

RESTORING THE RARE KENTUCKY LADY'S SLIPPER ORCHID IN THE KISATCHIE NATIONAL FOREST: OUTPLANTING STUDY RESULTS

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Cypripedium kentuckiense

CHARACTERISTICS OF KENTUCKY LADY'S SLIPPER (*Cypripedium kentuckiense* C.F. Reed)

- A tall, stately perennial plant with large flowers
- Underground rhizomes allow periods of plant dormancy
- Its range is the SE United States, but it is rare throughout the range
- Kisatchie National Forest has 4 sites on 600,000 acres.
- Occurs in mesic forests on stream slopes or acid steep sites



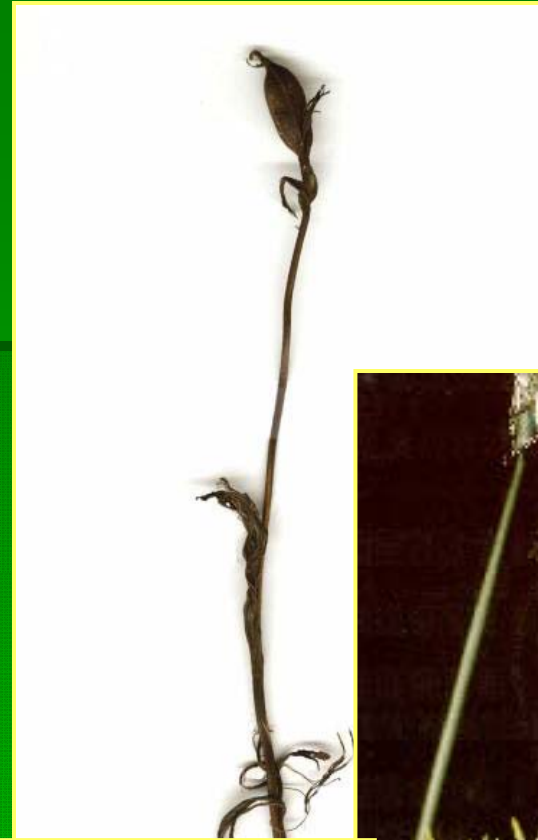
An Opportunity...

- Kevin Allen, amateur botanist and HS student in 2004—hand pollinated orchids.
- After 3 years of monitoring, discovered a seedpod at a Catahoula Ranger District site.
- Kevin has completing his master's degree and teaching in a Shreveport high school.



It's Alive...

- Kevin sent his seed pod to Spangle Creek Lab for germination.
- Seedpod proved to contain 1000s of viable seeds!
- Kevin contacted the KNF with his idea of re-introduction.



A COOPERATIVE EFFORT

- The KNF contacted the Central Louisiana Orchid Society (CLOS) and Southern Research Station (SRS) on a native orchid reintroduction project.
- CLOS was receptive. A research proposal was developed to fund the project.
- The Southwest Regional Orchid Grower's Association (SWROGA) provided a grant to buy plants and the project began in 2006.
- Additional grants from the Forest Service were obtained in 2007 and 2008.

OBJECTIVES OF THE STUDY

- Primarily to reintroduce up to several hundred rare orchids to KNF lands.
- Research various growing media and horticultural techniques from seedling to juvenile plant.
- Reintroduction methods: site selection, season, protection, age.
- In the spring of 2006 & 2007 plantlets were distributed to 10 orchid growers for development of cultural techniques.
- Growers kept records of their techniques.

EARLY SEEDLING DEVELOPMENT

- Early seedling growth was rapid. These seedlings were about 4 weeks old.
- Two or three leaves developed quickly.
- Most of the seedlings that were received were vigorous and healthy.



Many growers used trays like this.

Based on 2006 results, the medium used in 2007 was a commercial mix (Promix) with addition of sand. This combination resulted in a loose mix that favored seedling development and lifting for planting.



Dormancy “recognized”



Photos on October 9, 2006 of seedlings with dead and live shoots. Root systems of these seedlings were washed to evaluate condition.

Status of root systems of seedlings dormant and alive?



Condition of root systems of seedlings with a recently dead shoot (left) and one that has a green shoot (right). Both root systems are live and the seedling on the left is in a dormant state.

Size of planting stock

- Bill Cullina, New England Wildflower Society, indicates that 1-year-old seedlings have been outplanted with success. However, older plants may result in better establishment.



Lifting seedlings from trays

- Different methods were tested to evaluate the best way to lift seedlings from the trays.
- The method that worked best was to lift a block of the medium and a seedling with a spatula.
- Since the media is loose, the seedlings are easily separated without injury.
- Most seedling still have the dead shoot growth.



ROOT DEVELOPMENT OF 2006 AND 2007 SEEDLINGS AT PLANTING

- Before planting, numbers of roots and growing points were counted on each plant.
- Averages of data follow:

	2006 Crop		2007 Crop	
Rep.	Roots (#)	Shoots (#)	Roots (#)	Shoots (#)
1	7.6	1.0	9.0	1.2
2	6.2	1.2	6.6	1.4
3	6.6	1.0	7.2	1.0
4	5.6	1.0	10.2	1.6
Avg.	6.5	1.0	8.3	1.3



NATIVE CYPRIPEDIUM SEEDLINGS ON THE CATAHOULA RANGER DISTRICT



Two native *Cypridium* seedlings located on the Kisatchie National Forest's Catahoula Ranger District.



Photo taken April 15, 2008

PLANTING SITES/SOILS

- Kentucky lady's slippers require a site with few hours of direct sun a day and filtered or dappled shade for the rest.
- Soils should be moist, but not wet, and reasonably well drained.
- The pH should be slightly acid to nearly neutral.

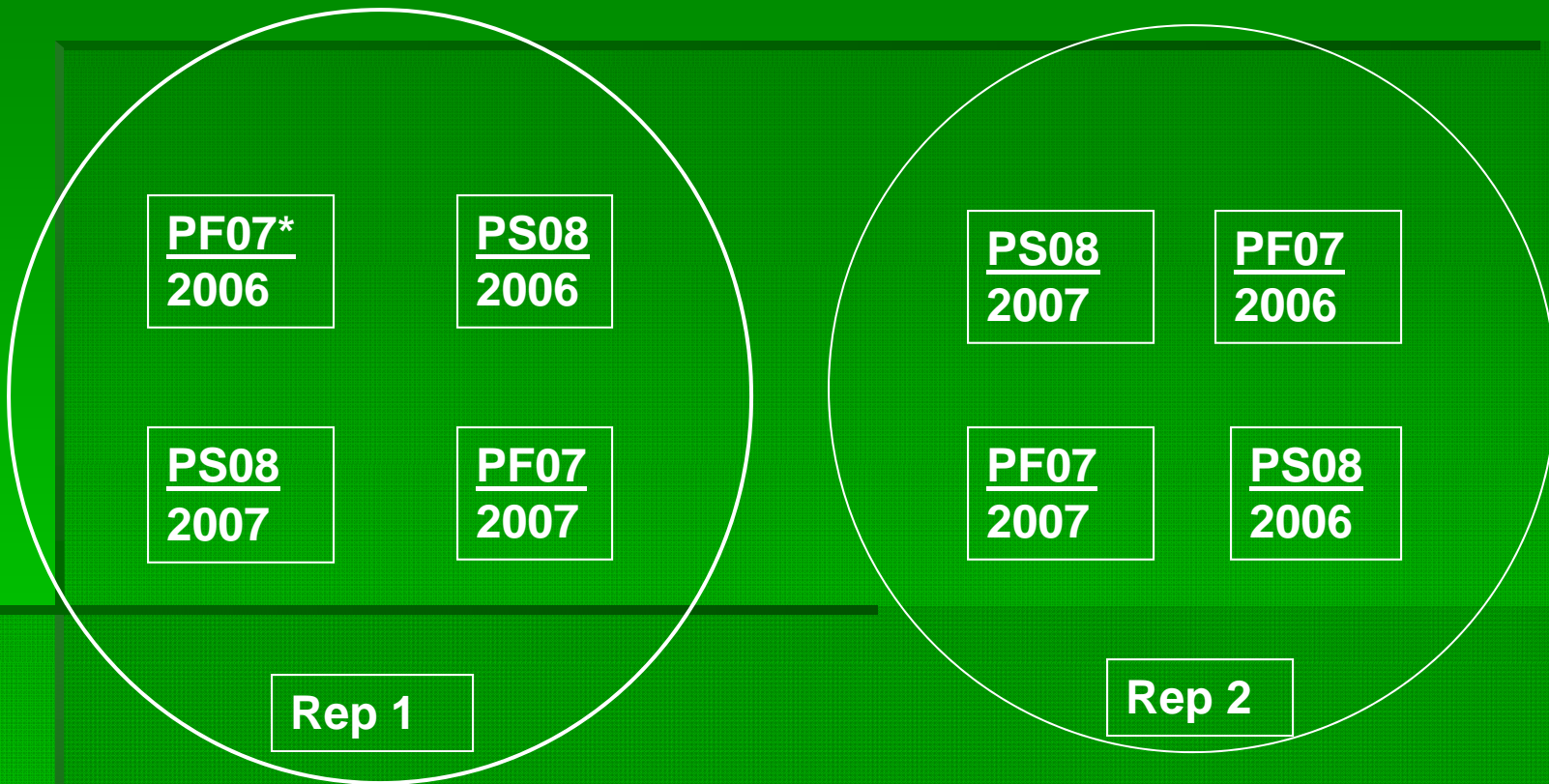


INDICATOR PLANTS

- Plants found on typical lady's slipper sites are beech and holly trees, Christmas ferns, and cranefly orchids.
- *Tipularia discolor* is a frequent orchid on the sites. Its leaves are bright green with a purple back in winter. The leaves die when flower stalks appear in the summer.



Layout of outplanting test, four replications of five seedlings



*In each 5-seedling plot, the top identification, e.g., PF07, represents the season of outplanting and the lower date represents the year of the crop.

PLANTING TECHNIQUES

- Work compost into the soil.
- Spread roots to preserve the root pattern and dribble soil to cover.
- When the plant is almost covered, keep tamping the soil lightly as you fill while holding the bud(s) so as to keep them at a proper depth.
- When complete the bud should be just breaking the surface.
- Cover the site with mulch.



Seedling characteristics and survival of various treatments when planted on December 18, 2007 and March 6, 2008

Planting variable	Characteristics at planting		Survival	
	# Roots	# Shoots	% 4/1/08	% 7/7/08
December 18, 2007 planting				
2006 crop	6.5 ^a	1.0	70	25
2007 crop	8.3	1.3	50	15
March 6, 2008 planting				
2007 unfertilized	8.4	1.3	85	15
2007 fertilized ^b	8.1	1.2	65	10

^a The numbers represent an average of 4 replications of 5 seedlings each.

^b Slow-release nutrients (one teaspoon of Osmocote 19-6-12) were applied at each planting spot after each seedling was planted.

Treatments imposed to seedlings at the December 30, 2008 planting of 2007 stock.

- Depth of planting is the primary treatment being evaluated in this test—1/2 and 1 inch depth.
- Seedling size/age comparisons will be made with earlier plantings.
- In a related test, inoculation with fungal material is being evaluated in side test.



Performance of 2007 crop seedlings grown in trays, lifted, and planted December 30, 2008, and measured in the field May 20, 2009 and April 6, 2010

Variables measured	Planting Treatments ^a	
	Shallow	Deep
Characteristics at planting—December 30, 2008		
Roots (number)	10.8 ^b	10.2
Shoots (number)	1.0	1.0
Measurements on May 20, 2009		
Survival (percent)	92	60
Measurements on April 6, 2010		
Survival (percent)	15	10

a The planting depths were ½ inch (shallow) and 1 inch (deep) below the soil surface.

b The numbers presented are averages of 40 seedlings (8 reps of 5 seedlings each).

CONCLUSIONS

- Shallow planting is more successful than deep
- Fertilizing at planting was not successful
- If dormant when planted, season of planting is not important
- Seedling age or stage of development is very important
- Caging of planting site is not needed
- Inoculation with mycorrhizae seems important
- Additional research is needed to establish reintroduction guidelines



Effects of fungal inoculation of potted seedlings

Seedling treatment	Survival at 6 months
Inoculated with soil humus	52 percent
Control (not inoculated)	37 percent

CONTINUING RESEARCH

- Grants have been received to fund Kevin Allen's high school honors chemistry class' efforts to develop *Cypripedium* seed germination and developmental techniques.
- Kevin Allen will train with Dr. Zettler at Illinois College to begin fungal relation studies with seed germination and seedling establishment.
- Plantlets from these germination studies will in grown in garden situations to develop larger plants for field outplanting.
- Tests of seedling size at planting, depth of planting, fungal inoculation, and planting site will continue.



Our goal is to reintroduce these orchids