ECOLOGICAL STUDIES OF WILLOW (*SALIX CAROLINIANA*): MONTHLY STATUS REPORT #11

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Ecological Studies of Willow (*Salix caroliniana*): Monthly Status Report #9
Covering the time period from December 1-31, 2009

This status report summarizes progress made on the Ecological Studies of Willow project through December 31, 2009, 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 Years 1 & 2. Tasks initiated and underway in this reporting month are highlighted in blue, completed tasks in red.

YEAR 1

Quarter	Months	Tasks accomplished
1st	Oct – Dec, 2008	Initiate and complete Task 1 (*Finalize research plan*)
2nd	Jan – Mar, 2009	Initiate Task 2.1 (*Germination & early survival and growth experiments*) Initiate Task 2.4 (*Life history*)
3rd	Apr – Jun, 2009	Continue Task 2.1 (*Germination experiment*) Initiate Task 2.2 (*Willow transplantation*) Initiate Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*)
4th	Jul – Sep, 2009	Continue Task 2.4 (*Life history*) Complete Tasks 2.1 & 2.2 (*Germination experiment & Willow transplantation*) Complete Task 3.1 (*Data analysis and final report, Year1*)

YEAR 2

Quarter	Months	Tasks accomplished
1st	Oct – Dec, 2009	Continue Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*)
2nd	Jan – Mar, 2010	Continue Task 2.3 (*Fire response*) Continue Task 2.4 (*Life history*) Initiate 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, Year2*)

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

In previous reports, the UCF team presented its initial analyses of greenhouse experiments on willow seedlings and cuttings. Data included in these reports were height and crown growth of seedlings, and height and stem diameter of cuttings. Below, we present initial analyses of changes in the number of leaves for both seedlings and cuttings. We also summarize willow responses for both experiments.

We analyzed variation in the change in number of leaves as:

$$change = \log\left(\frac{final\_count}{initial\_count}\right)$$

Non-treatment variation was controlled by using number of leaves (or its logarithm) at the beginning of each interval as a covariate. This allometric function accounted for significant variation in the change innumber of leaves for both stages (seedlings and cuttings; Fig 1, and Tables 1 and 2). The leaf count of one seedling was missed during one evaluation, so we analyzed data without this single plant.

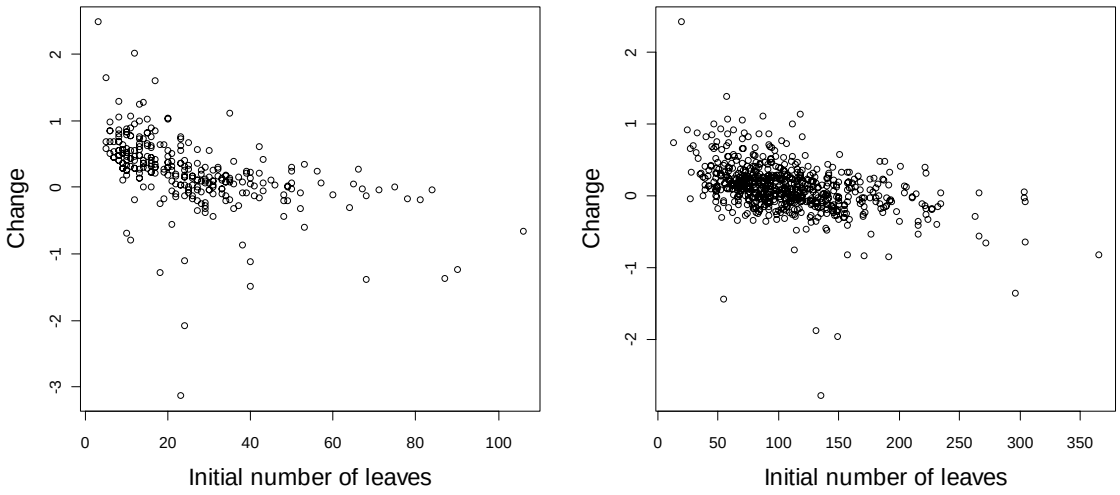

Fig. 1. Allometric relationship between initial number of leaves and change in number of leaves in willow seedlings (left) and cuttings (right).

Table 1. Analysis of covariance of change in number of leaves as a function of soil, nutrients and watering regime for seedlings. All plants on BC soil or its combinations were included except one plant without data in one evaluation. Log of initial number of leaves was used as a covariate (r^2=0.34). Table entries are sources of variation, degrees of freedom (df), sums of squares (SS), mean squares (MS), F-ratios (F) and their associated probabilities (P).

Response:	Seedlings				
	df	SS	MS	F	P
log(initial)	1	25.035	25.035	130.986	2.200E-16
Soil	2	2.615	1.308	6.842	1.263E-03
Nutrients	5	0.414	0.083	0.433	0.826
Watering	3	2.026	0.675	3.533	0.015
log(initial):Soil	2	0.55	0.275	1.439	0.239
log(initial):Nutrients	5	0.871	0.174	0.912	0.474
Residuals	271	51.795	0.191		

Table 2. Analysis of covariance of change in number of leaves with soil, nutrients and watering regime for cuttings. All surviving plants were included. Initial number of leaves was used as covariate (r^2=0.21). Table entries are sources of variation, degrees of freedom (df), sums of squares (SS), mean squares (MS), F-ratios (F) and their associated probabilities (P).

Response:	Cuttings				
	df	SS	MS	F	P
log(initial)	1	13.982	13.982	140.509	2.20E-16
Soil	5	3.357	0.671	6.747	3.89E-06
Nutrients	5	0.708	0.142	1.422	0.2142
Watering	3	0.896	0.299	3.001	0.0300
log(initial):Soil	5	0.426	0.085	0.857	0.5097
log(initial):Nutrients	5	1.463	0.293	2.940	0.0124
Residuals	623	61.993	0.100		

For both seedlings and cuttings, the most informative model of change in number of leaves included the main effect of soil and watering treatments and the covariable initial number of leaves (Tables 3 and 4). Residuals of the best models (Fig. 2) were randomly distributed, normal and independent, illustrating the effectiveness of the allometric covariate.

Table 3. AIC and AIC weights for models of change in number of leaves for seedlings

Models for change in the number of leaves for seedlings	df	AIC	Akaike weight
log(initial) + Soil + W	8	349.3	0.8623
log(initial) + Soil	5	354.3	0.0708
log(initial) * Soil	7	355.9	0.0318
log(initial) + Soil + Nutr + W	13	357.3	0.0158
log(initial) + Soil + Nutr + W + Soil:log(initial)	15	358.3	0.0096
log(initial) + W	6	358.6	0.0082
log(initial) + Soil + Nutr + W + Soil:log(initial) + Nutr:log(initial)	20	363.4	0.0007
log(initial)	3	363.6	0.0007
log(initial) + Nutr	8	371.7	0.0000
log(initial) * Nutr	13	374.4	0.0000

Table 4. AIC and AIC weights for models of change in number of leaves for cuttings.

Models for change in the number of leaves for seedlings	df	AIC	Akaike weight
Initial + Soil + W	11	366.7	0.6024
Initial + Soil + Nutr + W	16	369.6	0.1413
Initial + Soil	8	369.7	0.1344
Initial + Soil + Nutr + W + Soil:log(initial) + Nutr:log(initial)	26	370.2	0.1047
Initial * Soil	13	375.1	0.0090
Initial + Soil	21	375.3	0.0082
Initial * Nutr	13	387.0	0.0000
Initial + W	6	389.0	0.0000
Initial	3	392.1	0.0000
Initial + Nutr	8	396.8	0.0000

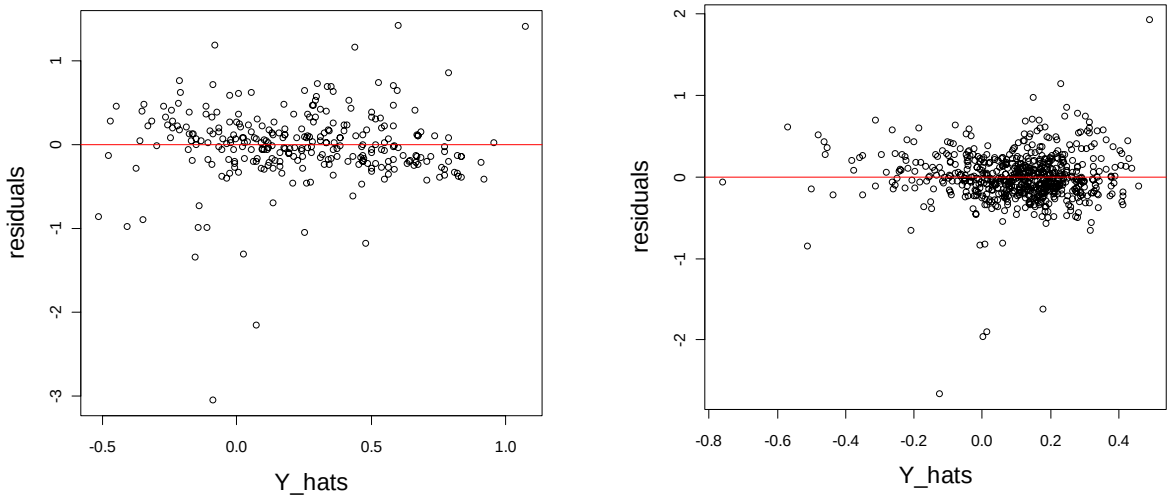

Fig. 2. Distribution of the residuals for the best models of change in number of leaves of seedlings (left) and cuttings (right).

For both seedlings and cuttings, growth in number of leaves varied with initial number of leaves entering the treatment period in the crossover design (Tables 5 & 6). Seedlings and cuttings with large numbers of leaves entering a treatment period tended to have more leaves at the end of the period than plants entering a period with few leaves.

Table 5. Simple contrasts among treatments for change in the number of leaves in seedlings. The reference treatment does not appear in the list. Significant contrasts in bold. Because contrasts were not independent we used Bonferroni $(\alpha' = \alpha / k)$ to obtain an experimentwise error rate of 0.017 for nutrient comparisons and 0.01 for watering treatments and soils.

Coefficients:				
	Estimate	Std.Error	t	Pr(>|t|)
(Intercept)	1.785	0.137	13.07	**2.00E-16**
log(initial)	-0.463	0.040	-11.72	**2.00E-16**
SoilBCRL	-0.013	0.064	-0.20	0.842
SoilBCSJ	-0.204	0.061	-3.35	**0.001**
T0Hal	-0.218	0.073	-3.00	**0.003**
T0Nor	-0.035	0.072	-0.48	0.631
T0Sub	-0.048	0.073	-0.65	0.516

Table 6. Simple contrasts among treatments for change in the number of leaves in cuttings. The reference treatment does not appear in the list. Significant contrasts in bold. Because contrasts were not independent we used Bonferroni $(\alpha' = \alpha / k)$ to obtain an experimentwise error rate of 0.017 for nutrient comparisons and 0.01 for watering treatments and soils.

Coefficients:				
	Estimate	Std.Error	t	Pr(>|t|)
(Intercept)	0.427	0.044	9.712	**2.00E-16**
initial	-0.004	0.000	-12.908	**2.00E-16**
SoilRL	0.175	0.046	3.783	**0.0002**
SoilRLBC	0.209	0.045	4.670	**0.0000**
SoilSJ	0.011	0.042	0.263	0.7929
SoilSJBC	0.116	0.044	2.650	**0.0083**
SoilSJRL	0.122	0.044	2.806	**0.0052**
T0Half	-0.060	0.035	-1.691	0.0914
T0Norm	0.013	0.035	0.358	0.7206
T0Sub	-0.074	0.035	-2.098	0.0363

Willow seedlings established on the 50% Blue Cypress:50% St. Johns soil mixture or in the one-half watering treatment had the least growth in leaf number (Figs. 3 & 4). The latter result is consistent with the field competition and island experiments, where seedlings died during the dry season and at the highest elevations above marsh level, where soil moisture was limited. Growth in leaf number of seedlings did not vary significantly among nutrient treatments.

In contrast, growth in leaf number by willow cuttings varied significantly among soil types but not among watering treatments (Table 6; Figs. 5 & 6). Willow cuttings had higher leaf growth in all except pure St. Johns soil, compared to the control. Growth in leaf number of cuttings tended to be lower in the one-half watering treatment than in the control, but the difference was not statistically significant once the Bonferonni correction was applied. As with seedlings, growth in leaf number by cuttings did not vary significantly among nutrient treatments.

An interesting pattern emerges when willow responses are summarized by life-history stages and growth parameters (Table 7). In both seedlings and cuttings, all growth parameters varied significantly with the initial size covariate. This result is not surprising because plants tended not to shrink in diameter or height, or to lose leaves.

In both seedlings and cuttings, all growth parameters responded to soil type (Table 7), which remained the same for each plant throughout the experiment. Nutrients also remained the same for each plant throughout the experiment, but only affected height and crown growth of seedlings. Willow cuttings did not respond to nutrient treatments.

Watering treatments varied from interval to interval due to the cross-over design. All three growth parameters of willow seedlings were affected by watering treatment. In contrast, willow cuttings only responded with changes in leaf growth.

Interactions between the initial covariate and experimental treatments were important for particular growth parameters. In seedlings, growth in height was influenced by interactions between initial height and the soil and nutrient treatments. In cuttings, growth in stem diameter and height were influenced by the interaction between initial height and the soil treatments (Table 7).

Overall, results of the greenhouse experiments conform well to field observations. Willow seedlings are more sensitive to environmental conditions than larger plants (cuttings) and soils tremendously influence growth of all willow stages. Seedlings are especially responsive to water treatments, while cuttings are less affected by any treatment except one-half normal water. Nutrients have less numerous effects on willow growth and responses sometimes vary with initial plant size. Willow seedlings are more responsive than cuttings to nutrient treatments (Table 7).

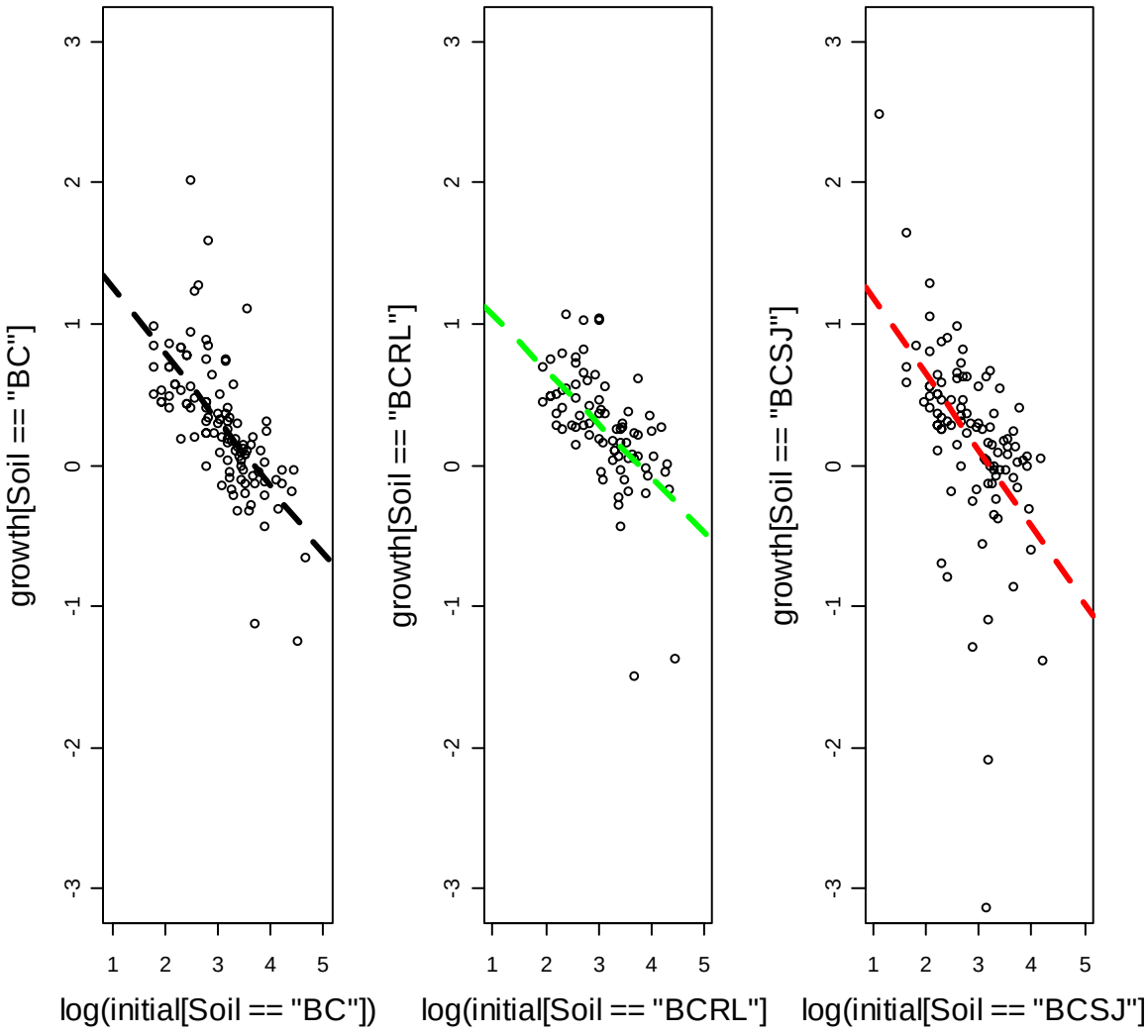

Fig. 3. Change in number of leaves ("growth") by substrate of seedlings.

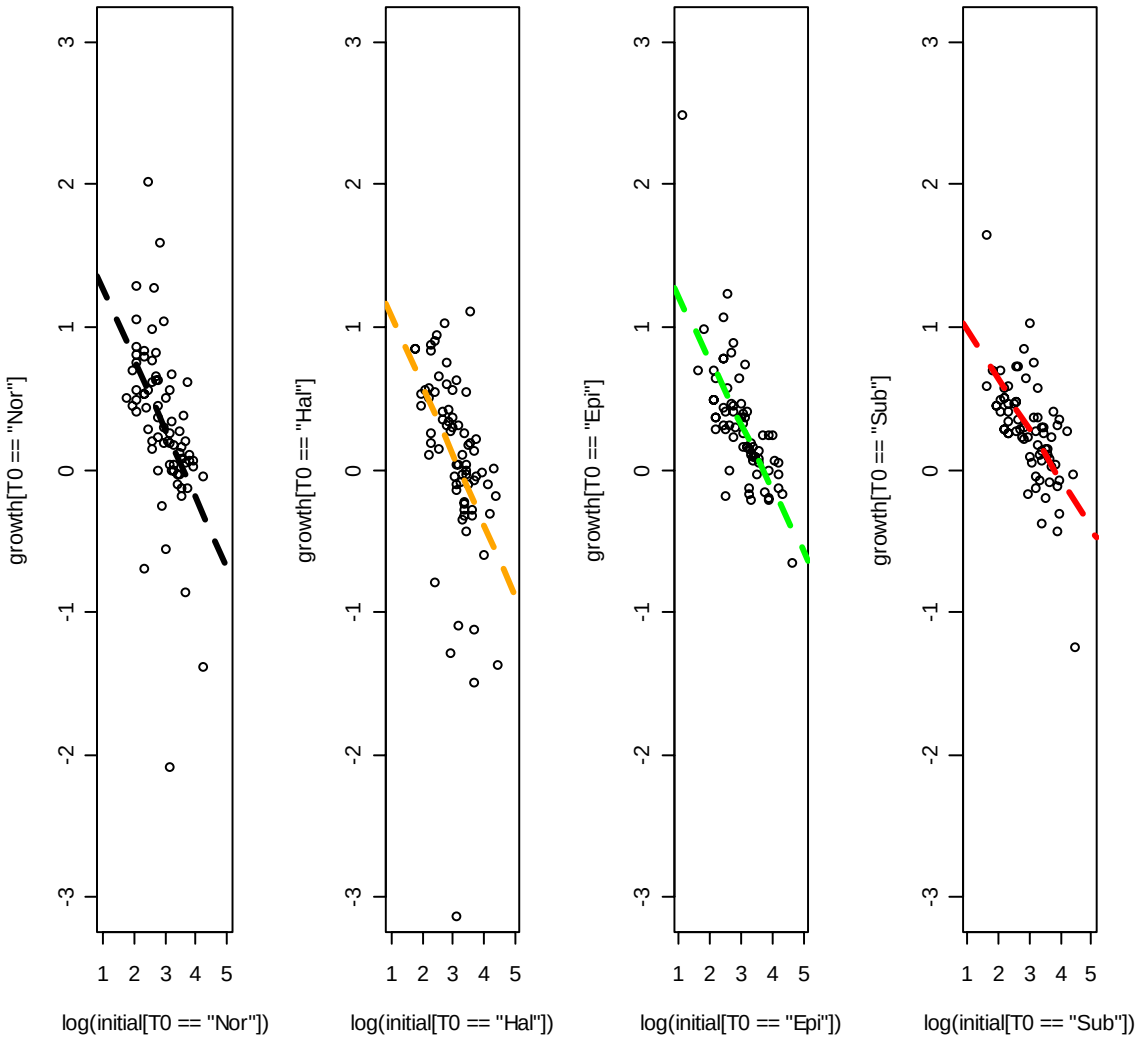

Fig. 4. Change ("growth") in number of leaves of seedlings. Notice lower change in number of leaves for seedlings with half watering compared to submerged and episodic watering.

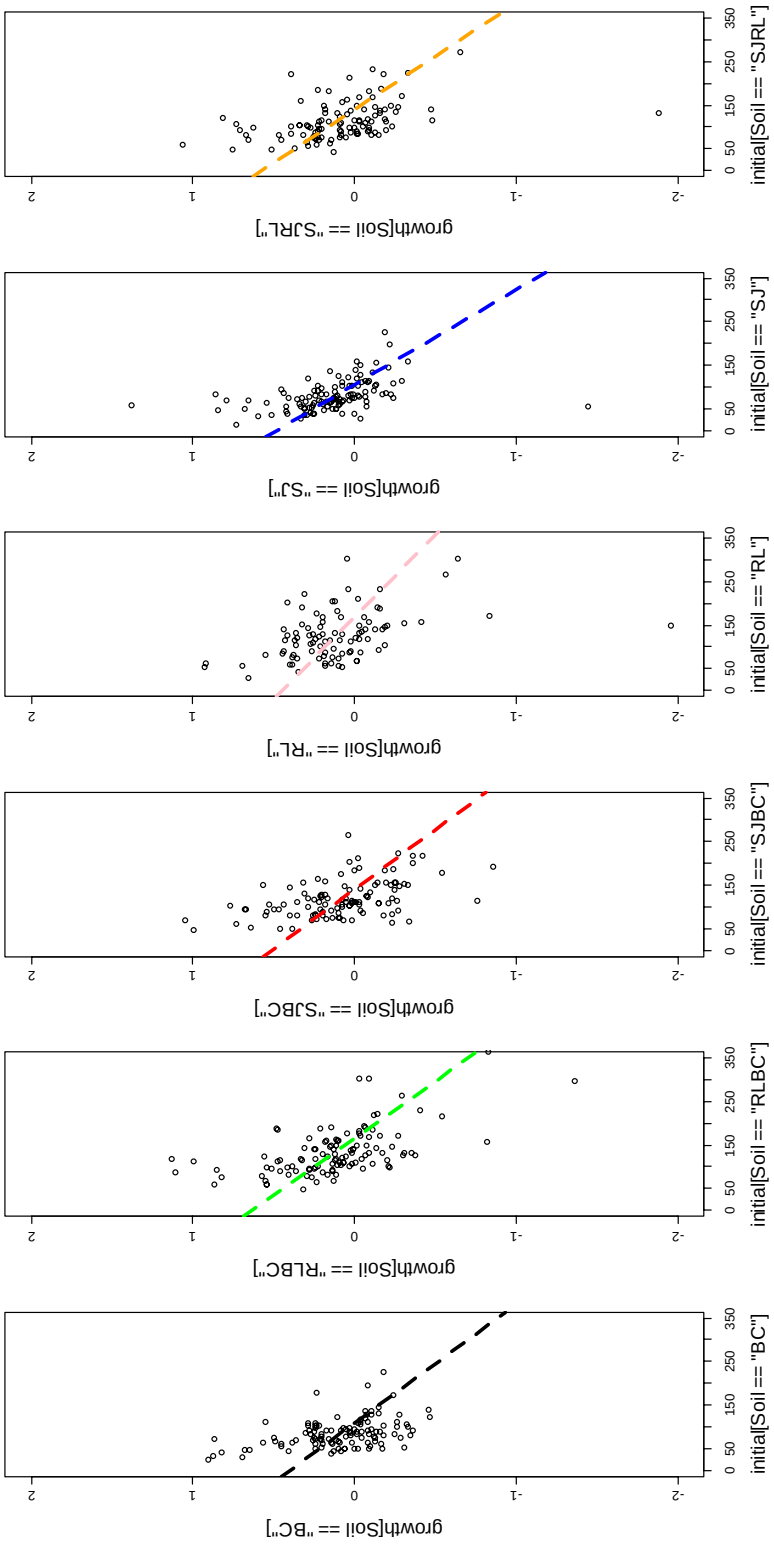

Fig. 5. Change in number of leaves ("growth") by substrate of cuttings.

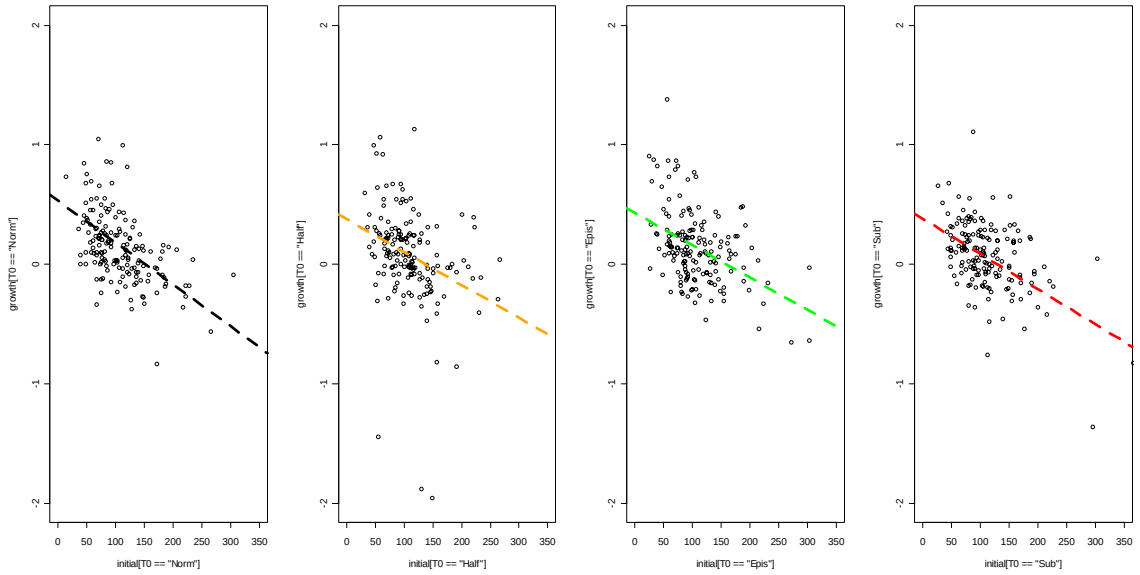

Fig. 6. Change ("growth") in number of leaves. Notice lower change in number of leaves for cuttings with submerged watering compared to control.

Table 7. Summary of effects of treatment or control variables related to the performance of willow seedlings and cuttings in the greenhouse experiments.

Variable analyzed		**Main effects**			**Interactions**	
	Covariable	**Soil**	**Nutrients**	**Watering**	**init:S**	**init:N**
Seedlings						
Height Growth	Log (initial)	√	√	√	√	√
Crown Growth	Log (initial)	√	√	√		
Change in # of Leaves	Log (initial)	√		√		
Cuttings						
Diameter Growth	Initial	√			√	
Stem length Growth	Initial	√			√	
Change in # of Leaves	Initial	√		√		

Progress on Task 2.2 – Willow Transplantation

A. Competition Experiment – We conducted our scheduled monitoring of the competition experiment during this quarter. The water level had subsided compared to October; only a few cuttings remained alive and none showed signs of flowering. We will monitor this experiment again in early February.

B. Hydrology Experiment – We also monitored the hydrology experiment during this quarter. Seedlings and cuttings remained alive on all four islands, which also were colonized by many other marsh plants, including *Juncus*. A few cuttings were quite large and had dropped their leaves, which we suspect may be an energy-conserving strategy to permit reproduction. We will monitor this experiment again in late January or early February and terminate it if any willows show signs of flowering.

Initiate Task 2.3 - Fire response

We designed our sampling strategy for this task and will present it to District staff during our scheduled meeting/field reconnaissance session on January 22, 2010.

Progress on Task 2.4 - Life History

We modified our approach to use a deterministic sampling scheme that incorporates information on soil types, vegetation assemblage, and human influences (e.g., proximity to roads and levees vs. open marsh). We will present our proposed experimental design to District during our meeting/field reconnaissance session on January 22, 2010.

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 completed additional analyses of the greenhouse cutting experiment; decided on designs for the fire experiment and additional experiments on tolerance to flooding and seed viability; and agreed on an improved design for demographic (life history) sampling.