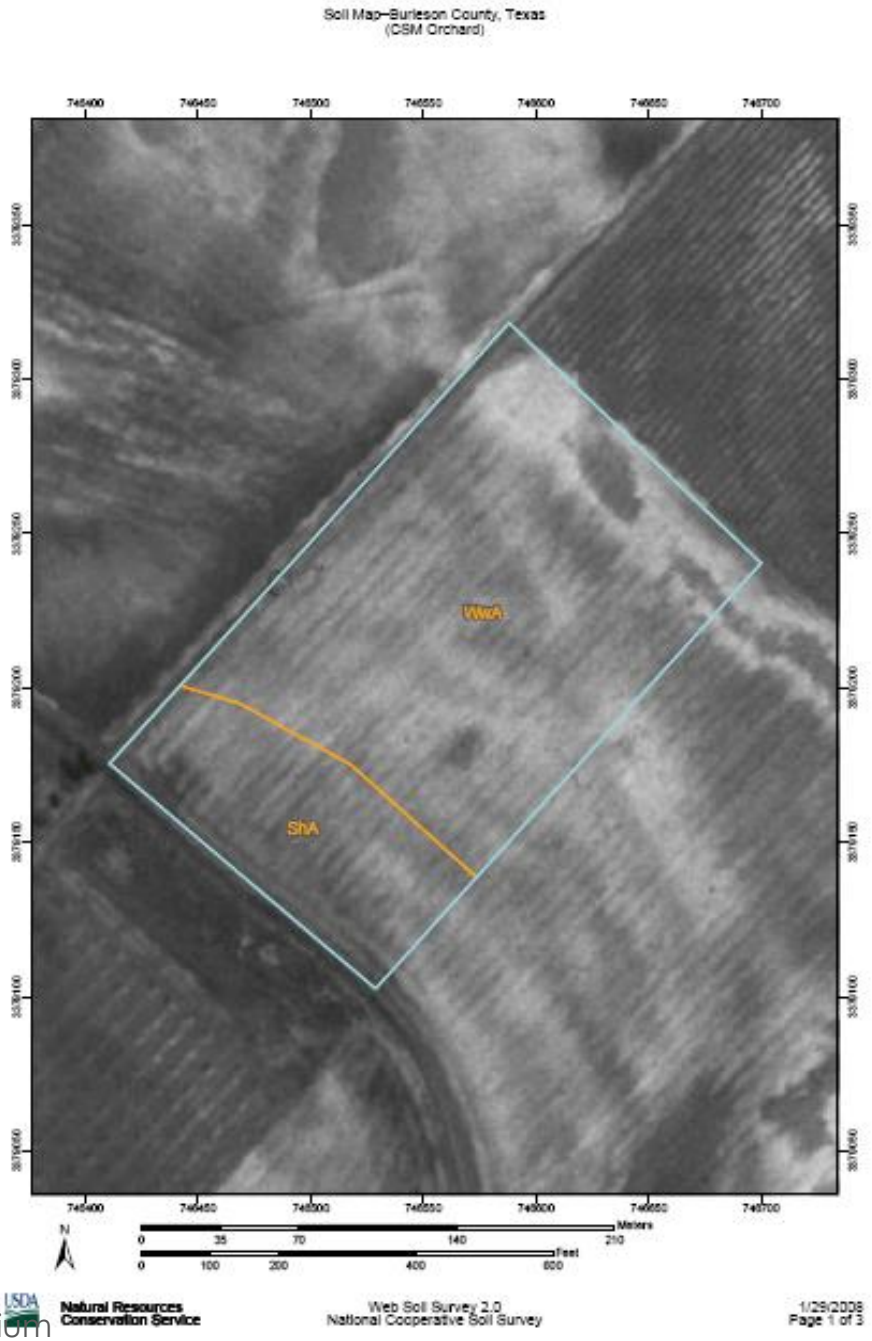
WwA Weswood silty clay loam, 0-1% slopes

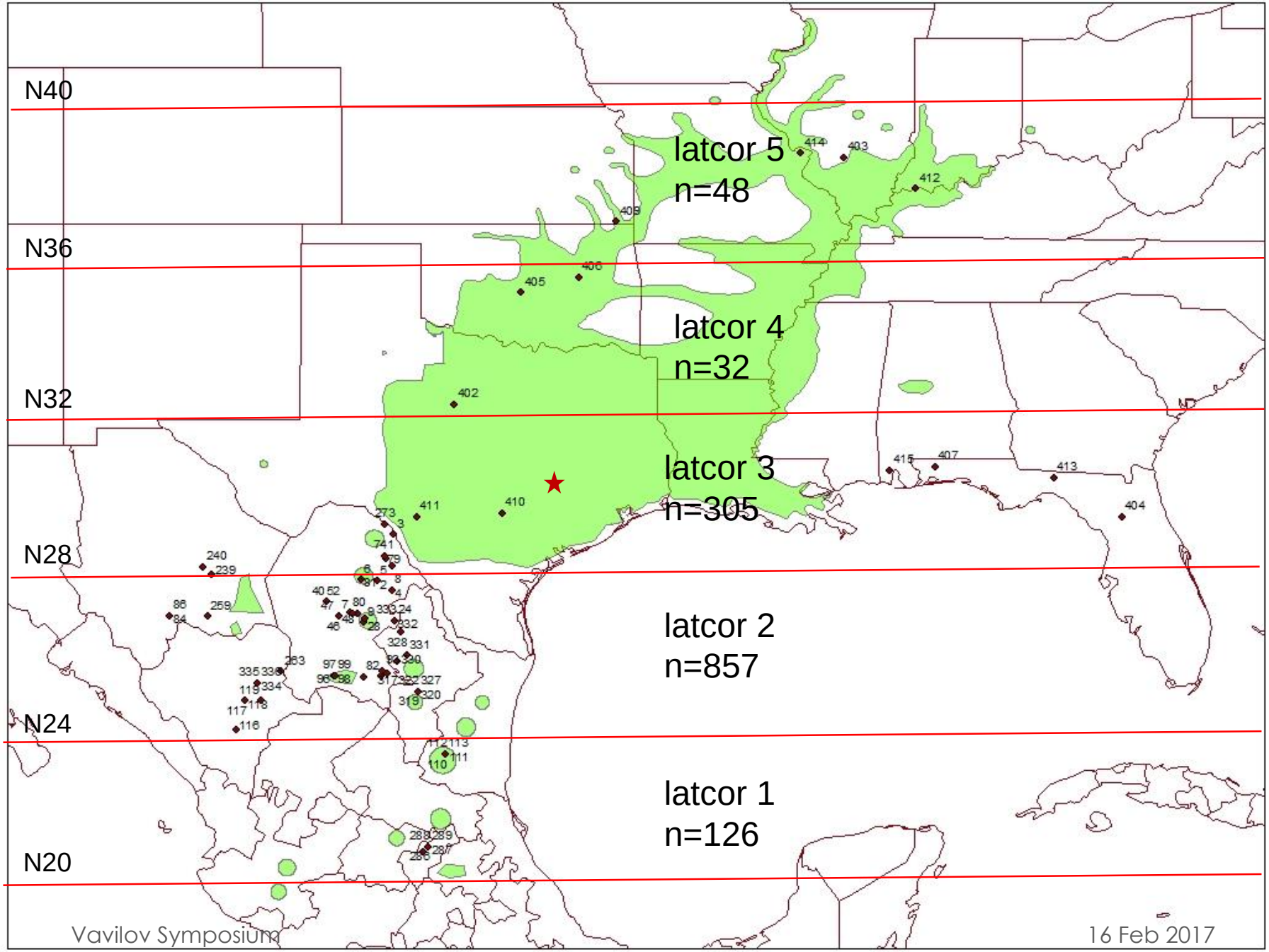
7.5 Acres

ShA Ships clay, 0-1 % slopes

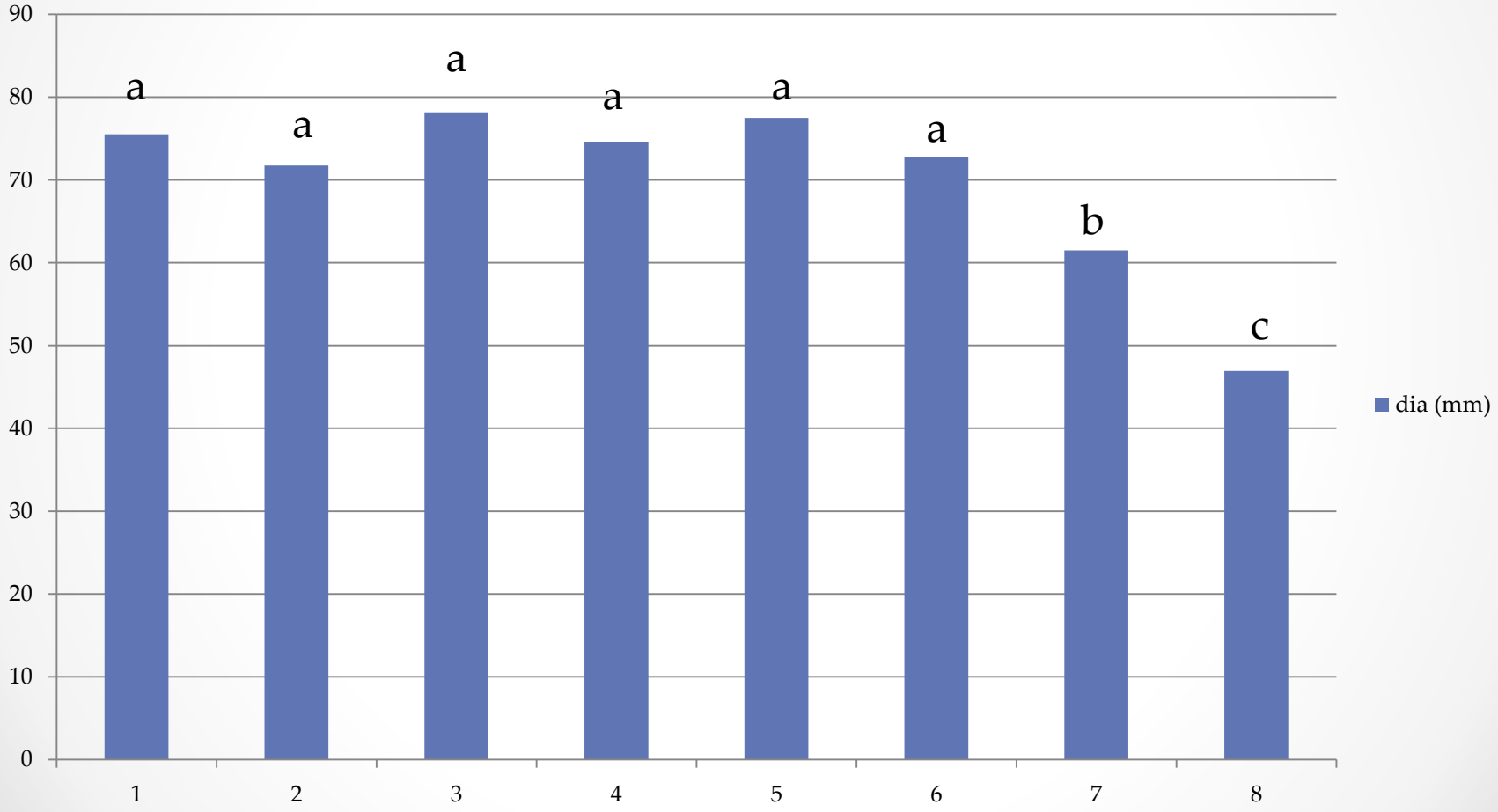
2.5 Acres

10.0 Acres

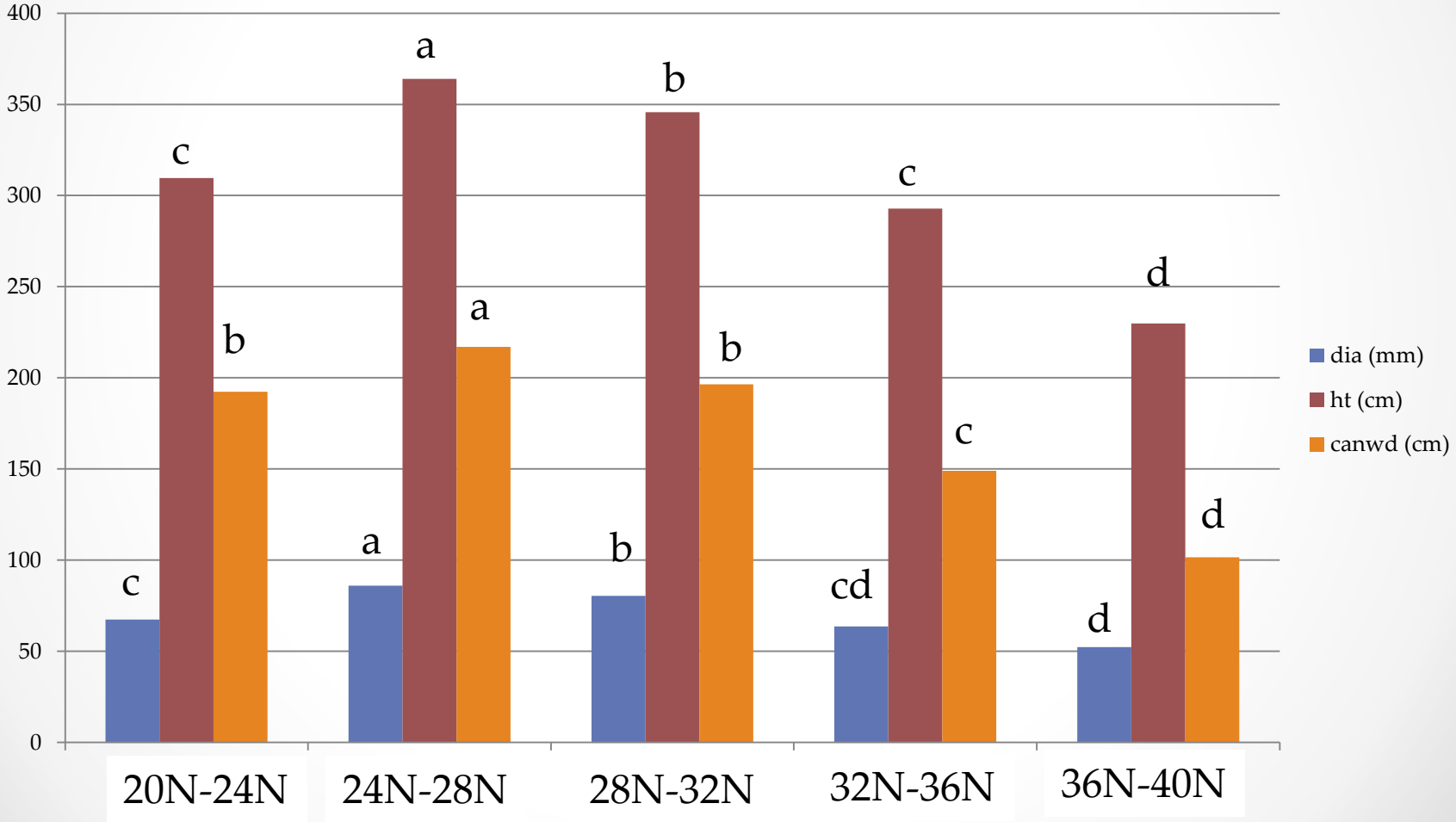



N40
N36
N32
N28
N24
N20
latcor 5
n=48
latcor 4
n=32
latcor 3
n=305
latcor 2
n=857
latcor 1
n=126

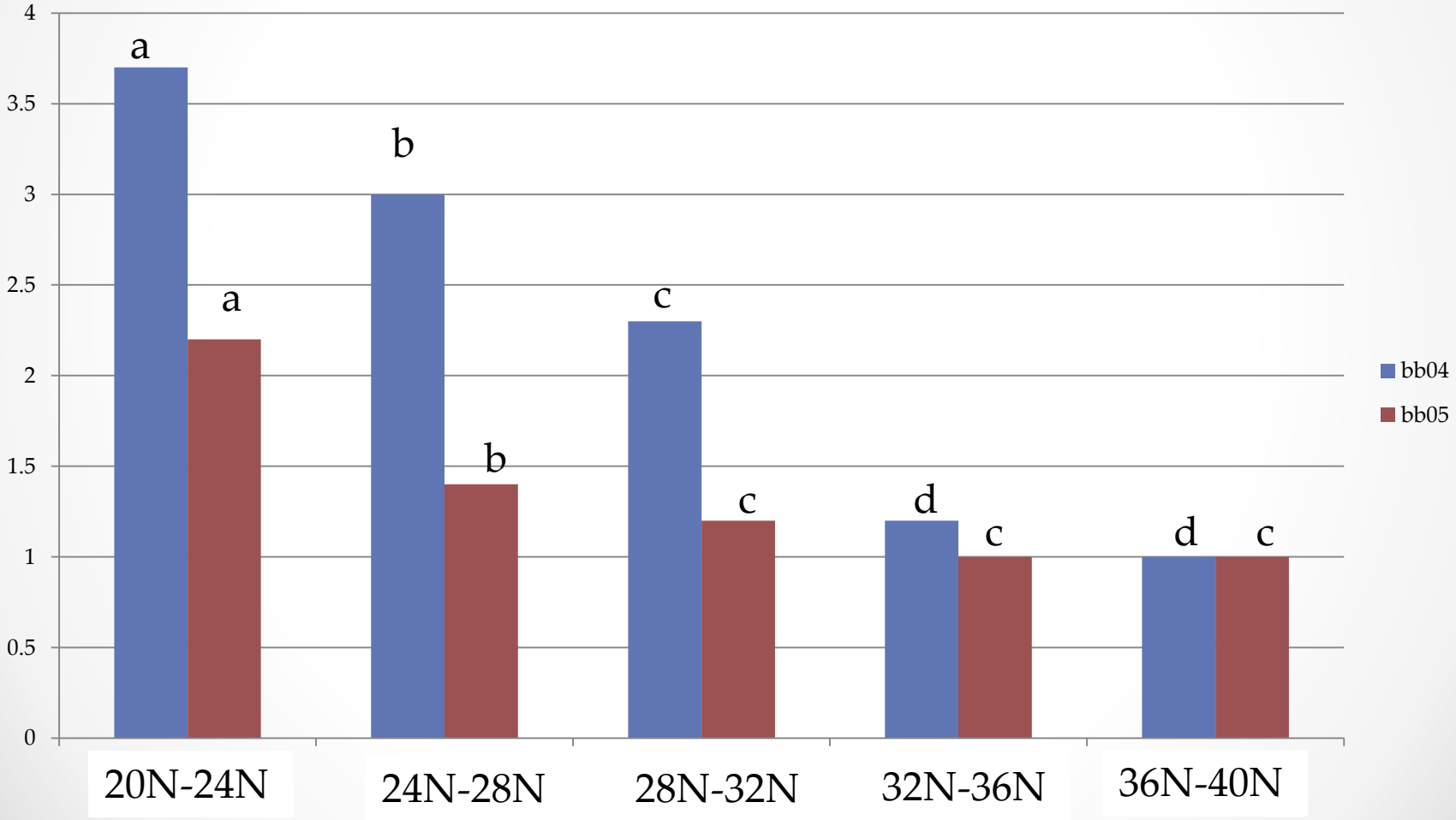
Trees growing in Ships clay (Block 8) are reduced in size compared to trees growing in the Weswood soil. Block 7 was intermediate.



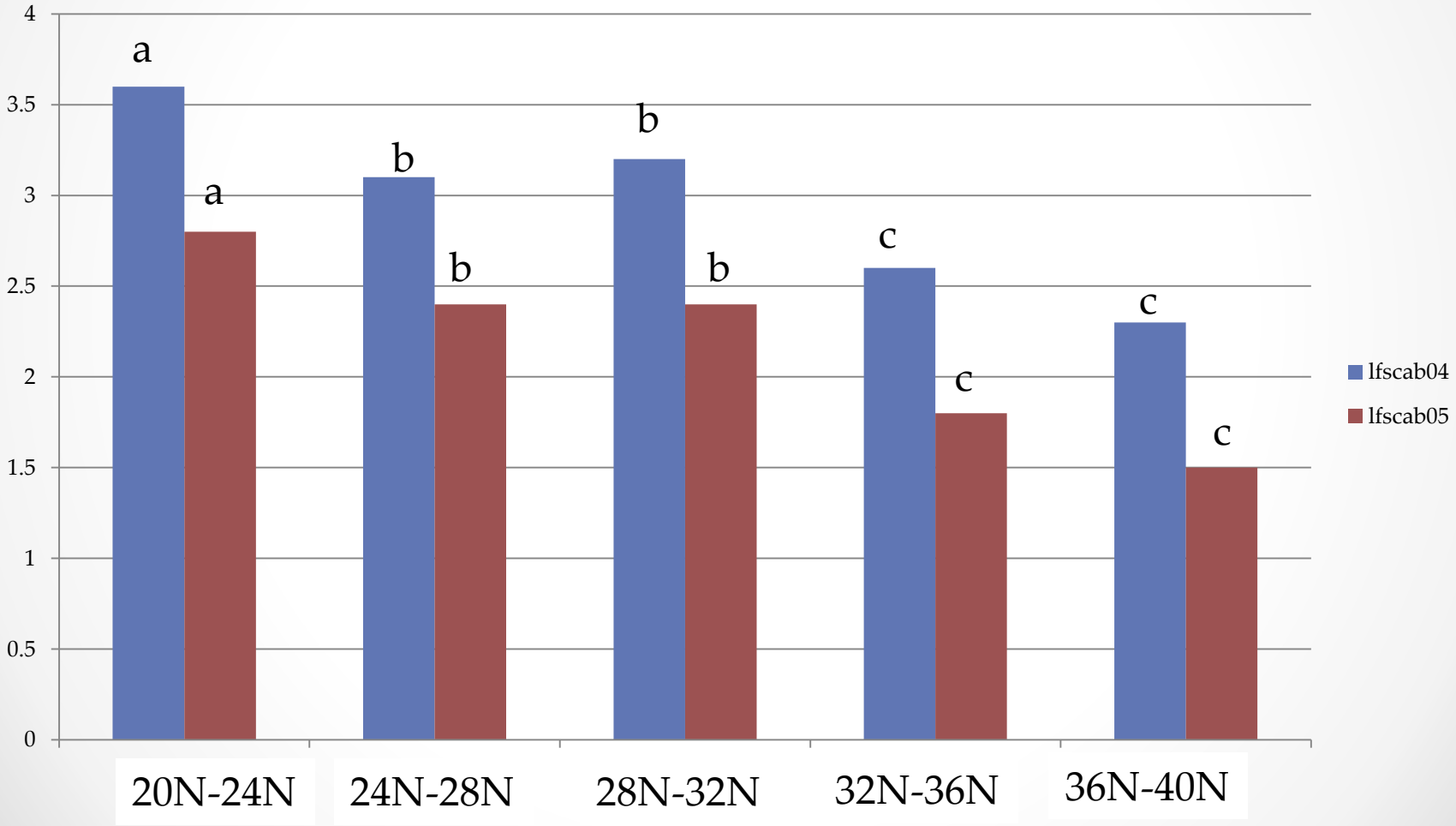
Largest seedlings grow from seed collected slightly south of the orchard site, and decrease in size with northern seed sources



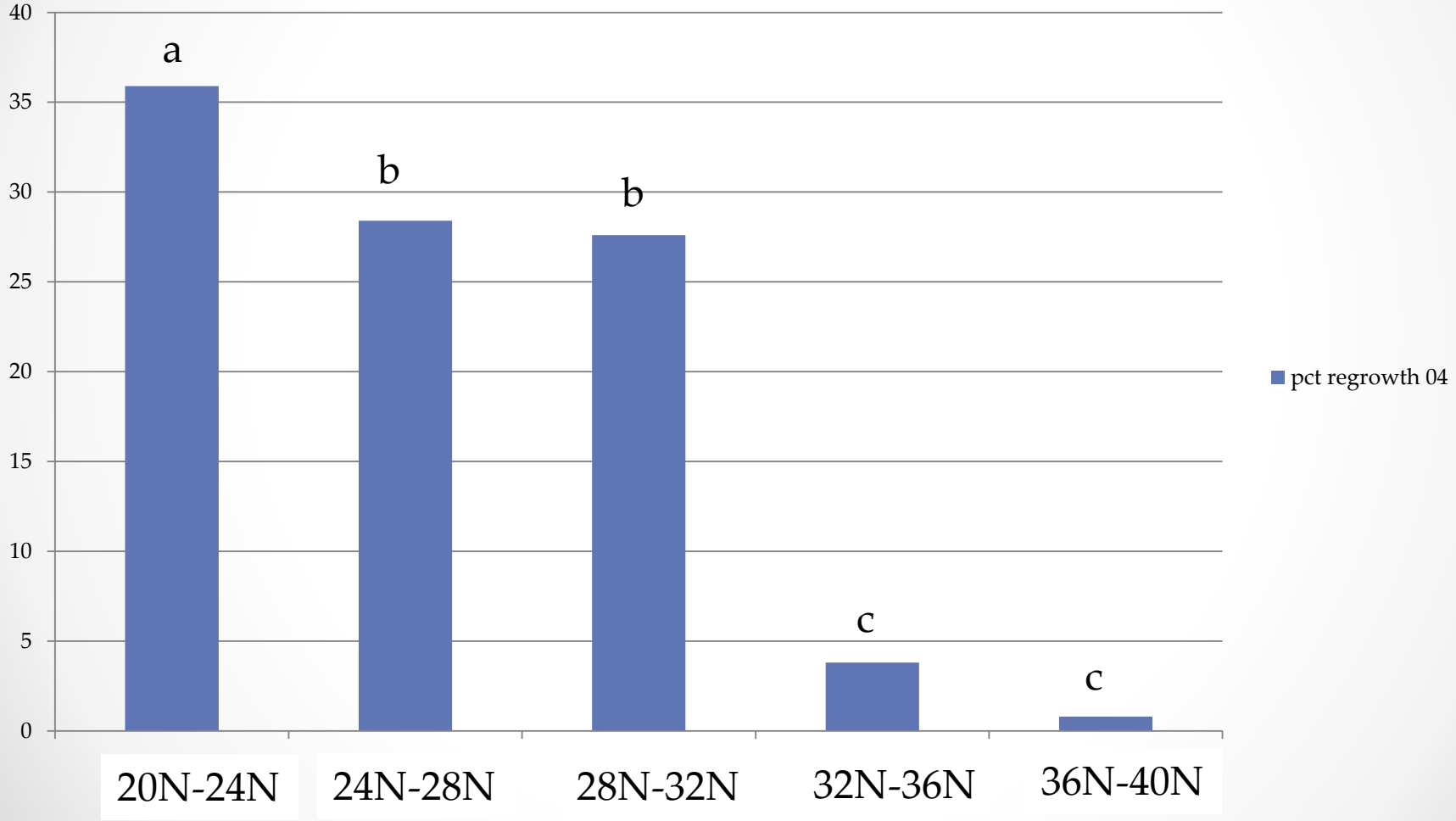
Initiation of growth occurs first in seedlings from southern sources.



Leaf scab decreased in seedlings from south to north



Late season regeneration growth (October flushes) is greatest in trees from southern sources



Pecan microsatellite markers were initially developed in this program for cultivar identification and verification

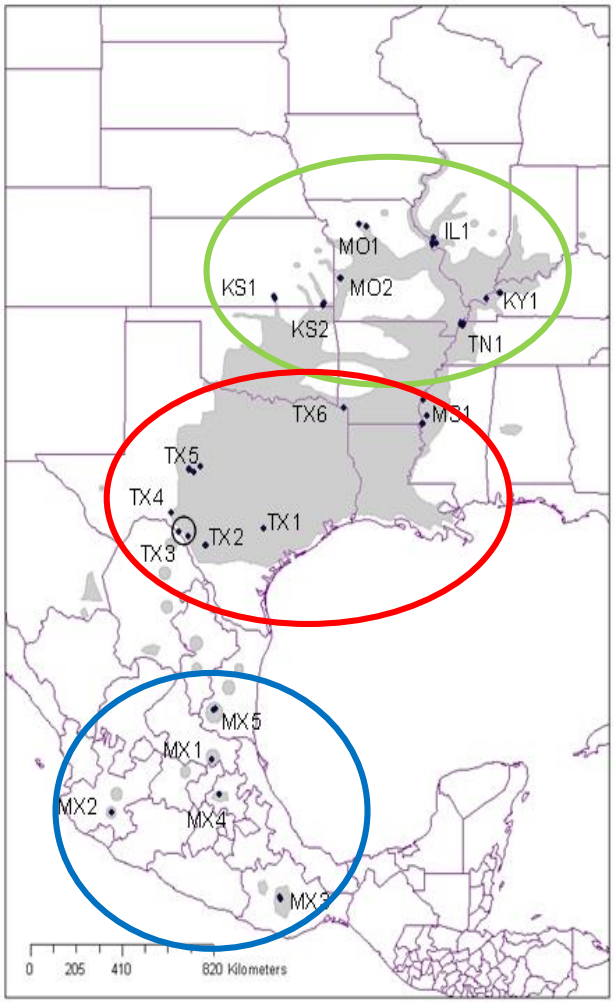
Repository:

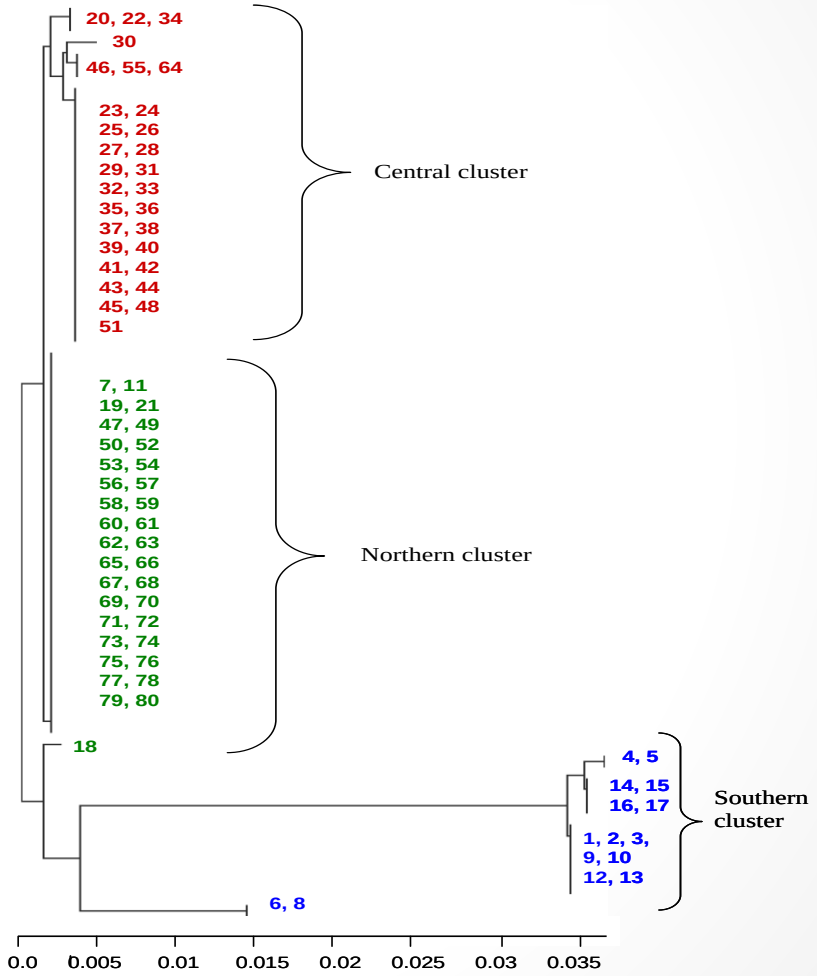
- Distinguish species
- Distinguish geographic populations
- Verify cultivar identity
- Verify parentage
- Accumulate profiles for comparison

Breeding program

- Associate phenotype with genotype
- Understand adaptations related to distribution
- Provide genetic markers for selection

Patterns of nuclear vs plastid microsatellite variation analyzed as bp alleles differed in native pecan populations collected across the range of the species. Greatest plastid diversity was in southern populations, consistent with age and isolation in glacial refugia. Greatest geographic information content is in plastids.

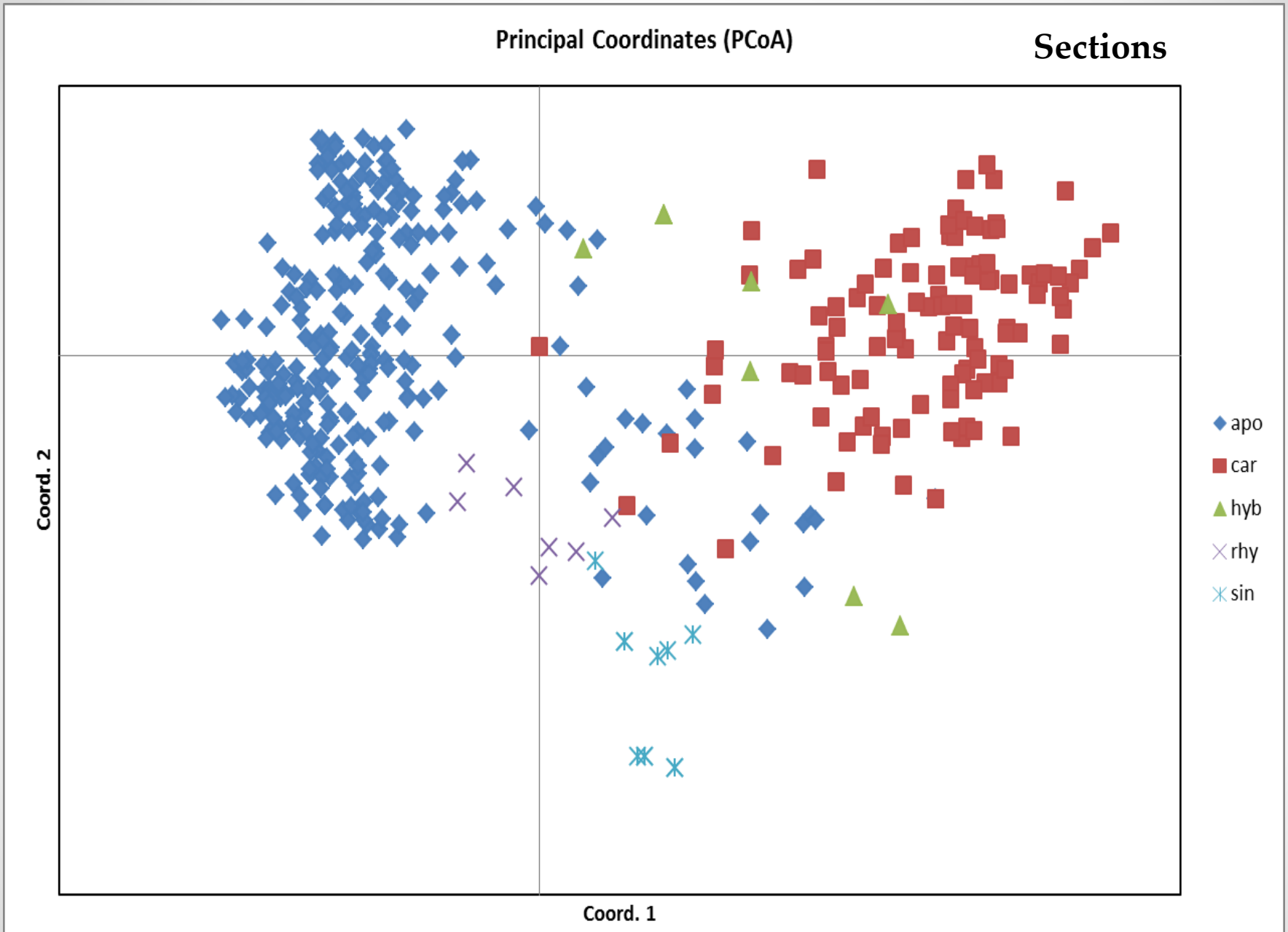


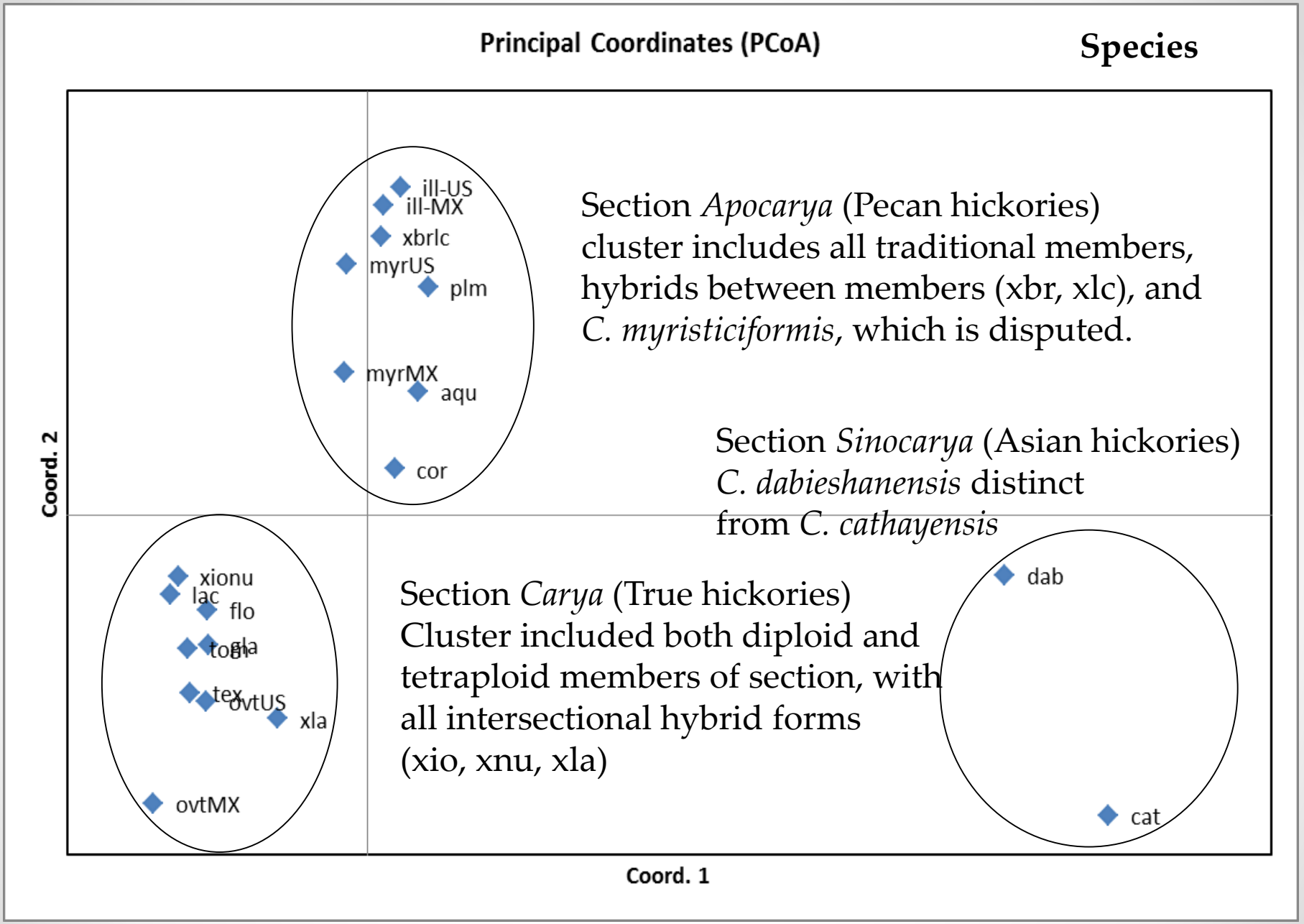


Grauke L J, Mendoza-Herrera A M, Miller A J, Wood B W. 2011. Geographic patterns of genetic variation in native pecans. Tree Genetics and Genomes 7:917-932.

Sections of Carya: analysis of molecular profiles of 3 plastid and 11 nuclear microsatellites in binary form

Total Band Patterns SECTIONS					
No. Loci	325				
No. Samples	418				
No. Pops.	5				
Population	**apo**	**car**	**hyb**	**rhy**	**sin**
n	282	110	8	7	11
No. Bands	246	255	84	62	48
No. Bands Freq. >= 5%	96	147	84	62	48
No. Private Bands	40	57	1	9	6
No. LComm Bands (<=25%)	0	0	0	0	0
No. LComm Bands (<=50%)	50	50	24	9	8
Mean He	0.058	0.085	0.058	0.046	0.039
SE of Mean He	0.006	0.006	0.007	0.006	0.006
Mean uHe	0.058	0.085	0.062	0.049	0.041
SE of Mean uHe	0.006	0.006	0.007	0.007	0.007


Principal Coordinates (PCoA)
Sections
Coord. 2
Coord. 1
apo
car
hyb
rhy
sin


Principal Coordinates (PCoA)
Species
Coord. 2
Coord. 1
ill-US
ill-MX
xbrlc
myrUS
plm
myrMX
aqu
cor
xionu
lac
flo
tom
gla
tex
ovtUS
xla
ovtMX
dab
cat
Section *Apocarya* (Pecan hickories) cluster includes all traditional members, hybrids between members (xbr, xlc), and *C. myristiciformis*, which is disputed.
Section *Sinocarya* (Asian hickories) *C. dabieshanensis* distinct from *C. cathayensis*
Section *Carya* (True hickories) Cluster included both diploid and tetraploid members of section, with all intersectional hybrid forms (xio, xnu, xla)

Sectional hybrids group with Section *Carya*

- *C. myristiciformis* is taxonomically classified in Section *Carya*, and has been recognized as a morphological intermediate between the Sections. With these few molecular markers, it groups with *Apocarya,* possibly because of hybridity with pecan, which is suspected. Its role in phylogenetic development of the genus might be explored with appropriate inclusion in core diversity panels. **The species is threatened across its disjunct range from the southeastern US into Mexico, and is included in efforts at *in situ* conservation.**
- Diploid and tetraploid species of Section *Carya* form a cluster that includes *Apocarya* hybrids. **Gene flow between the sections justifies inclusion of selected representatives in core diversity panels and increased attention to their horticultural value as rootstocks and for scion development.**

Molecular markers reveal 'Major'- a productive parent in the Breeding Program is interspecific hybrid. Genetic contributions of Crop Wild Relatives will be more accessible with development of better genomic tools

Kentucky native discovered 1907 on the Major farm, in a forest of mixed pecan and hickories.

It has good resistance to scab, passed to progeny: 'Osage', 'Kanza', and 'Lakota'. 'Osage' is the male parent of 'Mandan'. Its progeny are among the most scab resistant seedlings in the CSM provenance orchard.

SampNo	PlantID	LOCATION	ssp	A05a	A05b
LJ98-4	Mahan	MS	ill	160	165
LJ00-86	Pawnee	CC	ill	160	160
LJ01-115	Schley	MS	ill	160	160
LJ00-112	**Major**	KY	ill	115	150
LJ06-9	Kanza	CC	ill	115	160
LJ06-7	Osage	CC	ill	115	160
LJ06-105	Pleas (XBR)	OK	xbr	115	160
LJ06-62	Abbott Thinshell (XBR)	IL	xbr	115	160
LJ09-903	cordiformis (NY)	NY	cor	115	115
Stone 4407	cordiformis	NC	cor	115	115
LJ06-49	02-COR-LA-BF1	LA	cor	110	115
LJ06-50	02-COR-LA-BF2	LA	cor	115	120

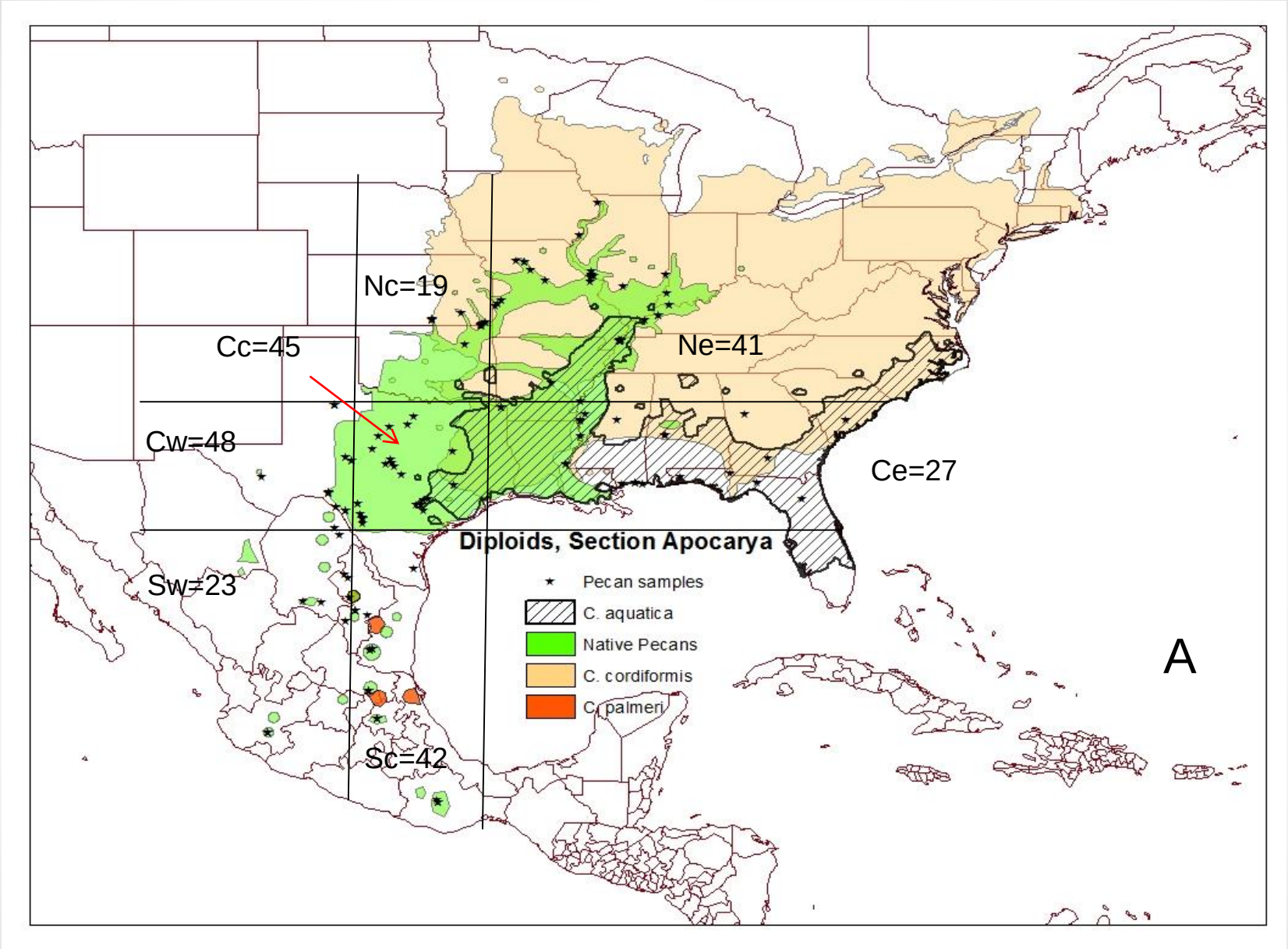
Grauke, LJ, Klein RR, Grusak MA, Klein P. 2015. Acta Horticulturae 1070:109-126

The 1-A05 locus is a pentanucleotide repeat $(GTTTT)_5$, developed from EST00005 in cooperative work with the Grusak and Klein labs.

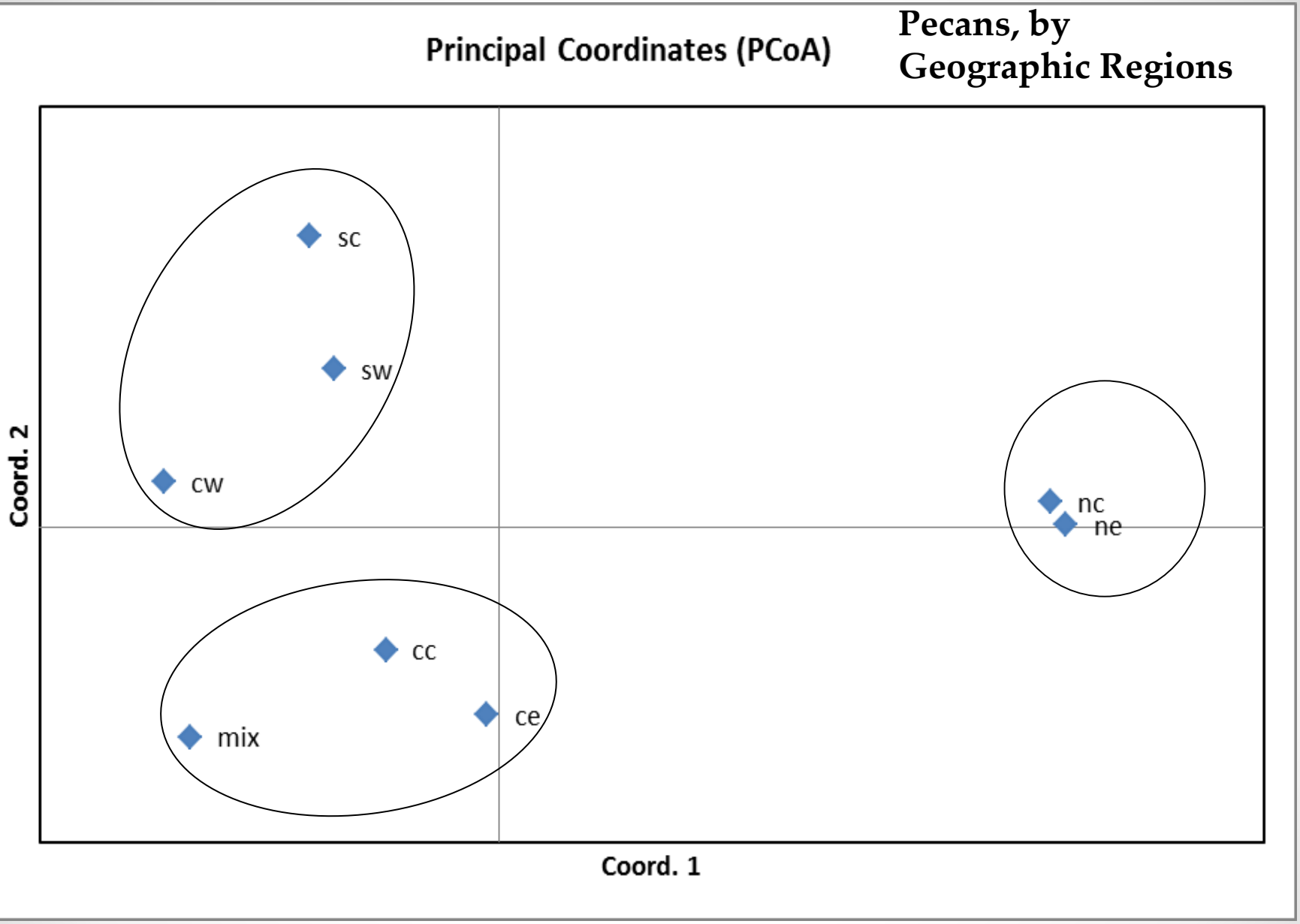
Mapping populations are being Developed between 'Lakota', whose admixture from CWR may aid resistance, and 87MX3-2.11, the homozygous reference genome.

Native pecans by geographic region: analysis of molecular profiles of 3 plastid and 11 nuclear microsatellites in binary form

Total Band Patterns PECANS by GEOGRAPHIC REGIONS								
No. Loci	325							
No. Samples	252							
No. Pops.	8							
Population	**cc**	**ce**	**cw**	**mix**	**nc**	**ne**	**sc**	**sw**
n	45	27	48	7	19	41	42	23
No. Bands	130	97	110	69	100	122	113	100
No. Bands Freq. >= 5%	79	74	80	69	100	73	76	60
No. Private Bands	6	2	4	0	1	9	9	10
No. LComm Bands (<=25%)	12	9	10	6	8	12	9	7
No. LComm Bands (<=50%)	39	25	34	20	34	37	30	23
Mean He	0.050	0.050	0.051	0.045	0.045	0.049	0.046	0.049
SE of Mean He	0.006	0.006	0.006	0.006	0.005	0.006	0.005	0.006
Mean uHe	0.051	0.051	0.052	0.049	0.046	0.050	0.047	0.050
SE of Mean uHe	0.006	0.006	0.006	0.007	0.006	0.006	0.005	0.006


Nc=19
Cc=45
Ne=41
Cw=48
Ce=27
Diploids, Section Apocarya
Pecan samples
C. aquatica
Native Pecans
C. cordiformis
C. palmeri
Sw=23
Sc=42
A

Pecans, by Geographic Regions



Pairwise Population Matrix of Mean Population Binary Genetic Distance
Pecan populations labeled as sampled within state of origin
Highest affinity between populations shown in green, greatest distance in orange

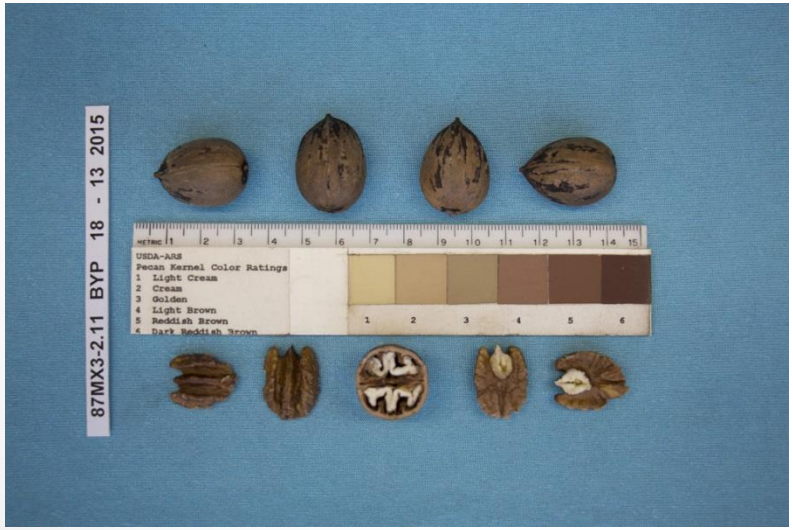
ALM	DC	IL	KS	KY	mix	MO	MX1	MX2	MX3	MX4	MX5	NLC	se	TN	TX1	TX2	TX3	TX4	TX5	TX6	TXSD		n
24.27	29.10	24.15	23.95	23.65	27.23	23.18	25.14	22.84	25.31	29.00	24.33	26.25	25.26	23.45	26.19	25.80	27.28	26.42	27.77	23.87	26.68	**ALM**	10
29.10	15.49	28.81	28.18	28.70	27.91	27.60	26.38	23.83	23.77	27.44	28.17	28.21	27.36	29.69	26.97	27.87	28.55	27.04	28.65	27.72	27.33	**DC**	25
24.15	28.81	23.77	23.05	23.03	27.89	22.41	25.25	23.82	26.51	30.01	24.01	26.60	25.53	22.40	26.29	26.61	27.71	27.38	28.17	23.61	27.11	**IL**	13
23.95	28.18	23.05	22.77	22.03	27.20	21.72	25.62	23.52	26.03	29.54	23.53	26.41	25.47	21.99	25.60	25.78	26.83	27.03	27.37	23.34	26.94	**KS**	13
23.65	28.70	23.03	22.03	20.04	25.84	21.61	25.52	22.10	26.88	29.90	23.48	26.11	23.97	21.88	24.64	25.59	26.08	26.33	26.72	23.01	25.88	**KY**	8
27.23	27.91	27.89	27.20	25.84	23.52	26.77	27.47	24.56	26.16	29.26	26.40	26.19	21.98	28.21	24.38	24.05	25.40	25.11	25.87	27.02	22.69	**mix**	7
23.18	27.60	22.41	21.72	21.61	26.77	20.00	25.10	21.82	25.87	28.91	22.97	25.62	24.95	21.44	25.59	26.18	26.72	26.43	27.43	22.75	26.03	**MO**	13
25.14	26.38	25.25	25.62	25.52	27.47	25.10	15.33	17.48	19.59	22.55	24.93	26.87	25.81	25.57	26.73	25.84	26.66	25.43	28.62	24.16	27.49	**MX1**	7
22.84	23.83	23.82	23.52	22.10	24.56	21.82	17.48	11.44	19.13	22.19	21.44	24.35	23.54	23.46	24.81	24.26	24.53	24.47	26.50	22.98	25.93	**MX2**	9
25.31	23.77	26.51	26.03	26.88	26.16	25.87	19.59	19.13	8.29	17.64	24.58	25.35	25.54	26.81	25.57	24.95	26.20	25.00	28.04	25.52	26.91	**MX3**	7
29.00	27.44	30.01	29.54	29.90	29.26	28.91	22.55	22.19	17.64	16.06	28.24	28.00	28.53	29.81	28.51	28.33	29.23	28.10	30.45	28.06	29.73	**MX4**	12
24.33	28.17	24.01	23.53	23.48	26.40	22.97	24.93	21.44	24.58	28.24	20.80	24.11	24.67	24.11	24.73	24.73	25.50	25.19	27.04	23.29	26.04	**MX5**	14
26.25	28.21	26.60	26.41	26.11	26.19	25.62	26.87	24.35	25.35	28.00	24.11	24.04	23.97	26.72	24.63	23.76	25.29	24.40	26.56	25.35	24.74	**NLC**	17
25.26	27.36	25.53	25.47	23.97	21.98	24.95	25.81	23.54	25.54	28.53	24.67	23.97	19.44	25.85	22.16	21.04	23.42	21.90	23.89	24.11	21.16	**se**	18
23.45	29.69	22.40	21.99	21.88	28.21	21.44	25.57	23.46	26.81	29.81	24.11	26.72	25.85	20.91	26.82	26.42	27.07	27.88	28.05	23.11	27.30	**TN**	12
26.19	26.97	26.29	25.60	24.64	24.38	25.59	26.73	24.81	25.57	28.51	24.73	24.63	22.16	26.82	22.12	22.17	24.85	24.06	24.15	24.88	23.08	**TX1**	16
25.80	27.87	26.61	25.78	25.59	24.05	26.18	25.84	24.26	24.95	28.33	24.73	23.76	21.04	26.42	22.17	20.75	22.13	20.93	23.82	24.07	22.48	**TX2**	8
27.28	28.55	27.71	26.83	26.08	25.40	26.72	26.66	24.53	26.20	29.23	25.50	25.29	23.42	27.07	24.85	22.13	23.00	22.20	25.49	26.42	23.40	**TX3**	5
26.42	27.04	27.38	27.03	26.33	25.11	26.43	25.43	24.47	25.00	28.10	25.19	24.40	21.90	27.88	24.06	20.93	22.20	19.33	24.46	25.20	22.56	**TX4**	10
27.77	28.65	28.17	27.37	26.72	25.87	27.43	28.62	26.50	28.04	30.45	27.04	26.56	23.89	28.05	24.15	23.82	25.49	24.46	26.10	26.94	23.88	**TX5**	13
23.87	27.72	23.61	23.34	23.01	27.02	22.75	24.16	22.98	25.52	28.06	23.29	25.35	24.11	23.11	24.88	24.07	26.42	25.20	26.94	22.06	25.42	**TX6**	9
26.68	27.33	27.11	26.94	25.88	22.69	26.03	27.49	25.93	26.91	29.73	26.04	24.74	21.16	27.30	23.08	22.48	23.40	22.56	23.88	25.42	21.40	**TXSD**	5

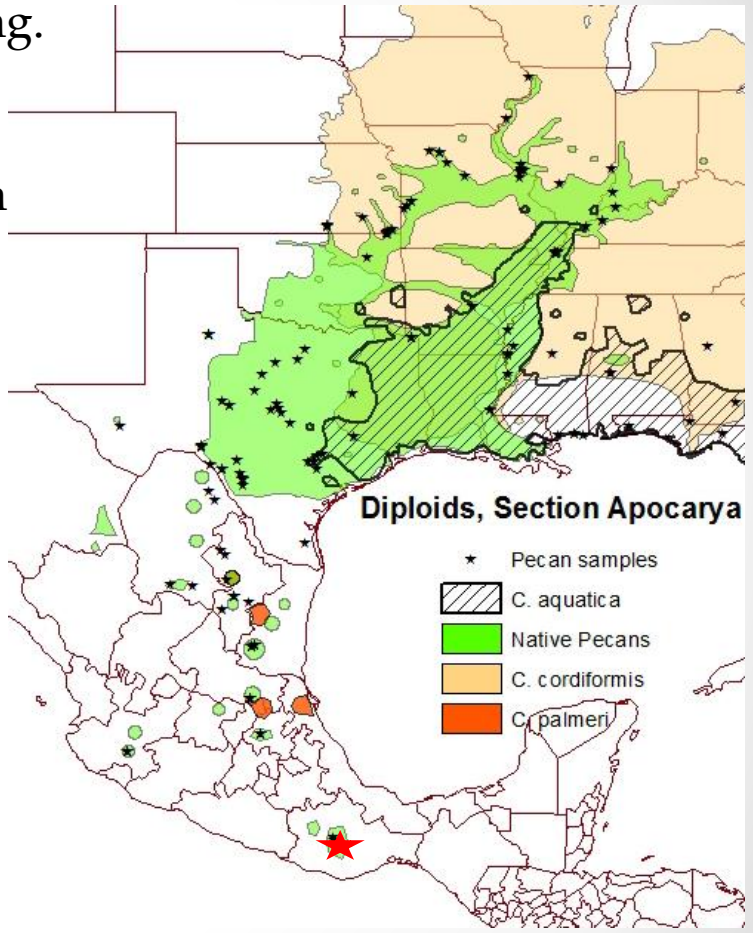
87MX3-2.11 : Reference Genome

Found to be homozygous at 11 nuclear microsatellite loci, possibly due to topographic isolation and inbreeding.

Evaluated by HudsonAlpha Genome Sequencing Center: “Contains lower variation than the common genotypes in the pecan breeding program and is an excellent choice from which to produce a reference genome.”

Jenkins, J. et al., 2015. Acta Hortic. 1070:101-108.





87MX4

Self-rooted seedlings grown from seed collected in Ixmiquilpan, Hidalgo, Mexico, in 1987.

Break buds early in the spring, but are the very last to cease growth in the fall, often continuing active growth into December.

As a group, they manifest indeterminate growth resulting in weeping habit.

With limited markers, this population had the greatest genetic distance to US populations. Does that predict heterosis?

In Cotton Root Rot trials with Mark Black (TAMU) in Uvalde Texas, open-pollinated seedlings from this population are by far the most vigorous.

In Salinity trials with Seichi Miyamoto's (TAMU) in El Paso Texas, seedstocks from this population produced vigorous seedlings considered "tolerant to both Na^+ and Cl^- ions".

Trees grown from seed collected in Ixmiquilpan, Mexico

Trees grown from seed collected in Jaumave, Mexico: how do we characterize tree architecture?

Jones Hybrid Hickory

BWV 5-31, 17 Apr 2012.

The molecular profile indicates hybridity between *C. cordiformis* and *C. ovata*

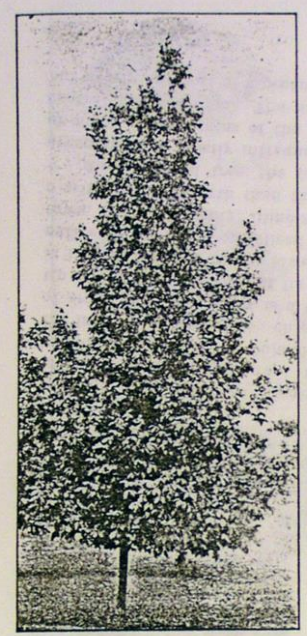


Figure 25—A Siers hybrid tree (bitternut x mockernut).

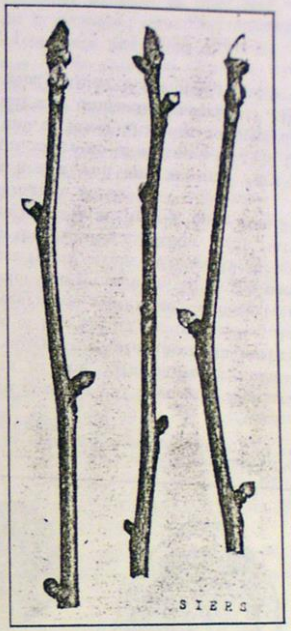


Figure 27—Dormant buds of the Siers hybrid hickory. Photograph by Miss L. A. Guernsey. 1945. (Natural size).

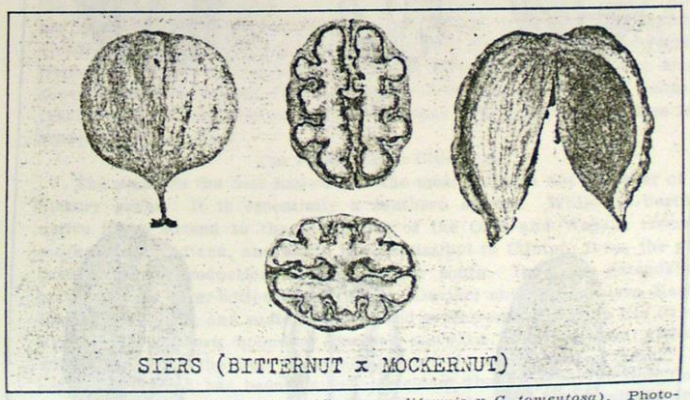
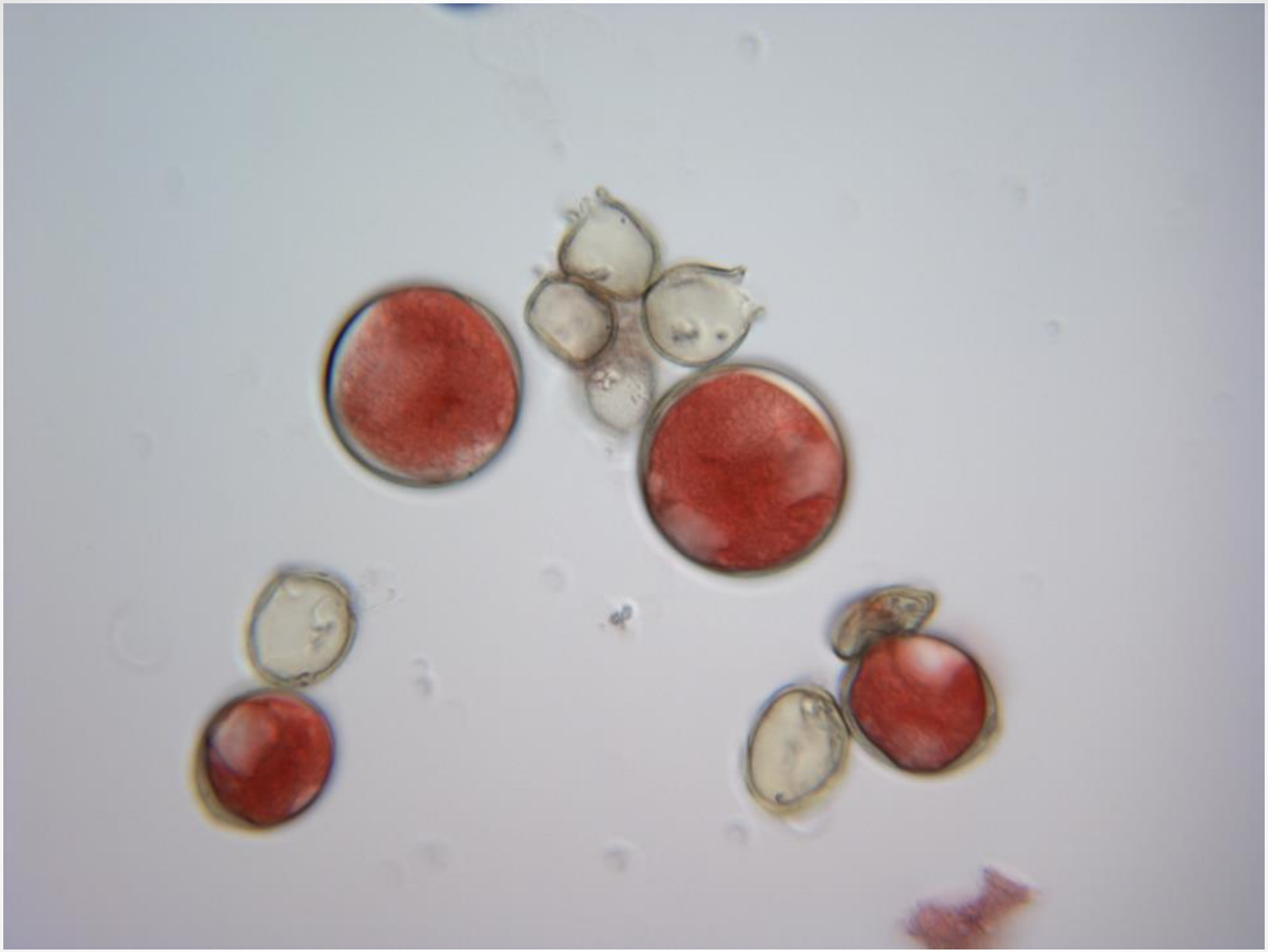


Figure 26—Siers hybrid hickorynuts (*Carya cordiformis* x *C. tomentosa*). Photograph by Miss L. A. Guernsey. 1945. (Natural size).

[109]

I believe our "Jones Hybrid Hickory" is actually the 'Siers' hybrid, described by C. A. Reed as a cross between bitternut and mockernut (C. tomentosa). That cultivar was propagated by J. F. Jones Nursery

Figures 25, 26 and 27 of 'Siers 'tree form, nuts with shucks, and buds (respectively) from C. A. Reed, 1944.



Jones Hybrid pollen, 25x. Showing giant pollen grains that are possibly unreduced gametes (and might fertilize a tetraploid).
Photo courtesy of Dr. David Stelly, TAMU

A strategy for coordinated development of molecular tools for pecan.

- Develop a reference sequence
  - 87MX3-2.11 is being sequenced by HudsonAlpha as a reference genome.
- Standardize phenotypic descriptors for traits of group interest
  - Refine observations on floral, leaf and tree structure
- Establish a segregating population in multiple locations for shared use in developing those descriptors, providing information on genotype x environmental interaction
  - 'Lakota' ('Mahan' x 'Major') x 87MX3-2.11 cross will be made in 2016.
  - Admixture from the hybridity of 'Major' may help resolve sources of disease resistance in 'Lakota'
  - Extreme variation in north-south phenology
  - Extreme variation in tree architecture

Windows of opportunity for the next generation

- *Xylella fastidiosa*, a xylem dwelling pathogen that causes Pecan Bacterial Leaf Scorch was identified in the recent Crop Vulnerability Report as an emerging issue on pecan. Working with a national team, we revised detection methods, screened broadly, and found PBLS in NCGR-Carya, as well as commercial orchards in TX, NM, AZ and CA.
- We need to characterize the microbiome as part of genomic characterization of geographic distribution.
- In developing, deploying and distributing genetic resources, we should observe the Hippocratic Oath and "First, do no harm."
- Improved "core" collections will provide access to the exploration of available diversity, both domestically and internationally, with much of the exchange as DNA.
- As individual programs are reduced in size, cooperation across programs becomes more important.
- Existing NCGR-*Carya* collections are an excellent foundation for the next generation of scientists, and the next generation of trees.

Cooperators

USDA ARS

Plains Area, Southern Plains Area Research Center, Crop Germplasm Research Unit, College Station, Texas

Tommy E. Thompson (retired), Xinwang Wang, Robert Klein, Lynn Johnson, Keith Kubenka, Toni Cox, Scott Chaloupka

USDA ARS Southeast Area, Southeastern Fruit & Tree Nut Research, Byron Georgia

Bruce Wood (retired), Jerry A. Payne (retired), Clive Bock, Jim Stuckey

USDA ARS, National Arboretum

Alan Whittemore, Kevin Conrad

Michael Grusak

USDA ARS, Baylor College of Medicine

Michael Grusak

Texas A&M University, College Station, Texas

Marvin Harris, Patricia Klein, Maria Azucena Mendoza-Herrera, Mark Black, James Starr, Siechii Miyamoto, David Stelly, Carol Loopstra, Marla Binzel, Nolan Bentley, Young-Ki Jo, Angelyn Hilton,

University of Georgia

Patrick Conner,

Louisiana State University

Randy Sanderlin

Kansas State University

William Reid

HudsonAlpha Institute for Biotechnology, Huntsville Alabama (HA)

Jeremy Schmutz, Jerry Jenkins, Jane Grimwood

New Mexico State University

Jennifer Randall, Rio Stamler

Saint Louis University, Saint Louis, MO

Allison Miller