

Are seed collection zones needed for sourcing plant materials for longleaf pine ecosystem restoration?

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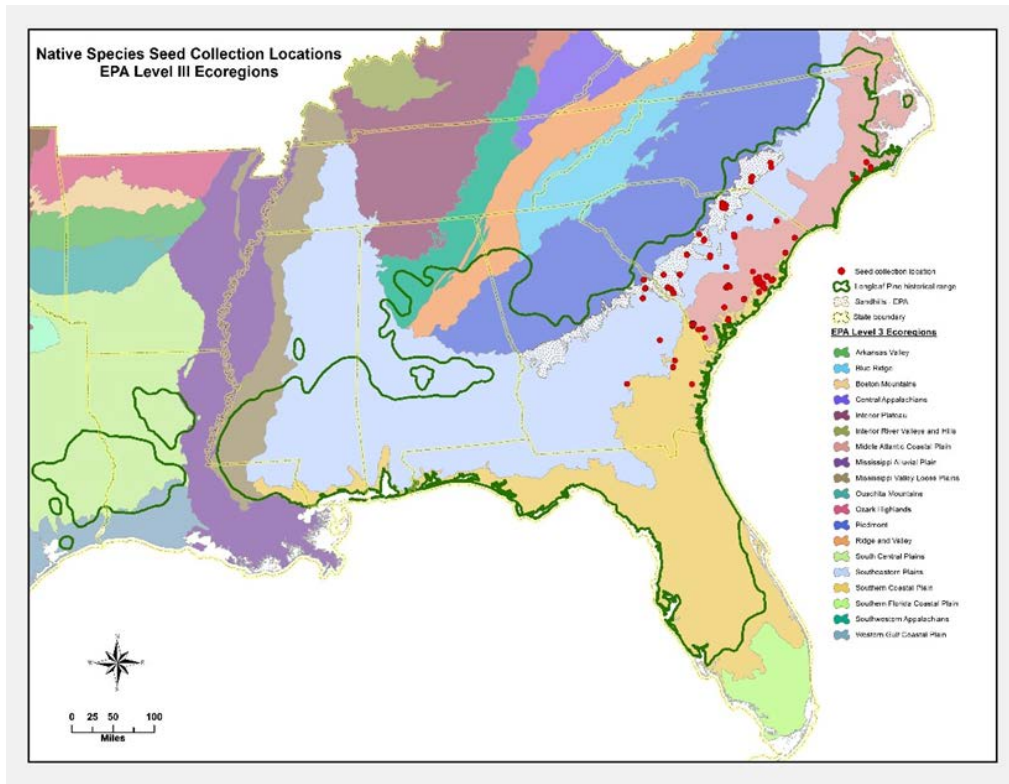
Presentation Objectives

- The LLP ecosystem and Need for seed, seed zones, strategies for addressing need
- Common garden studies in the LLP range – coordinated efforts
 - Gulf Coast study (Giencke et al 2018)
- Summarize results from Atlantic Coastal Plain study
- The need for seed collection zones?
- Genetic variation



Longleaf Pine Ecosystem Essentials

- Broad geographic extent
- Local variation is subtle, but controls diversity patterns (locally: soil texture, moisture availability, nutrient status)



Open pine canopy
Diverse ground layer
Maintained by frequent,
surface fire

Need for ecologically suitable seed for restoring longleaf pine communities



- Plant materials that will thrive in a restoration project
- Plant materials that do not threaten indigenous populations, conserve genetic resources
- Functional traits of interest are those that affect capacity to establish from seed in restoration projects

The safe solution

- Select seed sources from the closest possible location and collect from a site that physically matches the target site. ('Local is Best')
- May minimize risks of getting individuals maladapted to the restoration site, but provides no guidance for seed sources for wider distribution.

Recipe for success...

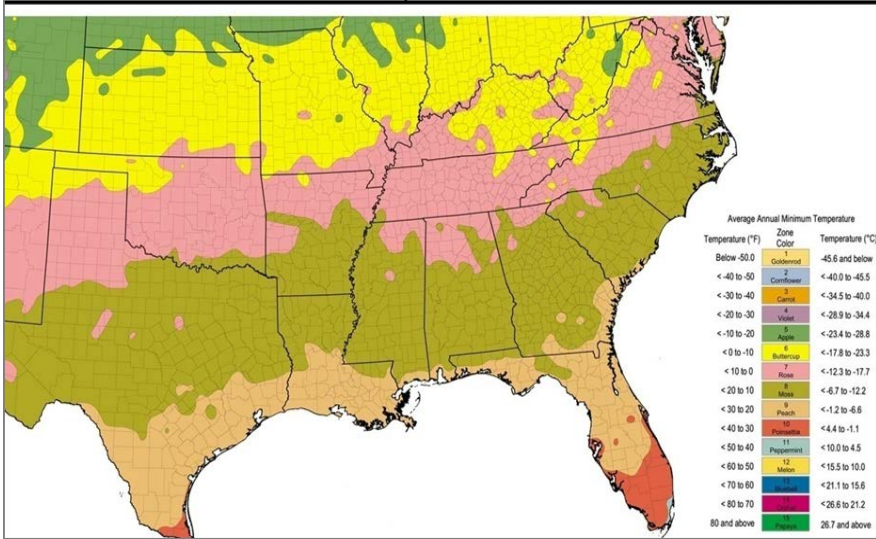
Dry
upland
seed



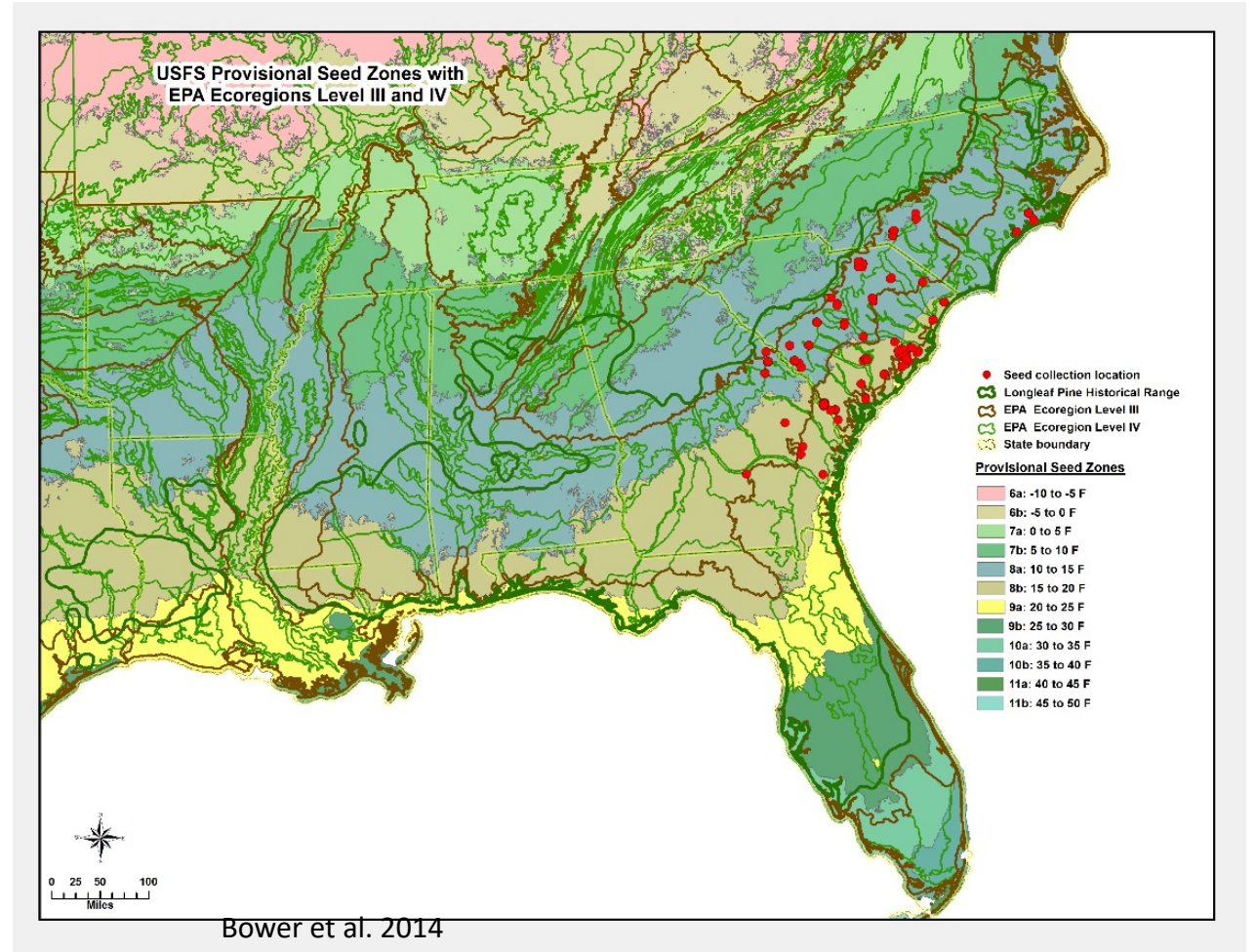
Nearby
dry
upland
site



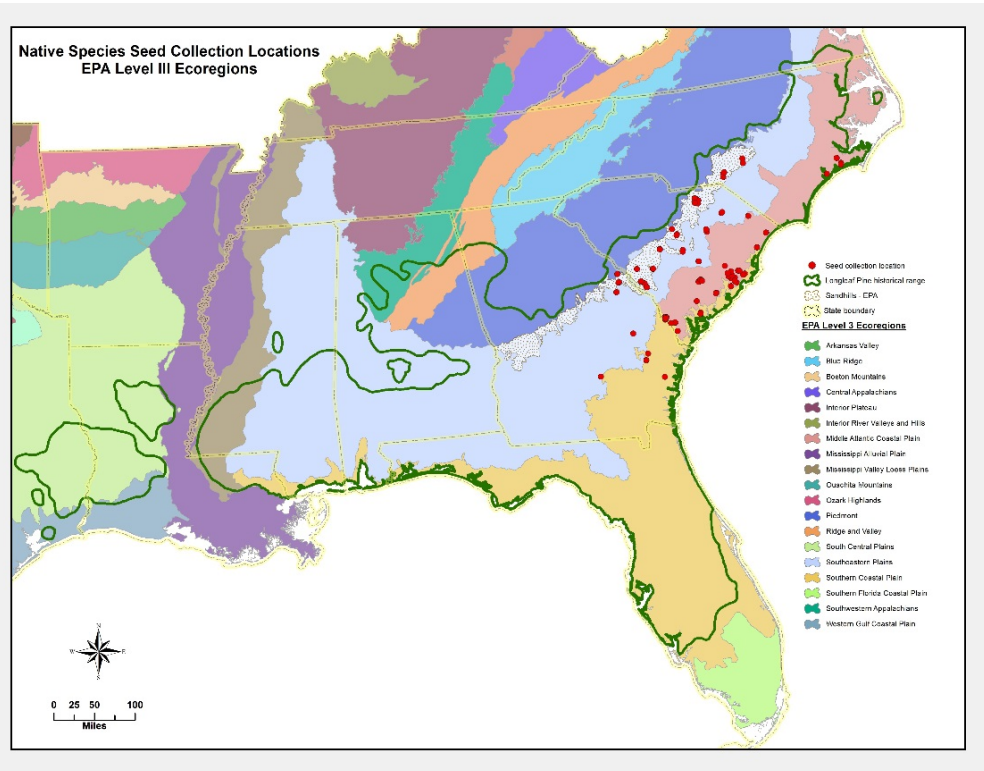
USDA Plant Hardiness Zone Map for the Southeastern United States



General Seed Zones

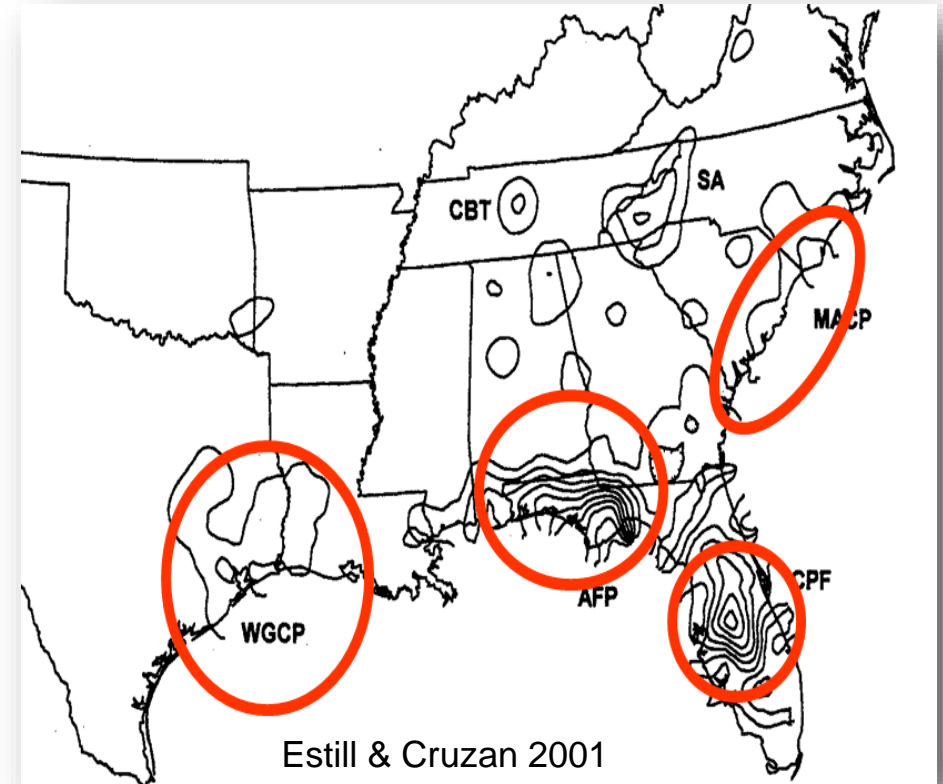


Applicability in SEUS to be determined with garden studies



Biogeographic considerations

- High levels of endemism throughout the ecosystem: opportunities, occurrences of speciation
 - Do patterns of endemism provide useful information related to intraspecific variation (fitness related) of widespread species, or genetic diversification related to geographic isolation?
- Species turnover across the range (legumes, mints, grasses)



Patterns in Endemism may be telling us something important...

An ecoregion model for biodiversity conservation planning... The Nature Conservancy

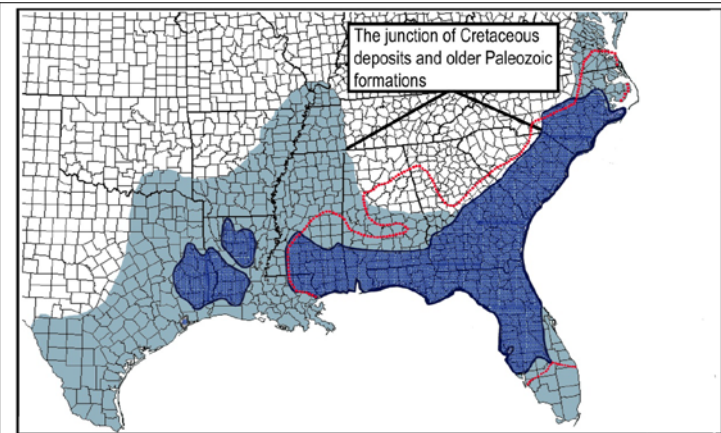


Figure 8. Phytogeographic pattern #18. Dark blue stippled area: core region of longleaf pine (*Pinus palustris*). Red dotted line: area of discontinuous or disjunct occurrences of longleaf pine. After Ware, Frost, and Doerr (1993). Light blue: Atlantic Coastal Plain, Gulf Coastal Plain, including the entire Mississippi Embayment. Adapted from Sorrie & Weakley (2001).

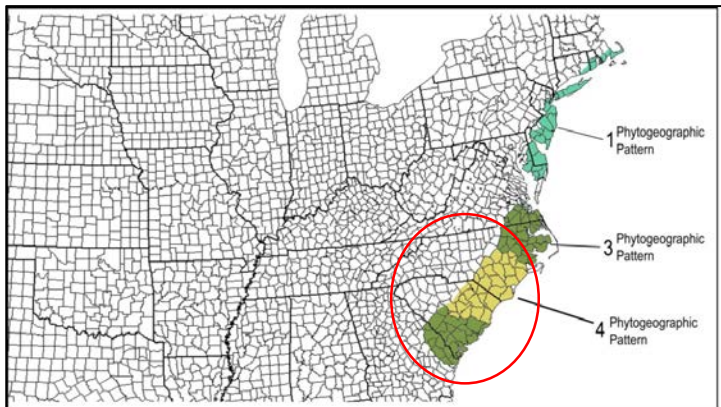


Figure 2. Phytogeographic patterns #1, #3, and #4. #1 (light blue): southeastern Massachusetts to southern New Jersey and adjacent Delmarva Peninsula. #3 (green): southeastern Virginia to southeastern Georgia. #4 (tan): southeastern North Carolina to northeastern South Carolina. Adapted from Sorrie & Weakley (2001).

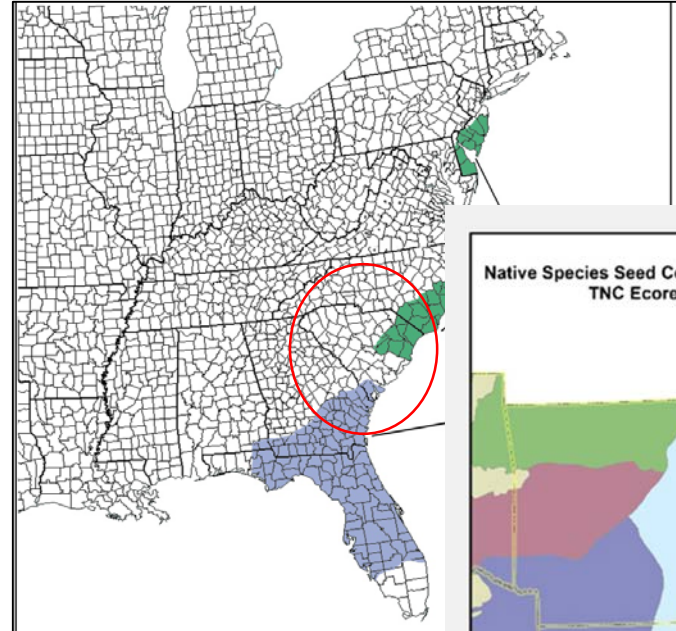
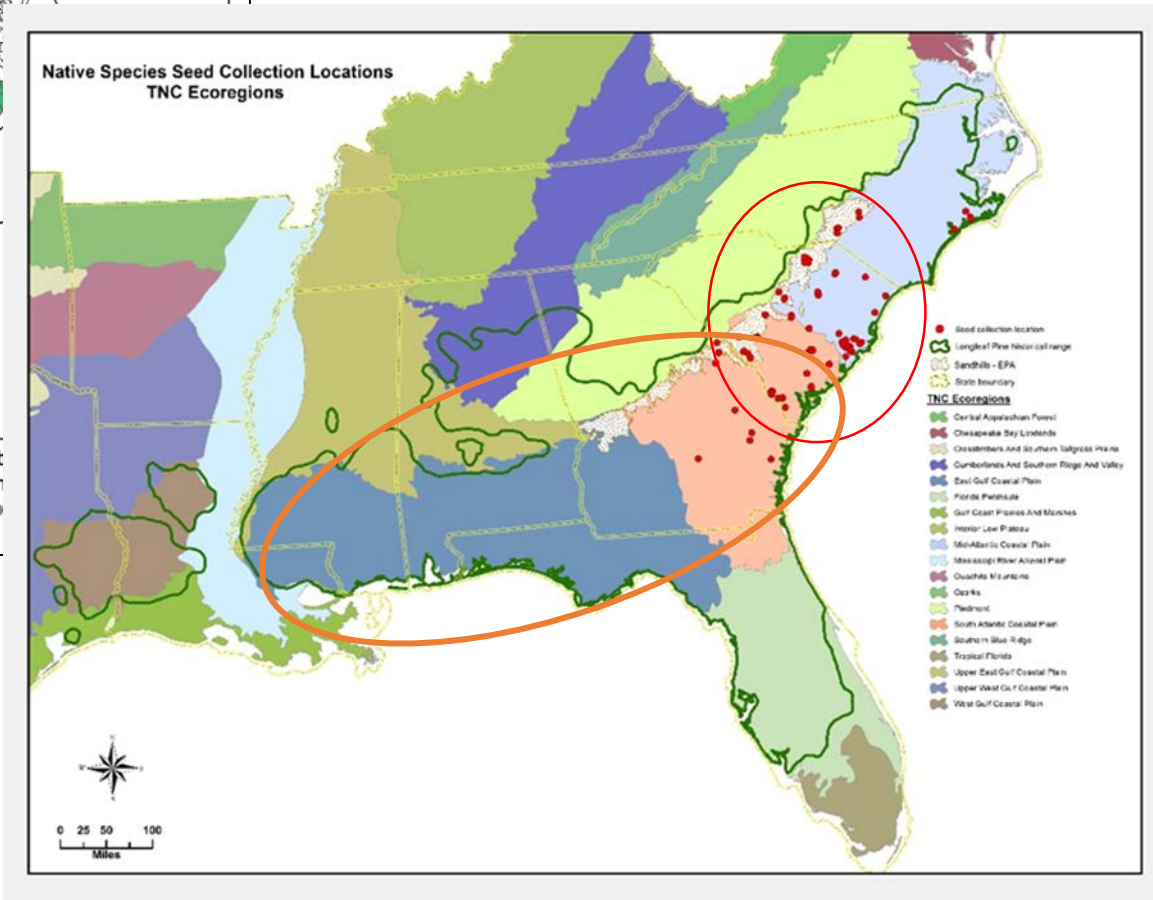


Figure 3. Phytogeographic pattern #2 and #6. #2 (green): Delaware/eastern North and South Carolina. #6 (blue): south Florida westward to the central panhandle of Florida and south Sorrie & Weakley (2001).



More information, more of the LLP range

RESEARCH ARTICLE

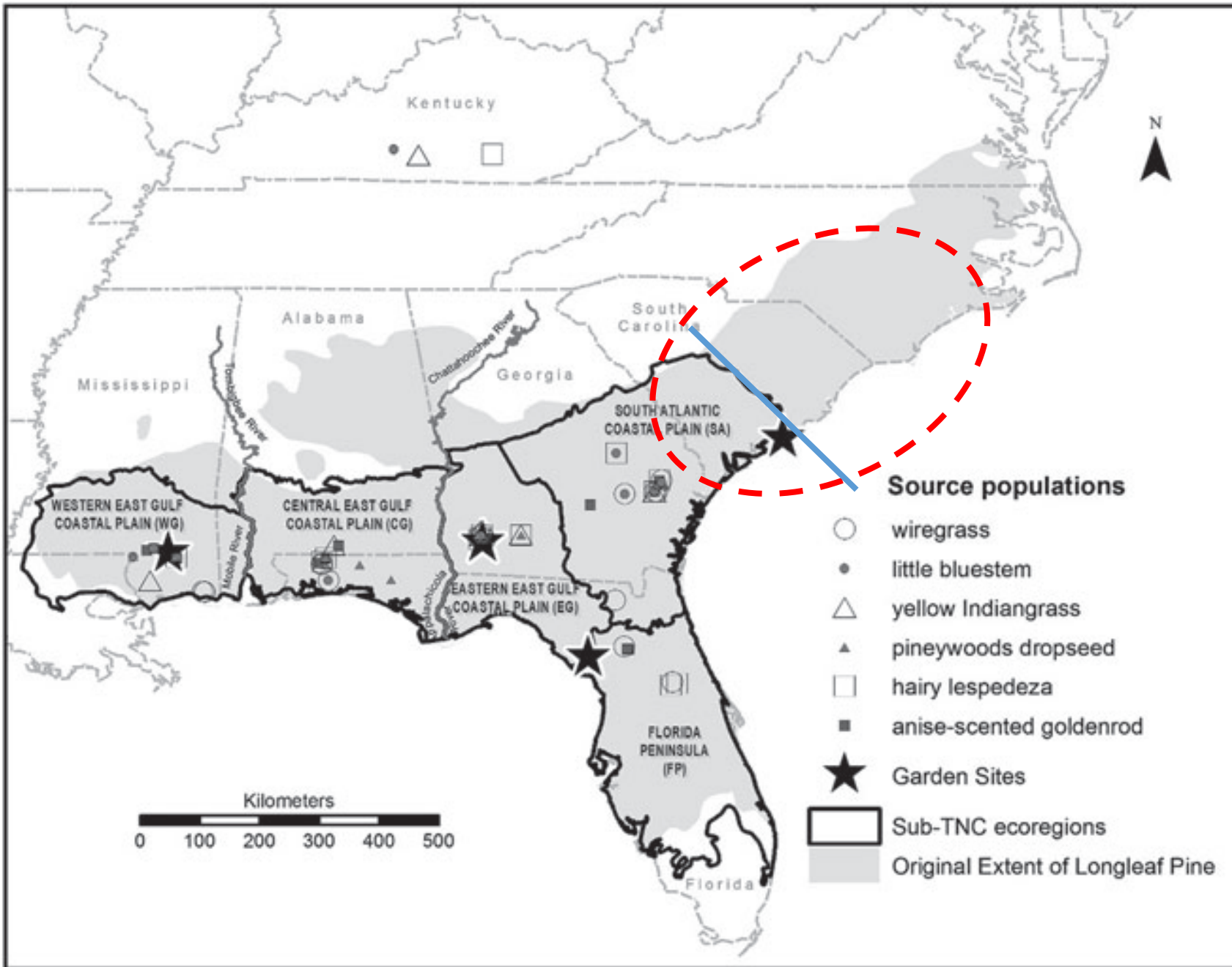
Seed sourcing for longleaf pine ground cover restoration: using plant performance to assess seed transfer zones and home-site advantage

Lisa M. Giencke^{1,2}, R. Carol Denhof³, L. Katherine Kirkman¹, O. Stribling Stuber¹, Steven T. Brantley¹

Seed sourcing is a concern for restoration practitioners in all regions and habitats. The possibility that plants are most suited to their home environments due to genetic adaptations to local biotic and abiotic conditions prompts questions of how far plant material can be moved from home sites and remain ecologically appropriate in a restoration setting. We tested a suite of provisional seed transfer zones at multiple geographic scales to assess their ability to capture potentially adaptive genetic variability among populations in the southeastern United States. Furthermore, we examined the effects of seed source and phenotypic plasticity on plant performance and whether locally sourced individuals have adaptive advantages relative to more distantly sourced individuals. With a reciprocal transplant and a common garden study, we show that, although seed source is the best predictor of differentiation in plant performance, performance differences among populations across larger seed transfer zones within the longleaf pine ecosystem are relatively minor. These findings suggest that consideration of larger seed zones within the longleaf pine ecosystem that are also more logistically and economically viable is warranted. However, earlier flowering of individuals from seed sourced outside the longleaf pine ecosystem suggests that moving plant material greater distances is more likely to result in phenological mismatches between plants and pollinators. Species-specific differences, however, indicate there is insufficient evidence to support a recommendation for a single set of seed transfer zones within the southeastern United States for all species.

Key words: common garden, ecotype, genetic variation, longleaf pine ecosystem, reciprocal transplant experiment, seed transfer zones

- Similar species
 - 4 grasses (wiregrass, pineywoods dropseed, little bluestem, yellow Indiangrass)
 - 1 legume (Hairy lespedeza hirta)
 - 1 composite (anise-scented goldenrod)
- Similar traits measured
- Overlapping time frame
- East-West gradient, 4 ecoregions



Some results

- Most variation occurred between populations
- “Northern” populations flowered much earlier
- Recommended a seed zone map

Methods- Species selection criteria

ECOLOGICAL VALUES	MARKET VALUES	PRODUCTION VALUES/COSTS
dominant groups/common	wildlife value	habitat breadth
continuous fine fuels	fuels/management cost	range
wildlife value	floral display	ease of growing in gardens/fields
generalized pollinator		ease of collection
representative of intact habitat (conservative)		availability
		ease of germination

Grasses, composites, legumes



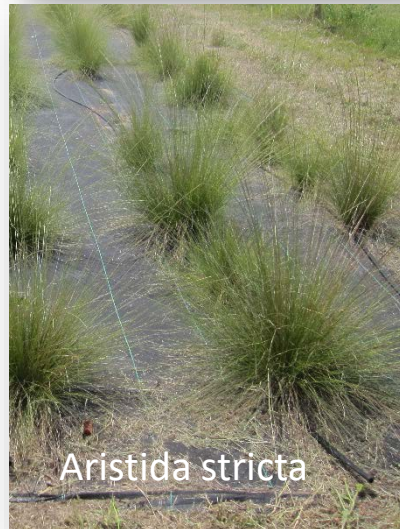
Schizachyrium
scoparium



Sorghastrum elliotii



Pityopsis graminifolia



Aristida stricta



Solidago odora



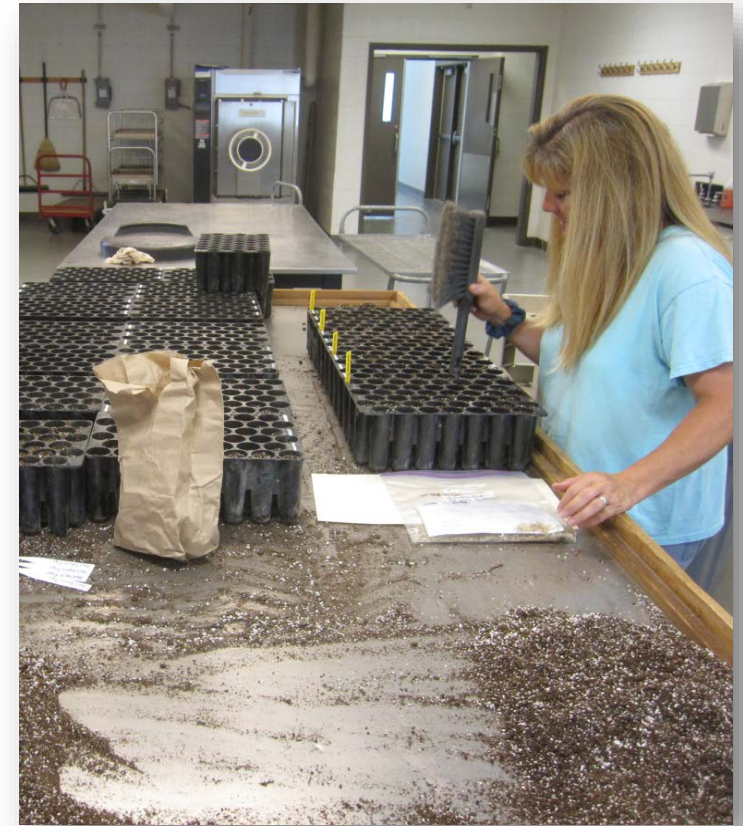
Lespedeza capitata



Tephrosia virginiana

Germination, Greenhouse culture

- Sowed seed in Fafard 3B media in blocks of cells 2.5 cm diameter x 15 cm deep
- Germinated under mist & transferred to hand-watering regime
- Grown in a glass house x 8 weeks, under natural light supplemented on cloudy days
- Fertilized 1/10 strength commercial liquid NPK or slow release

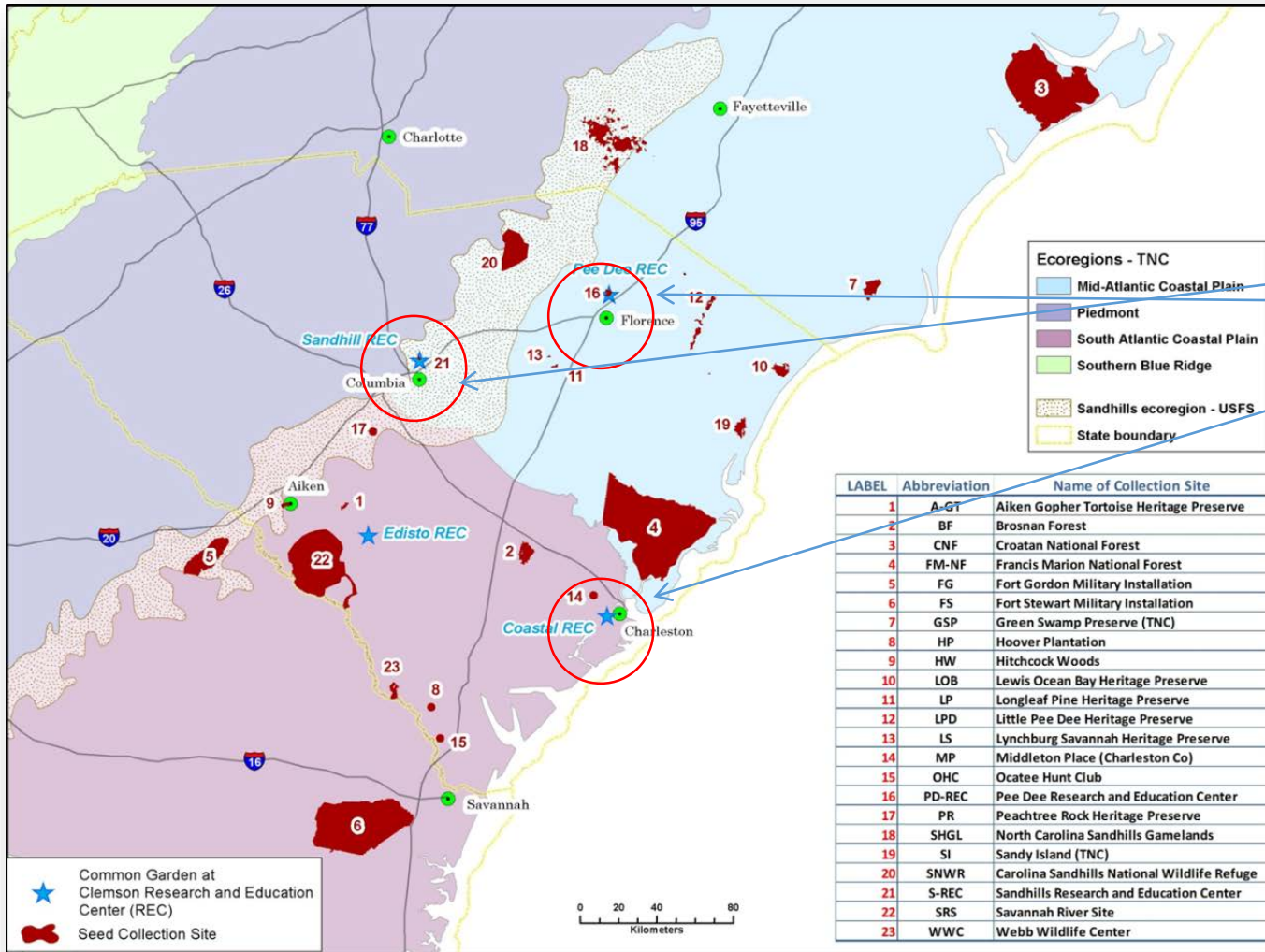


How to synchronize legume seeds germination?

Experiment: hot water, scarify sand paper, nick with razor blade, control



3 Gardens



Locations

- Middle Atlantic Coastal Plain (MACP)
- Middle Atlantic Coastal Plain-Sand Hills (MACPSH)
- South Atlantic Coastal Plain (SACP)
- Clemson University Research and Education Centers (REC)

Garden Installation

Sites were
experimental fields

Fallow at least 1 year
prior to garden
establishment

Not tilled

Landscape fabric laid
down for weed
control, moisture
retention



Response variables

- Traits that may be related to performance in the target planting area
- Height, number of leaves
- Biomass
- Flowering phenology
- Reproduction (biomass; seed mass)
- Specific Leaf Area, Carbon Isotopes
- C, N content in leaves



Preliminary data analysis

- Considered the effects of garden location, ecoregion
 - Analysis of Variance Models
- Data Analysis Design:
 - 3 (2) Garden locations
 - 3 different Ecoregion Models (represent collection zone)
 - ECO2: SACP, MACP
 - ECO3: SACP, MSC, SH
 - ECO4: SACP, SACPSH, MACP, MACPSH
- Separate analysis of variance for each trait, species, ecoregion model

Response = GARDEN Ecoregion GARDEN*INTERACTION



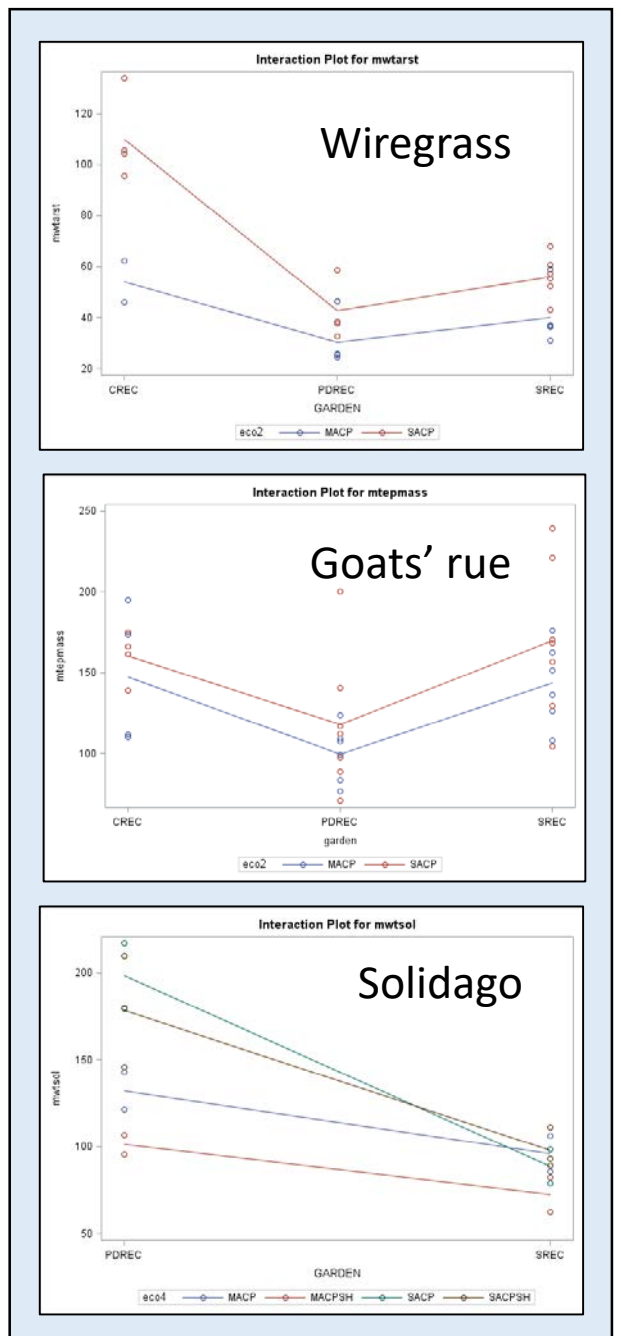
Recently
planted



Established Goats' Rue

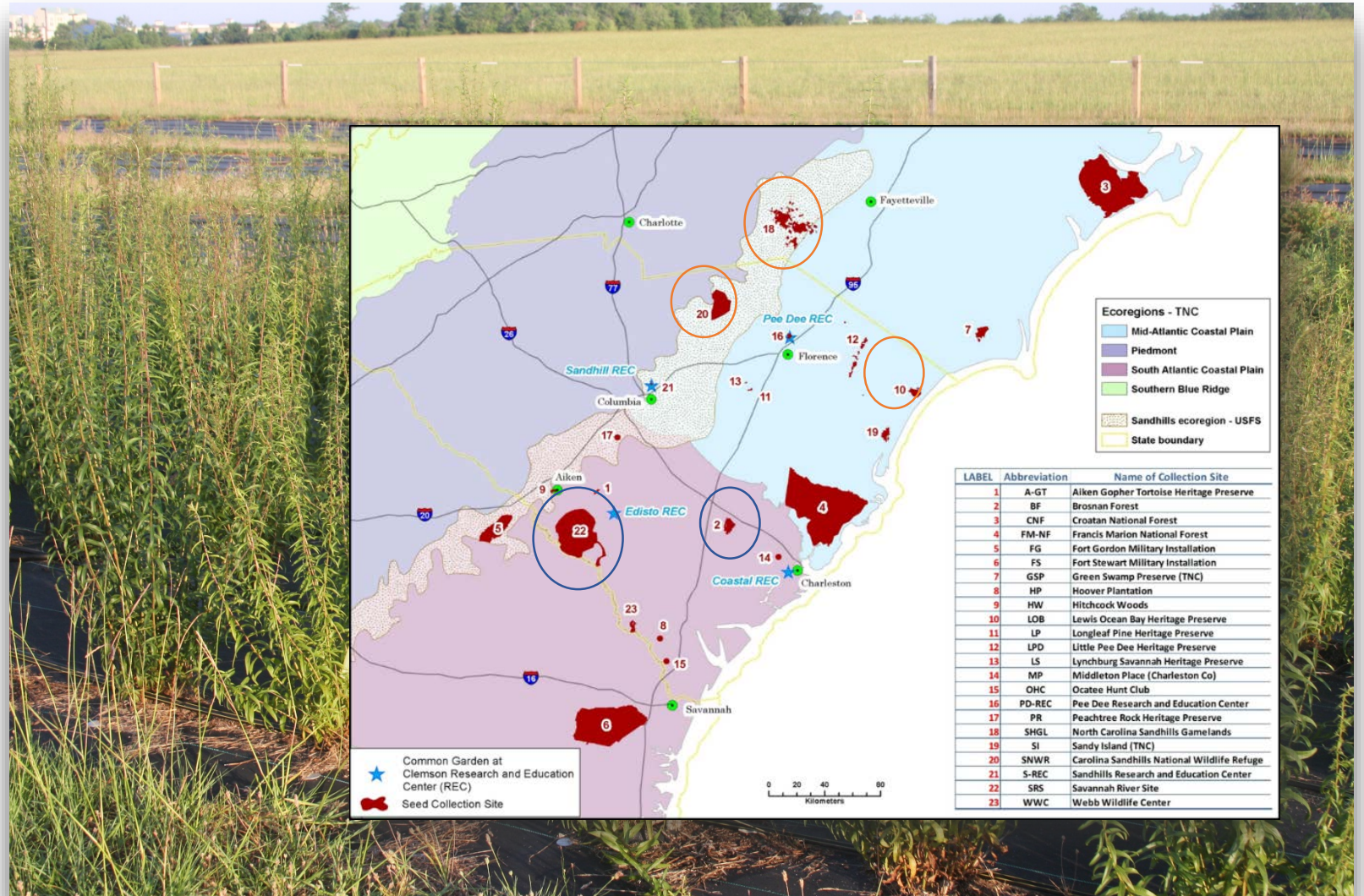
Biomass: model main effects

Species	Model	Garden	Ecoregion	Interaction
Wiregrass	ECO2 <.0001	<.0001	<.0001	.004
	ECO3 .0001	<.0001	.039	.168
	ECO4 <.0001	<.0001	.003	.089
Little bluestem	ECO2 .044	.009	.492	.469
	ECO3 .115	.012	.259	.875
	ECO4 .193	<.0001	.265	.907
Goats rue	ECO2 .017	.004	.136	.906
	ECO3 .138	.006	.744	.859
	ECO4 .192	.015	.350	.932
Round-headed Lespedeza	ECO2 .001	<.0001	.778	.798
	ECO3 .008	<.0001	.797	.975
	ECO4 --	--	--	--
Anise-scented Solidago	ECO2 <.001	<.0001	.001	.009
	ECO3 .011	.001	.350	.270
	ECO4 .000	<.0001	.007	.046



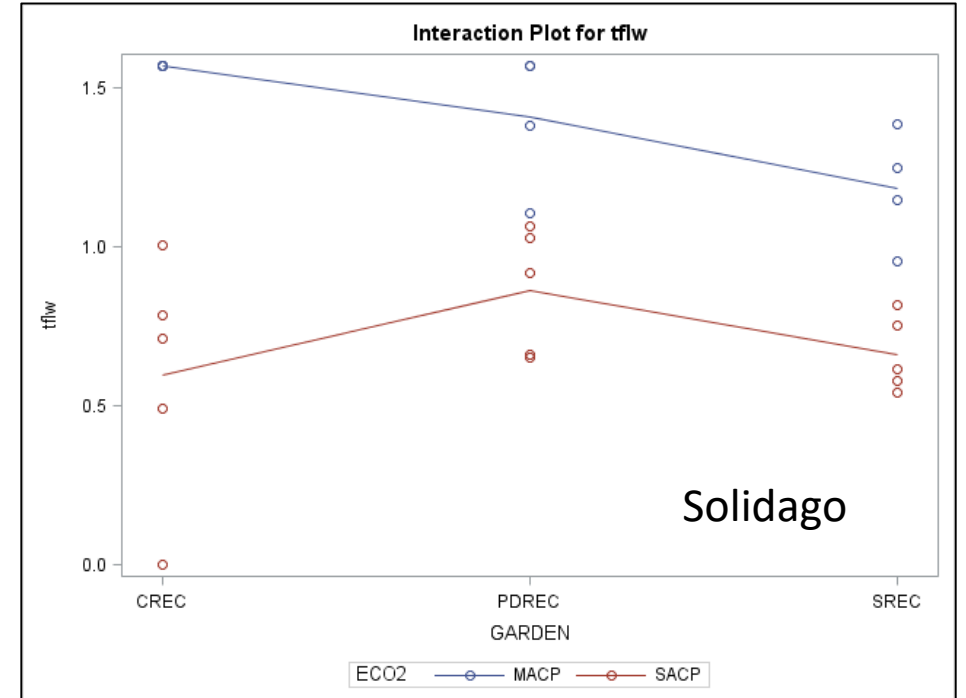
Results – Northern *Solidago* populations flower earlier

- 9 populations in the garden
 - 4 Middle Atlantic CP
 - 5 Southern Atlantic CP
- Flowering early
 - LOB, SHGL, SNWR
- Buds
 - PDREC, FG, WC, AGT
- Vegetative
 - SRS, BF

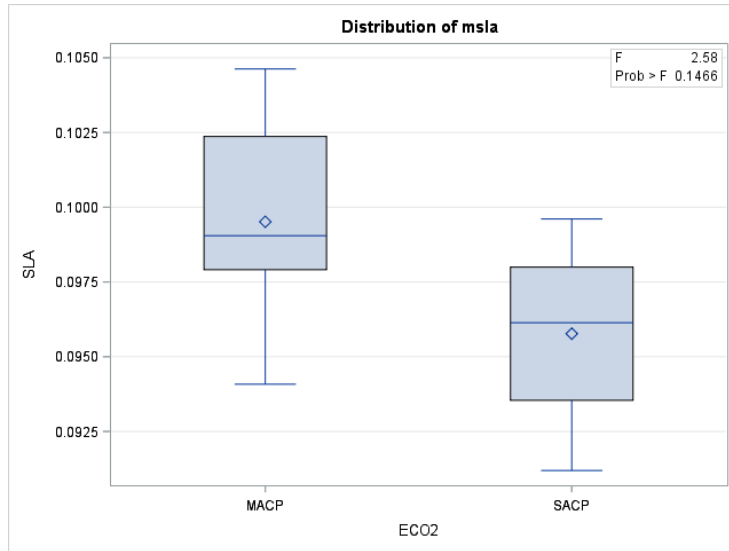


Percent flowering in September 2014 : main effects

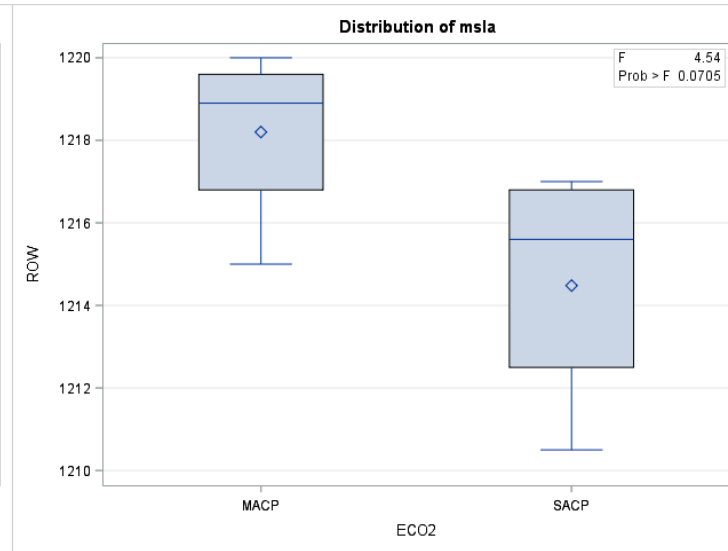
Species	Model	Garden	Ecoregion	Interaction
Round-headed Lespedeza	ECO2 .0016	.0002	.196	.963
	ECO3 .019	.0007	.471	.995
Anise-scented Solidago	ECO2 <.0001	.147	<.0001	.079
	ECO3 .396	.456	.076	.772
	ECO4 .0002	.089	<.0001	.095



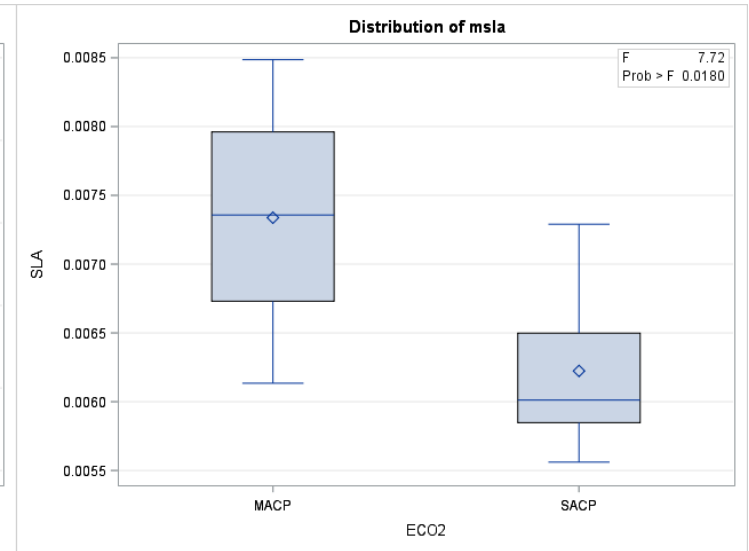
Specific Leaf Area, cm^2/mg



Lespedeza capitata



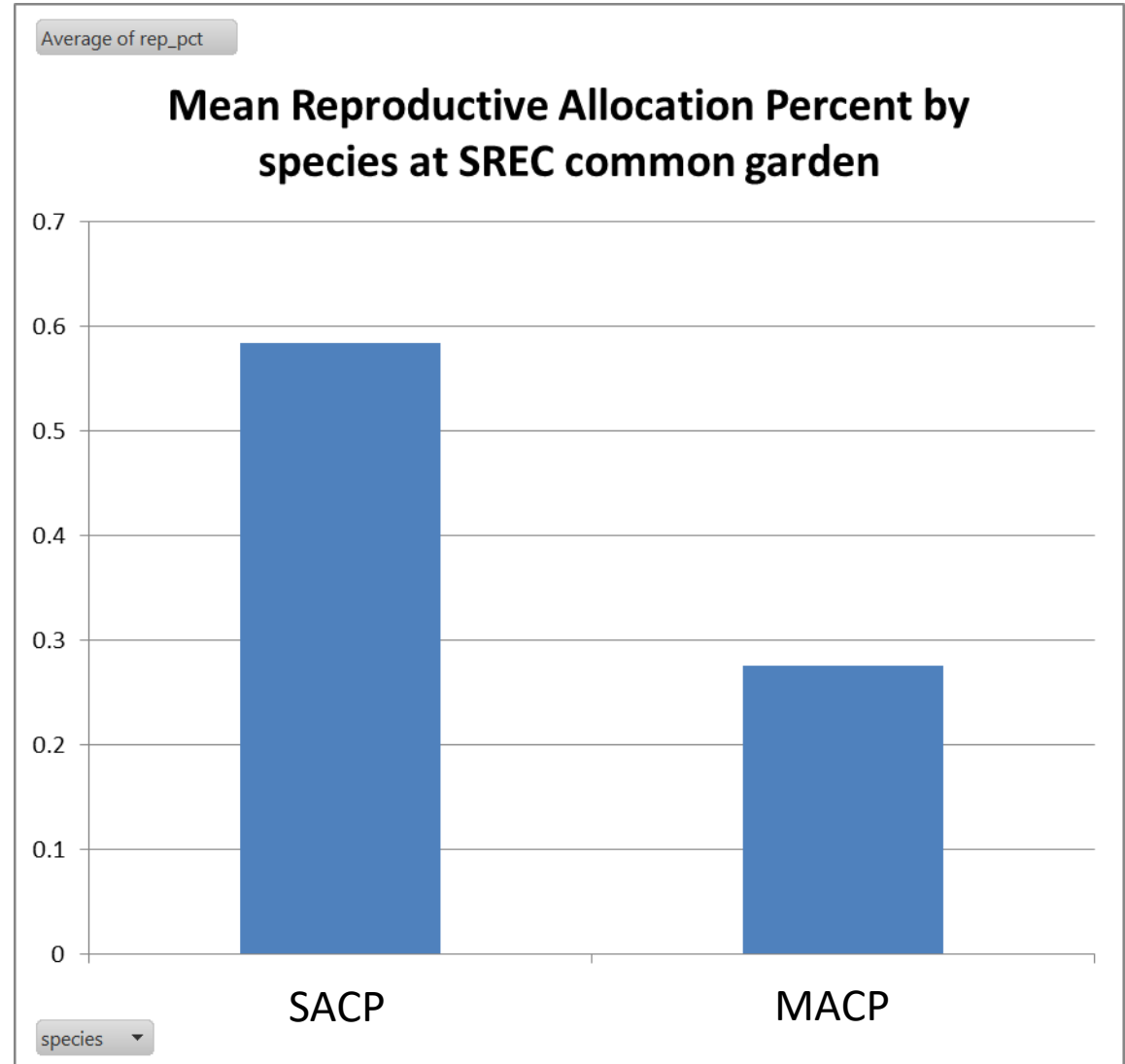
Solidago odora



Tephrosia virginiana

Results – *Aristida* spp reproductive effort

- Northern wiregrass (*A. stricta*) - MACP
- Southern wiregrass (*A. beyrichiana*) - SACP

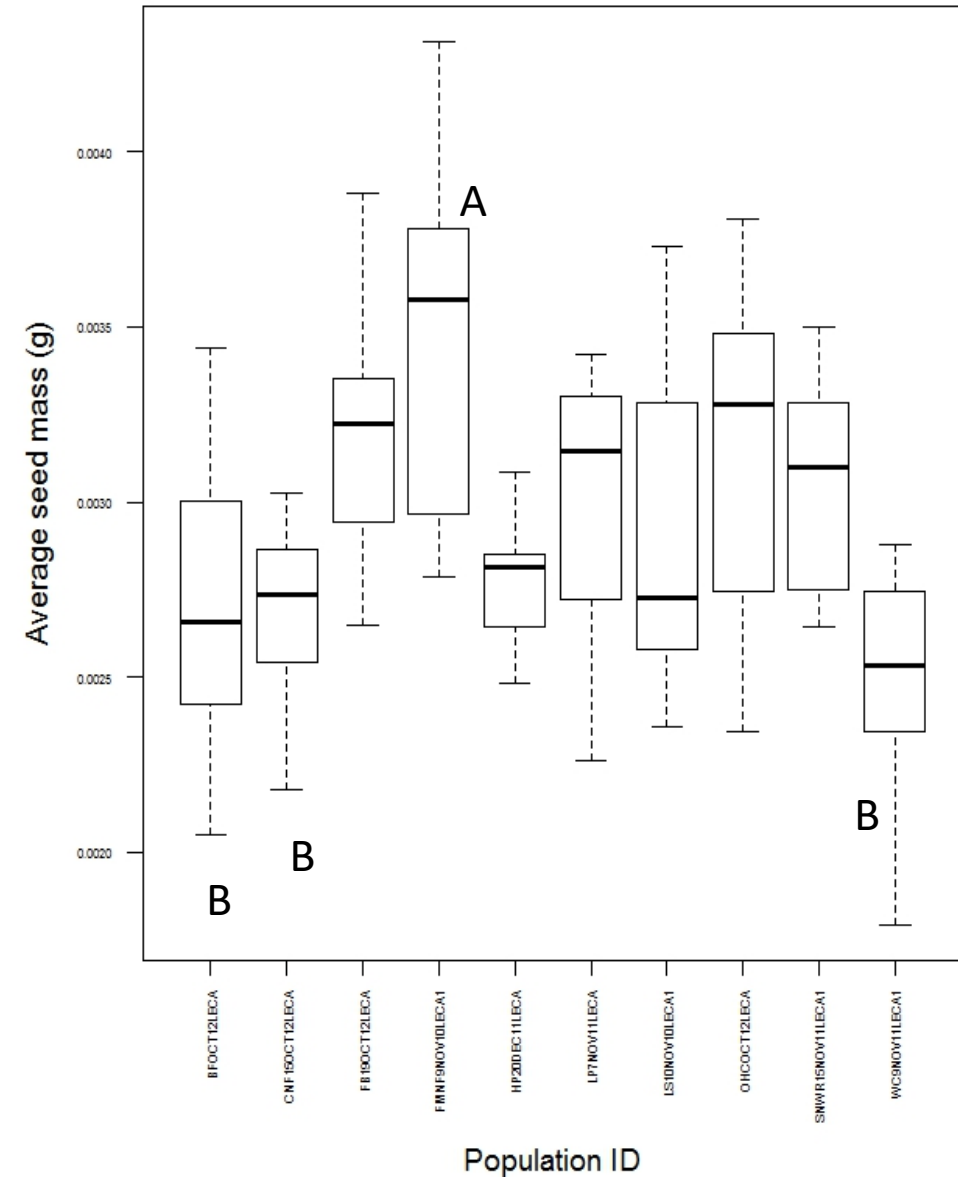


Results - Seed weight *Lespedeza*



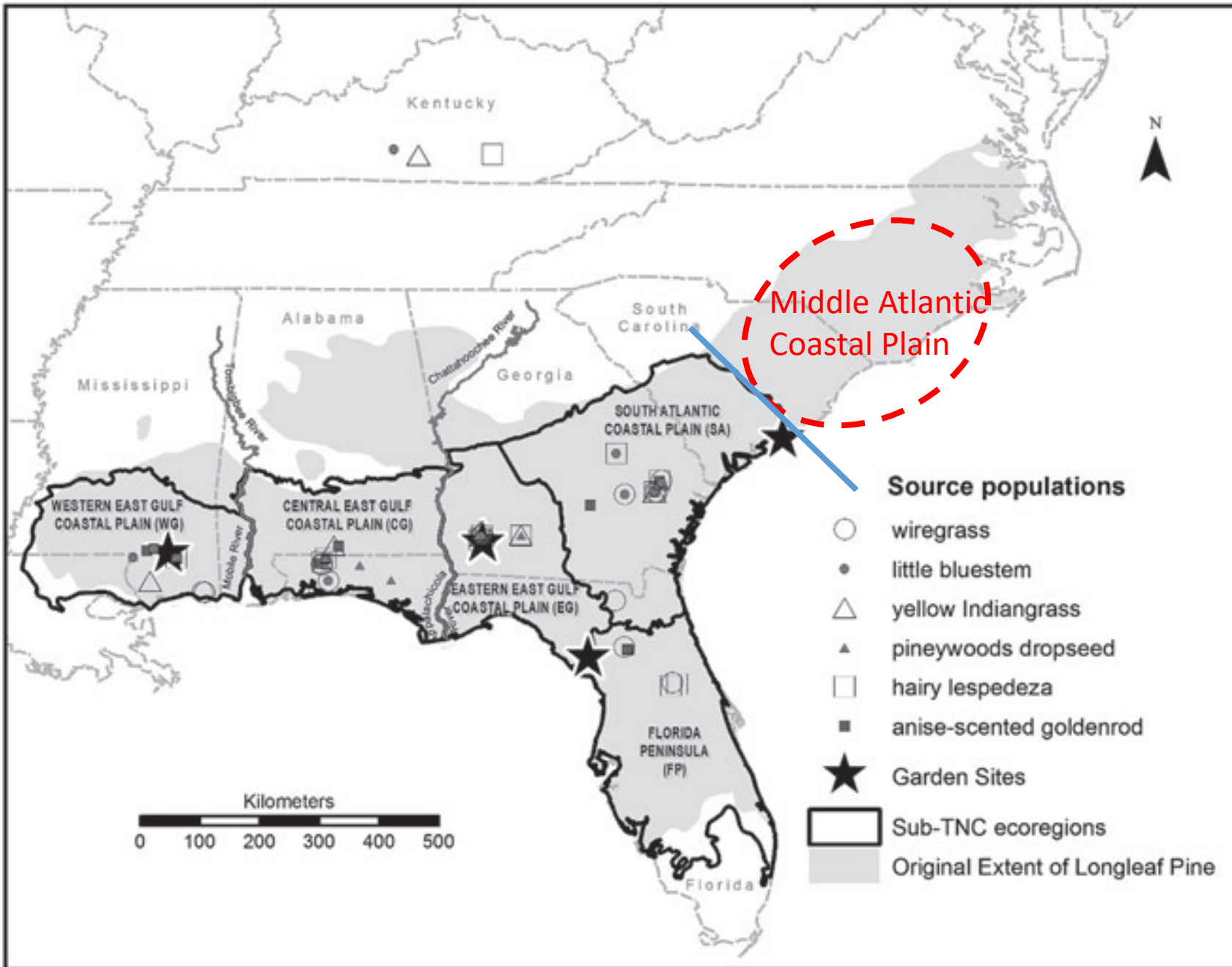
Populations vary, but not clearly related to geography

Average *Lespedeza capitata* seed mass at the SREC common garden



(Preliminary) Results Summary

- Always significant variation among populations of a species
- Evidence for geography related effect varied by species, trait
- Geographical models (ECO2, ECO3, ECO4) had significant effects for a subset
- Garden location had a significant effect on most traits
- Phenology differences may be strongest support for designating a separate Middle Atlantic Coastal Plain zone
- Interaction of Garden and Ecoregion effect were uncommon



Need for a seed zone collection model?

Need to know more...

- Performance in competitive situations
- More about germination and establishment
- Importance of within zone habitat variation
- Relationship to specific environmental factors that may change through time
- West Gulf Coast & Mountain Longleaf

Genetic diversity – relationship to geography

- Clemson University Graduate student, Jason Joines
- Expanded collections throughout the range of longleaf pine
- Preliminary study of genetic structure based on garden plants

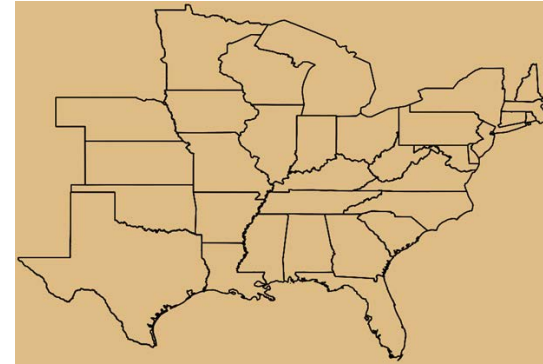
- Understand gene structure, inform seed source selection without 3 years of work



Goat's rue (*Tephrosia virginiana*)

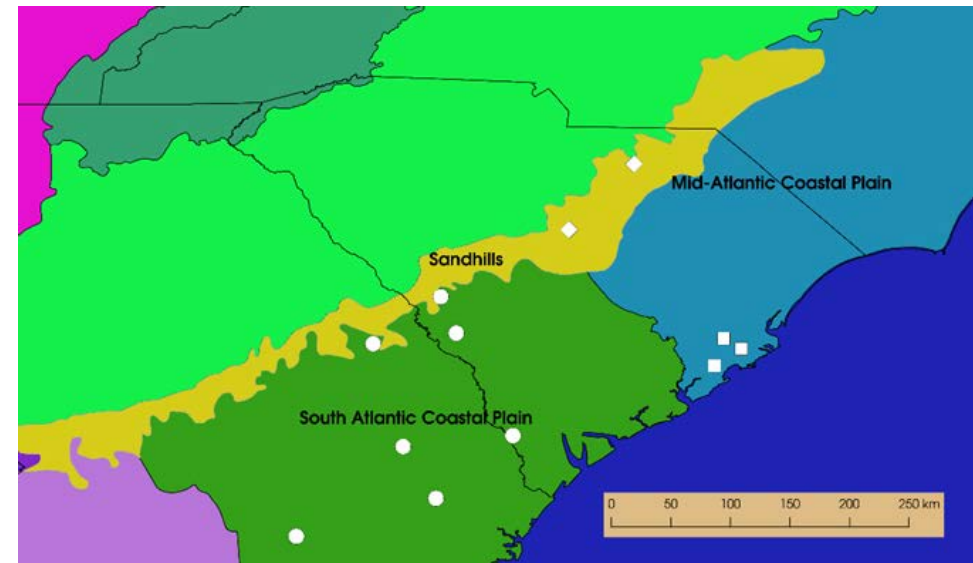
Genetic diversity related to geography

- 12 populations, 46 individuals
 - DNA extracted; GBS libraries created
- Libraries sequenced
- SNPs (single nucleotide polymorphisms) identified
- Mantel test of similarity between genetic & geographic distance
- AMOVA to examine the distribution of genetic variation among ecoregions, among collection sites within ecoregions, and among individuals within collection sites
- Principle Components Analysis to identify genetically similar groups

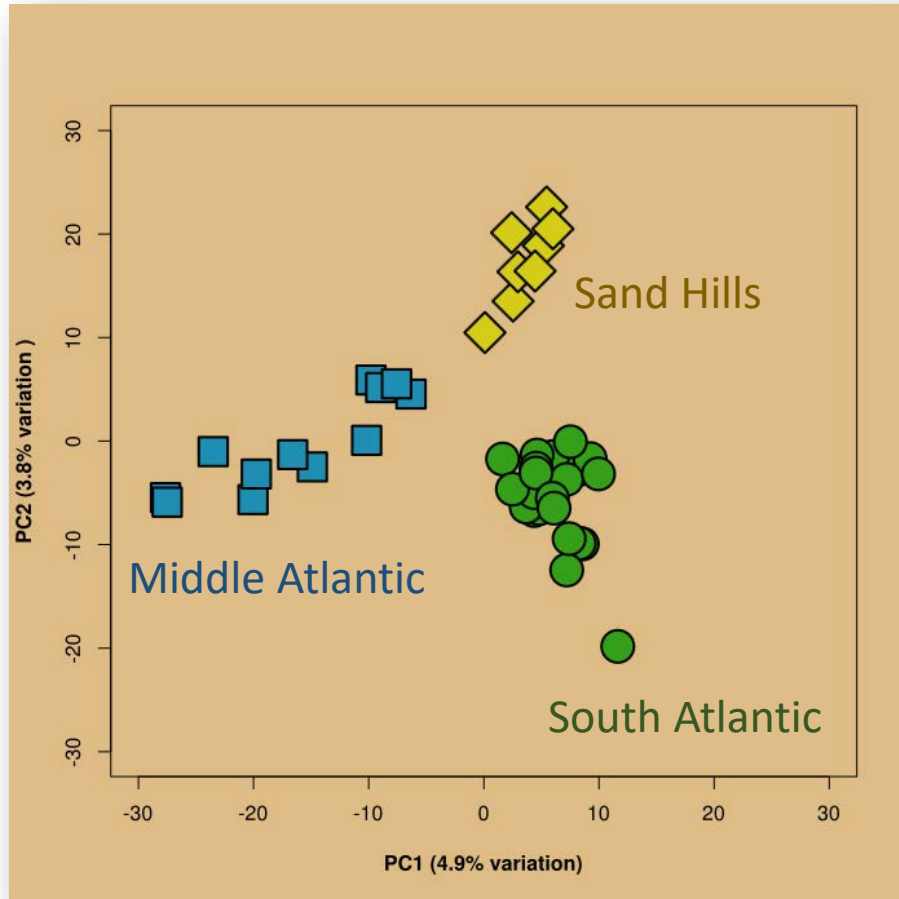


State level distribution of *Tephrosia*

Collection locations



Results



Variance component	% Variation	p-value
Among ecoregions	3.4	< 0.001
Among sites within ecoregions	12.3	< 0.001
Among Individuals within sites	84.3	

PCA of genetic dissimilarity: the first 2 principal components grouped individuals by ecoregion



So far, so good...

What's next?

Combine data into single
analysis (LLA Meeting)

Complete genetic studies
(2019)

Test results in field settings?



Thanks to our partners for collecting, testing, funding, providing garden space

- ❖ Jack Culpepper
- ❖ Nancy Jordan
- ❖ Bert Pittman
- ❖ Wayne Grooms
- ❖ Kathy Boyle
- ❖ Robin Mackie
- ❖ Bryan Mudder
- ❖ Gary Kaufman
- ❖ Jeff Glitzenstein
- ❖ Donna Streng
- ❖ Linda Lee
- ❖ Gary Burger
- ❖ Mike Walker

- ❖ Sudie Daves Thomas & family
- ❖ Lisa Lord
- ❖ Colette Degarady
- ❖ Allen Bridgmen
- ❖ Bob Franklin
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- ❖ Carol Denhof
- ❖ Ocatee Hunt Club Manager
- ❖ Hoover Plantation Manager
- ❖ Forest Service National Seed Lab
- ❖ Francis Marion Sumter NF
- ❖ RNGR staff
- ❖ Clemson Research & Education Directors and Staff

