ECOLOGICAL STUDIES OF WILLOW (*SALIX CAROLINIANA*): MONTHLY STATUS REPORT #15

Pedro Quintana-Ascencio
John E. Fauth
Luz M. Castro-Morales

Willows planted within UCF ponds to investigate effects of flooding and competition with sawgrass

Department of Biology, University of Central Florida,
4000 Central Florida Boulevard, Orlando, Florida 32816

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Ecological Studies of Willow (*Salix caroliniana*): Monthly Status Report #15

Covering the time period from April 1-30, 2010

This status report summarizes progress made on the Ecological Studies of Willow project through April 30, 2010, with reference to the tasks and timeline outlined in the Scope of Work and presented in Table 1 below.

Table 1. Timeline of tasks to be accomplished in Year 2. Tasks initiated and underway in this reporting month are highlighted in blue, completed tasks in red.

YEAR 2

Quarter	Months	Tasks accomplished
1st	Oct – Dec, 2009	Initiate Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*) Continue Task 2.5 (*Spatial analysis of willow distribution*)
2nd	Jan – Mar, 2010	Continue Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*) Continue Task 2.5 (*Spatial analysis of willow distribution*)
3rd	Apr – Jun, 2010	Initiate Task 2.2 (2nd iteration, *Willow transplantation*) Continue Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*) Continue Task 2.5 (*Spatial analysis of willow distribution*)
4th	Jul – Sep, 2010	Complete Task 2.2 (2nd iteration, *Willow transplantation*) Continue Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*) Continue Task 2.5 (*Spatial analysis of willow distribution*) Complete Task 3.2 (*Data analysis and final report, Year 2*)

Progress on Task 2.1 – Germination and Early Survival and Growth Experiments

During this quarter, we completed a growth chamber experiment that determined how long willow seeds remain viable. Briefly, between March 5, 2010 and April 22, 2010, we collected willow seeds from the southern (State Route 50) and northern (State Route 60) regions of the Upper St. Johns River basin. Seeds were collected early (SR 60, March 11; SR 50, March 19), in the middle (SR 60, March 19; SR 50, April 8) and late (SR 60, March 8; SR 50, April 22) in each region's flowering season. We collected viable (green) seeds from three different individuals per region, and organized seeds into groups of thirty, which were placed onto filter paper within a Petri dish. We then randomly selected and applied one of six different treatments: watering on the same day seeds were collected, or 2, 4, 6, 8 or 10 d later. During this time, seeds were maintained in a CMP 4030 (CONVIRON, Canada) growth chamber that reproduced spring temperature regimes for either Titusville, FL (north) or Lake Wales, FL (south). Thus, the experiment had all possible combinations of four main factors: Season and location of seed collection, temperature regime in the growth chamber, and the number of days that seeds were held prior to watering (Table 2).

Analysis of variance revealed that all four main effects were statistically significant, as well as three 2-way interactions and two 3-way interactions (Table 2). By far the single most important factor was the number of days before watering. Almost no willow seeds germinated when they were held 10 d without watering and seeds collected in mid-season remained viable the longest (Fig. 1). Temperature regime also was statistically significant, with seeds germinated under the cooler, northern temperature regime remaining viable slightly longer than those germinated under the warmer, southern temperature (Fig. 2). This result was independent of the number of days to watering (Table 2, non-significant D x T interaction). Similarly, seeds collected in the north location remained viable longer than those collected in the south, particularly toward the end of the 10-day holding period (Fig. 3).

Table 2. Analysis of variance of proportion of willow seeds germinating. Data angularly transformed to meet analysis assumptions. Adjusted R-squared = 0.94.

Source of variation	df	SS	MS	F	P
Days (D)	5	42.84	8.57	623.04	**<0.0001**
Location (L)	1	0.07	0.07	5.13	**0.025**
Temperature regime (T)	1	0.06	0.06	4.63	**0.033**
Season (S)	2	0.70	0.35	25.41	**<0.0001**
D x L	5	0.52	0.10	7.53	**<0.0001**
D x T	5	0.07	0.02	1.08	0.374
L x T	1	0.00	0.00	0.05	0.827
D x S	10	1.20	0.12	8.72	**<0.0001**
L x S	2	0.69	0.35	25.08	**<0.0001**
T x S	2	0.04	0.02	1.33	0.267
D x L x T	5	0.03	0.01	0.41	0.843
D x L x S	10	0.38	0.04	2.78	**0.003**
D x T x S	10	0.08	0.01	0.55	0.853
L x T x S	2	0.09	0.05	3.40	**0.036**
D x L x T x S	10	0.05	0.01	0.35	0.964
Residuals	144	1.98	0.01		
Total	215	48.8			

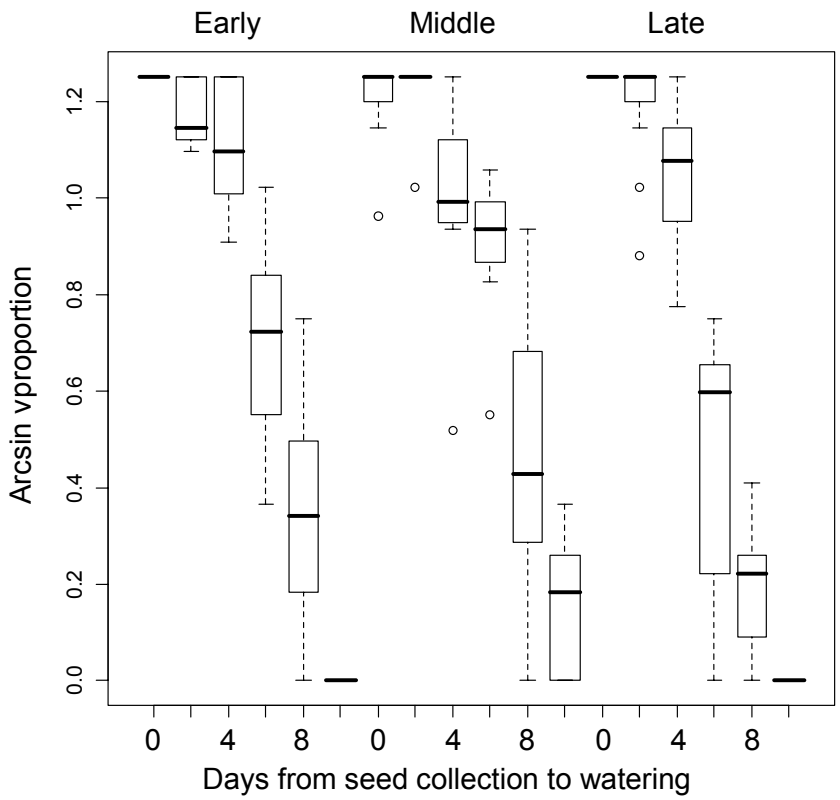

Fig. 1. Box plot of willow germination as a function of season of collection and days until watering.

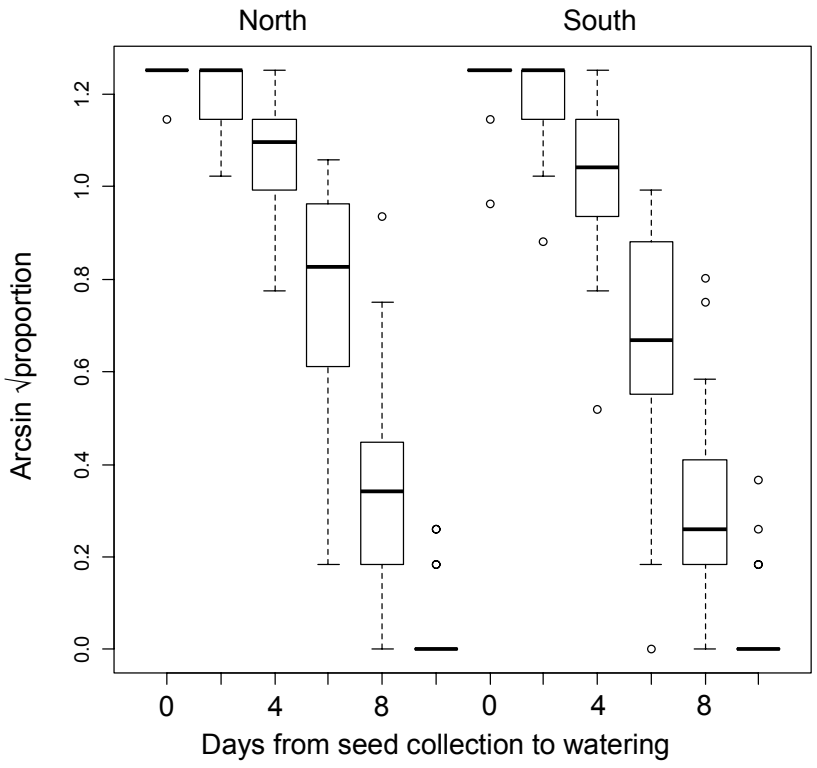

Fig. 2. Box plot of willow germination as a function of temperature regime and days until watering. The northern temperature regime was based on Titusville, FL and the southern regime on Lake Placid, FL.

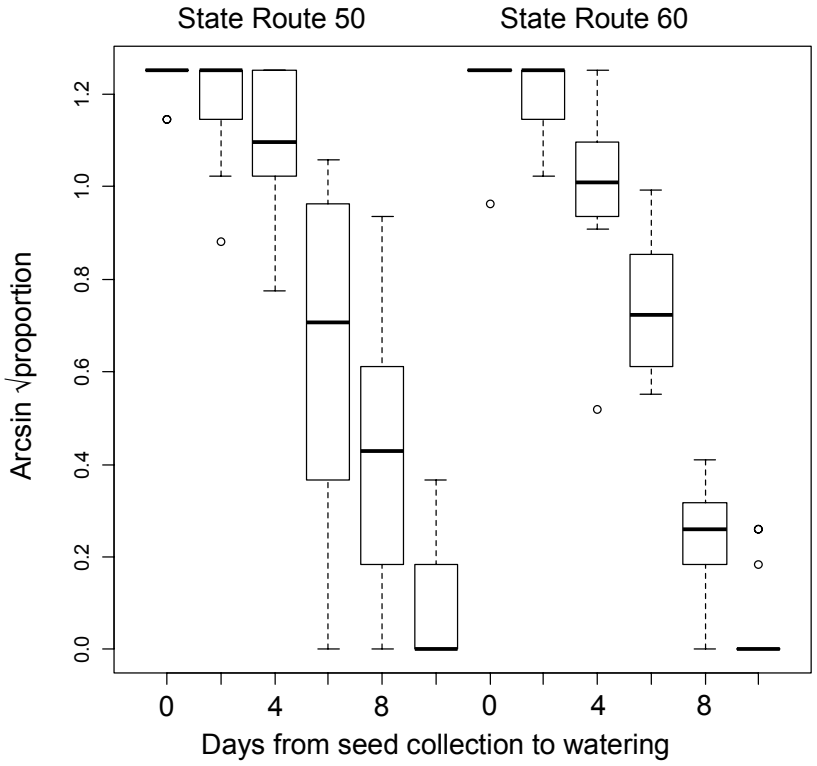

Fig. 3. Box plot of willow germination as a function of collecting locality and days until watering.

Progress on Task 2.2 – Willow Transplantation

A. Competition Experiment – We took advantage of the UCF experimental ponds to begin the flooding experiment (cover photo). Our design (Figure 4) overlaps this experiment with another, new experiment, which investigates competition between willow and sawgrass (*Cladium jamaicensis*).

In the flooding experiment, we planted three types of willow (seedlings, and short [15 cm] or tall [30 cm] cuttings) at each of four heights above the pond waterline: 0, 18, 35 and 50 cm. Each treatment was replicated twice within each of six experimental blocks, which are located on the north and south sides of three separate ponds.

In the competition experiment, we planted each willow type surrounded either by three or six plugs of sawgrass, which we obtained from a commercial supplier. Sawgrass plugs were spaced equidistant from each other and 15 cm from the central willow plant (Fig. 5). We placed this design only at the 0.25 and 0.38 cm heights above the waterline, because

we expected the sawgrass would die at the two extreme heights. We replicated the sawgrass competitor treatments once in each block.

Willow Competition Experiment

In Osborne's Ponds

Depth (m)	Willow alone	Low Competition with sawgrass (n = 3)	High Competition with sawgrass (n = 6)
0	N = 12		
0.25	N = 12	N = 6	N = 6
0.38	N = 12	N = 6	N = 6
0.50	N = 12		

x **3 willow types:**
Seedlings
Short cuttings
Tall cuttings

Fig. 4. Design of the combined experiments on flooding and competition. The flooding design is in the second column from the left and the competition experiment is in the middle two rows. The experiments overlap, sharing the treatments of willows grown alone at 0.25 and 0.38 cm heights above the pond level. The number of replicates is provided in each cell.

We are watering the plants as necessary while they become established, and then will gradually raise the water level until plants at the highest level are at the waterline. At this point, the other plants will be 0.12, 0.25 or 0.5 m underwater. We will monitor the plants monthly throughout the summer, recording survival, growth, and the competitive effect of sawgrasses on willows. Willows with no sawgrass planted around them serve as the no-competition control and also are part of the flooding experiment (Fig. 4).

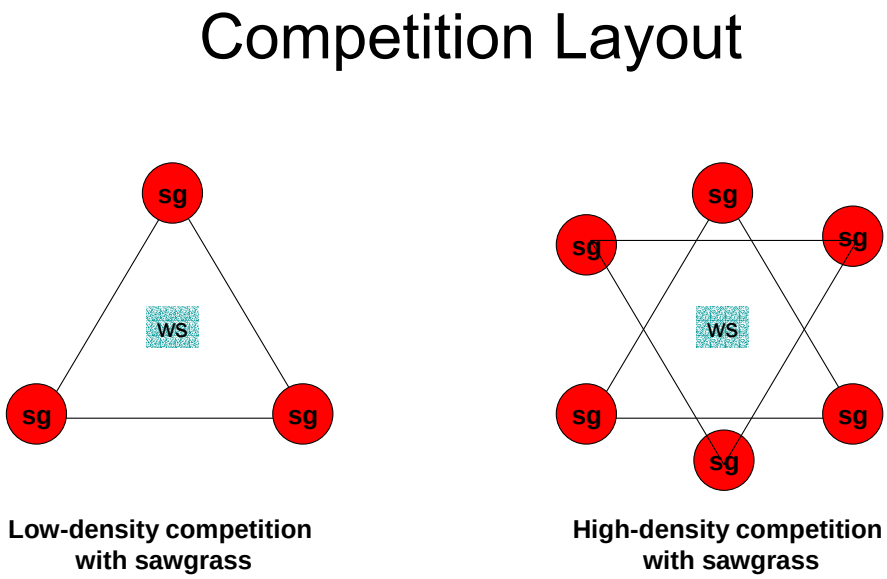

Fig. 5. Arrangement of willow (ws, blue squares) and sawgrass (sg, red circles) in the low- and high-density treatments of the competition experiment.

B. Hydrology Experiment – In previous reports, we summarized results of the hydrology experiment as they pertained to willow seedlings. Below, we summarize results of the island experiment for willow cuttings.

All but six willow cuttings survived until the end of the experiment, illustrating the plant's tolerance for different conditions once it grows beyond the seedling stage. Three-way factorial ANOVA revealed no significant variation in final cutting height or the percentage change in basal diameter among islands, height above the marsh or directions (all $F < 3.14$, $P > 0.096$). Thus, the overall plant morphology was similar despite the variation in environmental conditions that greatly affected seedlings. However, the percentage change in the number of leaves varied significantly among islands, heights, directions, and with the island x height and island x direction interactions (Table 3). The full model accounted for almost all of the variation in this parameter ($R^2 = 0.97$). On average, willow cuttings on Island 3 gained leaves (3%) while cuttings on the other islands lost 0.1-1%. Willow cuttings planted 35 cm above the initial marsh waterline also gained a greater percentage of leaves (1%) than those planted 17.5 cm above the waterline, which lost 0.25% of theirs. And on average, willow cuttings planted on the

Table 3. Analysis of variance of the percentage change in number of willow leaves in the island experiment.

Source of variation	df	SS	MS	F	P
Island (I)	3	42.31	14.10	27.1	**0.0003**
Height above marsh (H)	3	7.42	2.47	4.75	**0.0412**
Direction (D)	3	11.59	3.86	7.42	**0.0141**
I x H	9	20.78	2.31	4.44	**0.0311**
I x D	9	22.84	2.54	4.87	**0.0243**
H x D	9	11.21	1.25	2.39	0.1315
Residuals	7	3.64			
Total	43	137.18			

south sides of islands gained a greater percentage of leaves (1.4%) than those planted on the north side, which lost 0.6% of theirs. Of the two statistically-significant two-way interactions, the island by height is the more interesting (Table 3). Willow cuttings on Island 3 and initial height 35 cm grew more leaves than cuttings at almost every other island and height combination (Fig. 6). The small net loss of leaves elsewhere reflects cuttings that shed leaves at the beginning of their reproductive season. Several of these plants already were producing flowers but had not yet set seed when the experiment was terminated on February 13, 2010.

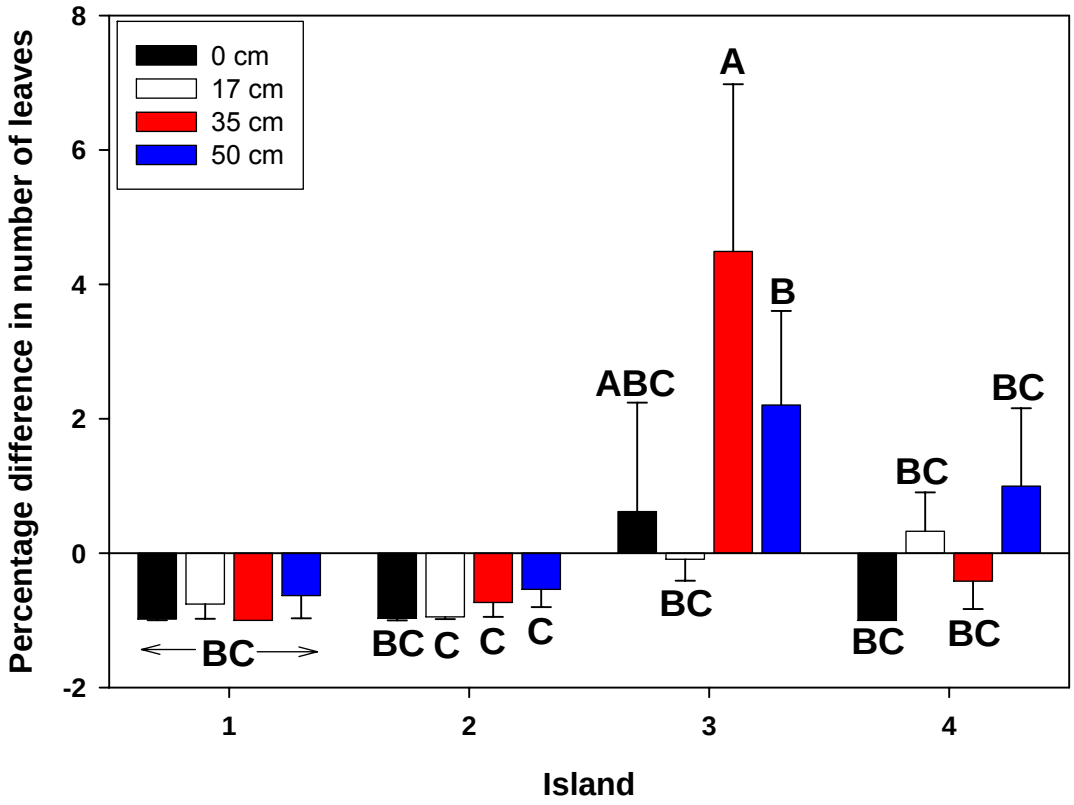

Figure 6. Mean (+ 1 SE) percentage difference in the number of leaves on willow cuttings planted on four replicate islands and at four different heights above the initial marsh water level.

Initiate Task 2.3 - Fire response

Unusually high water levels again prevented conducting the prescribed burns for this experiment.

Progress on Task 2.4 - Life History

We did not gather additional life history information during this period. Expanding this data set will be the focus of our summer work.

Progress on Task 2.5 – Spatial Analysis of Willow Distribution.

We did not modify our existing spatial model during this period.

Summary of Activity

During this reporting period, the UCF team maintained seedlings and cuttings for eventual use in the fire experiment; completed the seed viability experiment; and began two experiments that had some overlap in treatments. One experiment investigated the potential for flooding to prevent establishment of willows, and the other experiment investigated competition with sawgrass.