

VEGETATION DYNAMICS IN RESPONSE TO ORGANIC MATTER LOADING RATES IN A CREATED FRESHWATER WETLAND IN SOUTHEASTERN VIRGINIA

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Abstract: Created wetlands are often limited in soil organic matter, a product that usually accumulates with long-term ecosystem succession. Although many studies have tested the effect of adding organic material to these systems, few have quantified the effect of various loadings of organic matter (OM) in created wetlands. The purpose of this study was to determine how vegetation composition, standing crop biomass, and woody vegetation development varied in a created freshwater wetland with respect to different loadings (0, 56, 112, 224, or 336 Mg ha⁻¹) of a soil OM amendment. Soil C, N, and P were positively related to loading rate, as was soil surface elevation. Species richness, evenness, and diversity measurements, along with the Ellenberg Community Coefficient Similarity Index, suggested an overall similarity of plant assemblages regardless of loading rate. Standing crop biomass (580–790 g m⁻²) was not significantly correlated with OM loadings, but showed a significant curvilinear relationship with plot surface elevation. Woody vegetation development was correlated with OM loadings, plot elevation, and soil P, indicating a positive relationship with all three factors. An amendment loading of 112 Mg ha⁻¹ provided the maximum benefit because it provided soil nutrient levels that were within the range of natural wetlands while also minimizing changes in soil surface elevation due to the added bulk material.

Key Words: created wetlands, organic amendments, soil organic matter, wetland vegetation

INTRODUCTION

Permits to drain or fill wetlands granted under section 404 of the 1977 Clean Water Act are, as a final option, accompanied by compensatory mitigation requirements designed to replace the functions [i.e., ecosystem services (*sensu* Odum 1978)] once performed by the disturbed wetland. Therefore, the primary goal of such mitigation alternatives is to achieve functional equivalency with natural wetlands. The failure of created wetlands to achieve this goal is primarily due to a lack of proper hydrology (Mitsch and Gosselink 2000), but also can be attributed to improper geomorphic setting, lack of microtopography, extreme soil compaction (i.e., high bulk density), improper soil biogeochem-

ical conditions, and low soil organic matter (Whittecarr and Daniels 1999).

Variable but low amounts of soil organic matter have been reported for created wetlands in Pennsylvania [0.5%–0.9% (Stauffer and Brooks 1997), 2.3%–6.5% (Cole et al. 2001), 4% (Brooks et al. 2005)], Florida [2.6 ± 0.3% (Anderson and Cowell 2004)], and Virginia [0.5%–1.1% (Whittecarr and Daniels 1999), 3.5%–7.2% (Bruland and Richardson 2004)]. These values were low when compared to organic matter levels in natural reference wetlands [21.7 ± 20.4% in Pennsylvania (Bischel-Machung et al. 1996), 13.8% in Florida (Stauffer and Brooks 1997), and 2.4%–11% in Virginia (Whittecarr and Daniels 1999)].

Organic matter (OM) generally accumulates in ecosystems as succession proceeds (Odum 1969),

leading many studies to advocate amending wetland creation sites with organic material, either in the form of salvaged natural wetland soils or mulches, to help mitigation wetlands achieve functional equivalency more rapidly (Stauffer and Brooks 1997, Whittecar and Daniels 1999, McKinstry and Anderson 2003, Anderson and Cowell 2004, Bruland and Richardson 2004). Studies comparing environmental characteristics in amended and unamended created wetlands are common (Bischel-Machung *et al.* 1996, Brinson and Rheinhardt 1996, Cole and Brooks 2000, Stolt *et al.* 2000, Brooks *et al.* 2005). However, few studies have explored the differences in created wetland ecosystem functions along a gradient of organic carbon loading (Daniels *et al.* 2005).

The purpose of this study was to examine vegetation composition and structure in an early successional created forested wetland along a gradient of soil organic carbon. Our goal was to determine how vegetation composition, standing crop biomass, and woody vegetation development varied with respect to different loadings of a soil OM amendment. We hypothesized that plant communities would show differences in species composition and abundance among loading rates and that plant biomass and tree size would increase with OM loading. From our results we offer recommendations for the amount of an OM amendment that may be most beneficial in created wetland systems.

SITE DESCRIPTION

The Charles City Wetland Mitigation Site (CCW) is a 20.8 ha constructed wetland owned by the Virginia Department of Transportation (VDOT) in Charles City County, Virginia, USA (37°20'37"N, 76°55'33"W) (Figure 1A). The site can be classified as palustrine emergent headwater wetlands (Cowardin *et al.* 1979, DeBerry and Perry 2004), with 18.3 ha designed as forested wetlands. In 1996, the upper soil profile was excavated to a depth of 0.46–0.61 m (Daniels *et al.* 2005). Construction specifications called for the replacement of topsoil; however, there is no evidence that such replacement occurred in the study area. The site is characterized by surface exposure of a silty E or plastic argillic horizon (Btg) that typically exceeds 1 m in depth (Daniels *et al.* 2005). Precipitation is the dominant hydrologic factor in CCW, and fall and winter months are generally accompanied by up to 0.6 m of standing water. In the summer and fall, surface water appears to be perched over the restrictive Btg, particularly following heavy rain events. Following initial



Figure 1. Charles City Wetland Mitigation (CCW) site location, Charles City County, Virginia, USA, with A) indicating Charles City County in Virginia, B) showing the location of the experimental block within CCW, and C) showing the arrangement of plots within the experimental block.

grading, the site was stabilized with a seed mix of *Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot and *Panicum virgatum* L. (DeBerry and Perry 2004).

The study site consisted of a 680 m² area located along the northern edge of the CCW (Figure 1B) that contained twenty 4.6 × 3.1 m plots separated by 3.1 m alleyways. In late June 2002, the site was prepared by removing the existing vegetation with a bush-hog, ripping the soil with a root rake to a depth of 15 cm, and subsequently smoothing by a rototiller pulled by a tractor. In July 2002, each plot received one of five OM loading rates, ranging from 0–336 megagrams per hectare (Mg ha⁻¹) (Table 1), intended to 1) bracket the currently used and recommended rates employed by the wetland design and construction industry, and 2) coincide with rates reported for other disturbed land revegetation studies in Virginia. The plots were arranged in a randomized complete block design (Figure 1C), creating four plot replicates per loading rate. The double rectangular shape of the plot block was a result of fitting the experimental area into a zone containing consistent hydrophytic vegetation before plot preparation in 2002. The experimental design also attempted to block against an observed (no data available) soil moisture gradient from northeast to southwest; however, we found no block × treatment effects for plot elevation (two-way

Table 1. Mean (± 1 SE) surface elevation, soil carbon (C), nitrogen (N), extractable phosphorus (P), and C:N ratios in plots with different organic matter loadings on July 18, 2005 (elevation) or August 22, 2005 (soil nutrients) in CCW, Charles City County, Virginia, USA. Treatment means indicated by different letters denote significant differences based on Wilcoxon Rank Sum Test (C and N) or Tukey's Family Error Rate (Elevation, P, and C:N).

Soil Variable	Loading Rate (Mg ha ⁻¹)					n
	0	56	112	224	336	
Elevation (m)	10.36 \pm 0.00 ^a	10.36 \pm 0.01 ^a	10.38 \pm 0.01 ^a	10.43 \pm 0.01 ^b	10.47 \pm 0.01 ^c	24
Total C content (%)	1.66 \pm 0.28 ^a	3.37 \pm 0.14 ^b	6.66 \pm 1.08 ^c	16.53 \pm 3.06 ^d	15.08 \pm 3.51 ^{cd}	4
Total N content (%)	0.10 \pm 0.01 ^a	0.18 \pm 0.00 ^b	0.29 \pm 0.04 ^c	0.68 \pm 0.09 ^d	0.64 \pm 0.13 ^{cd}	4
Acid Extractable P content (mg kg ⁻¹)	2.8 \pm 0.2 ^a	6.3 \pm 1.3 ^a	8.3 \pm 2.1 ^{ac}	18.3 \pm 2.4 ^b	17.8 \pm 3.1 ^{bc}	4
C:N	17.1 \pm 1.0 ^a	18.3 \pm 0.6 ^{ac}	21.0 \pm 0.8 ^{ad}	239 \pm 1.2 ^{bd}	22.6 \pm 1.4 ^{cd}	4

ANOVA, $F = 0.29$, $df = 4$, $p = 0.876$) or plant biomass ($F = 1.57$, $df = 4$, $p = 0.207$) within the experimental site.

The organic amendment material (OAM) consisted of dry, mixed wood and yard waste compost processed by Grind-All LLC of Richmond, Virginia. The OAM was chosen due to its relative stability, history of use, moderate degree of decomposition, and relatively low total nitrogen content (Daniels et al. 2005). Chemical composition analysis of the OAM is listed in Table 2. Amendments were incorporated into the plots via disking and rototilling by tractor to a depth of approximately 12 cm.

From December 1–15, 2002, five *Betula nigra* and *Quercus palustris* saplings were planted on 1.2 m centers in each experimental plot. (In late September 2005, we discovered that three of the *B. nigra* saplings were actually *B. alleghaniensis*). Each sapling was fertilized with two 16-8-12 controlled

release fertilizer tablets buried within the planting pit near the tree roots. Besides the woody species specifically planted, experimental plots were allowed to re-vegetate naturally (via dispersal or seedbank). Further details regarding the preparation of the experimental plots can be found in Bergschneider (2005) and Daniels et al. (2005).

METHODS

Three years (two full growing seasons) after the OM amendments were incorporated into the soil, we returned to the CCW experimental block to collect the following environmental data from each plot: soil surface elevation, soil nutrients, plant assemblage composition and diversity, standing crop biomass, and woody vegetation development.

Elevation

Relative elevations of each plot were measured on July 18, 2005, using a Topcon AT-G7 Autolevel and a standard stadia rod. Elevation measurements were collected from the center point of each plot and from the base of each *B. nigra* sapling (up to six points per plot). The relative measures were then compared to a known surveyed elevation benchmark and converted to elevations above sea level. The average study site elevation was 10.4 m above sea level, although significant variability existed among plots.

Water table elevations were calculated using water table data from an adjacent, periodically monitored well and plot surface elevation data as previously described. The depth of the water table was measured bi-weekly in one of 30 wells maintained by VDOT at the CCW. Well # 2 data (VDOT, unpublished data) were chosen as the approximate level of the water table in the experimental plots due to the close proximity of the well to the experimental block (~10 m north of the experimental block). Water table elevation was estimated using relative

Table 2. Chemical analysis of organic matter amendment material performed by A and L Laboratories, Inc. Values are reported as means (± 1 SE), with $n = 5$.

Analysis	Chemical Concentration
Solids (%)	54.40 \pm 1.69
TKN (%)	0.86 \pm 0.02
P (%)	0.08 \pm 0.00
K (%)	0.38 \pm 0.01
S (%)	0.11 \pm 0.00
Ca (%)	1.25 \pm 0.03
Mg (%)	0.19 \pm 0.01
Mn (mg kg ⁻¹)	385.20 \pm 9.3
Cu (mg kg ⁻¹)	35.80 \pm 4.2
Zn (mg kg ⁻¹)	97.80 \pm 3.7
Fe (g kg ⁻¹)	6.08 \pm 0.13
Al (g kg ⁻¹)	7.16 \pm 0.15
Organic N (g kg ⁻¹)	8.24 \pm 0.34
Organic C (g kg ⁻¹)	358.97 \pm 22.70
C/N	43.60 \pm 1.12
EC (mS cm ⁻¹)	0.76 \pm 0.11

water table height data from Well # 2 and surface water depth data taken from an adjacent plot (Plot 12, LR 2). Since the plot surface elevation was known, elevation of the water table during ponded periods could be estimated by a regression of water table surface of Plot 12 (i.e., plot surface elevation + depth of surface water) vs. relative water table depth in Well # 2 ($F = 66.03$, $df = 3$, $r^2 = 0.96$, $p = 0.004$).

Soil Nutrients

One sample of the top 10 cm of the soil profile was collected in each plot on August 22, 2005, using a posthole digger. Each sample was thoroughly mixed and analyzed for total carbon (C) and nitrogen (N) content via a controlled combustion Elemental Analyzer (Nelson and Sommers 1996). Due to the assumed lack of carbonates present in the soil at CCW, total C was presumed to approximately equal organic carbon. Dilute double-acid extractable phosphorus (P) was determined by Inductively Coupled Plasma Spectroscopy (Donohue and Heckendorn 1996). All soil nutrient analyses were performed by the Virginia Tech Soil Testing Laboratories.

Plant Assemblage

Herbaceous vegetation was sampled monthly (April–October 2005) in each of the 20 plots. Measurements of vegetation cover were collected from two randomly placed 1 m × 1 m PVC quadrats in each plot. Percent cover per species was visually estimated directly in the field as a value of 1 to 100% or trace (<1%) using a modified Braun-Blanquet cover scale (Daubenmire 1959, DeBerry and Perry 2004) where: < 1% = trace, 1%–5% = 3%, 5%–25% = 15%, 25%–50% = 37.5%, 50%–75% = 62.5%, 75%–95% = 85%, and 95%–100% = 97.5%. Standing dead plant material and bare ground were treated as unique “species.” Plant taxonomy and nomenclature followed the Natural Resources Conservation Service (2006) Plants Database.

Estimates of cover and frequency were converted to relative measures, and Importance Values (IV) were calculated as the sum of relative cover and relative frequency for each species (Mueller-Dombois and Ellenberg 1974, Atkinson *et al.* 1993). The IV of each treatment replicate were averaged over the growing season to calculate IV at each loading rate. Dominant species for each treatment were selected by ranking in order of decreasing IV, with dominants comprising the first 50% of the total and

any additional species greater than 20% (50:20 rule). Species Richness (SR) was determined as the per quadrat (m^{-2}) average number of species for each loading rate during the 2005 growing season. Evenness (J') and the Shannon Diversity Index (H') values (Zar 1984) were calculated for each loading rate using IV and SR data. These values were calculated with standing dead and bare ground removed from the calculations, but including planted tree species *Betula nigra*, *B. alleghaniensis*, and *Quercus palustris*.

The Ellenberg Community Coefficient Similarity Index (SI_E) (Mueller-Dombois and Ellenberg 1974) was used as a measure of plant community similarity among loading rates. This calculation weights species presence and absence between two communities by IV, and is summarized in the following equation:

$$SI_E = (M_c/2)/(M_a + M_b + (M_c/2))$$

where M_c is the sum of IV of species common to both loading rates, M_a is the sum of IV of species unique to loading rate a, and M_b is the sum of IV of species unique to loading rate b.

Weighted Averages (WA) (*sensu* Wentworth *et al.* 1988) were calculated as the product of IV for each species and the indicator index (Region 2, Reed 1988) of that species. Indicator index values ranged from 1 (OBL) to 5 (UPL), with intermediate indicators assigned in between (FACW+ = 1.67, FACW = 2, FACW- = 2.33, FAC+ = 2.67, FAC = 3, FAC- = 3.33, FACU+ = 3.67, FACU = 4, FACU- = 4.33). These replicate values were then averaged over the growing season for each loading rate. This calculation is summarized in the following equation:

$$WA = (y_1u_1 + y_2u_2 + \dots + y_mu_m) / \sum_{i=1}^m y_iu_i$$

where y_1, y_2, \dots, y_m are the relative IV values for each species in a loading rate, and u_1, u_2, \dots, u_m are the indicator values of each species. WA was calculated including the planted tree species.

Standing Crop Biomass

Aboveground standing crop measurements were obtained by clipping all living and standing dead plant material at the soil surface from two randomly placed 0.25 m^2 (0.5 m × 0.5 m) PVC quadrats in each plot. These samples were collected on August 22, 2005, because seasonal plant biomass typically peaks during late summer in southeastern Virginia wetlands (Perry and Atkinson 1997, Perry and

Hershner 1999, DeBerry and Perry 2004). Harvested material was separated by species and dried at 40°C to constant mass.

Woody Vegetation Development

Woody vegetation development characteristics were sampled once on June 21, 2005 for all *B. nigra* saplings within the 20 plots. *Quercus palustris* saplings were omitted from these measurements due to high mortality. Morphometric characteristics measured were total height (using meter tape), crown diameter (using macro-calipers, Haglof, Inc. "Mantax Precision" Calipers), main stem diameter (using micro-calipers, SPI 6"/0.1 mm Poly Dial Calipers), and number of stems. Crown diameter was measured at three different angles at the visual diameter maximum and averaged. Main stem diameter was measured at the base of the main stem (trunk) or just above the visual top of stem base swelling (hypertrophy) that often developed in trees growing in flooded conditions.

Data Analysis

Simple regression (Systat Software, Inc. 2004) was used to explore most relationships between various parameters and loading rate, plot elevation, or soil nutrients. The non-parametric Kruskal-Wallis test (Minitab, Inc. 2005) was used to test for differences among treatments for biomass because the data did not fit a linear/curvilinear model. In this case, Levene's test was used to test for homogeneity of variance (Minitab, Inc. 2005). Tukey's family error rate was used for pair-wise comparisons of elevation and tree development among loading rates, while the non-parametric Wilcoxon Rank Sum Test was used for non-normal data including soil C and N, plant community diversity measurements (SR, J', H') and weighted averages, and biomass (Minitab, Inc. 2005).

Tree morphometrics were analyzed using Principal Components Analysis (PCA), which enabled the data to be analyzed simultaneously, providing a visualization of data structure not available using simple regression techniques. Development of *B. nigra* saplings was quantified using an index represented by the scores on the first Principal Component in the PCA. Multivariate statistics were performed on MATLAB software (The MathWorks, Inc. 2005).

Tests for normality, homogeneity, and pair-wise comparisons were performed using Minitab, release 14 (Minitab, Inc. 2005), while various regressions were performed using SigmaPlot, version 9.0 (Systat Software, Inc. 2004). Unless otherwise indicated,

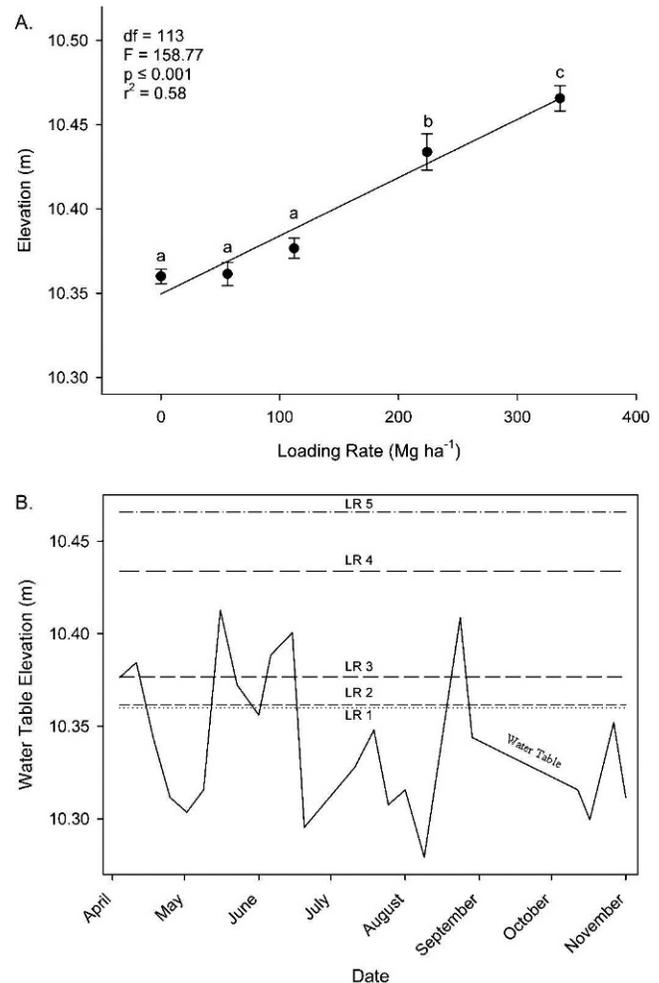


Figure 2. Mean (± 1 SE) elevation vs. A) Loading Rate and B) Water Table Elevation relative to average soil surface elevation for each Loading Rate in CCW, Charles City County, Virginia, USA, from April to October 2005. In A, the regression line is shown, and letters above treatment means denote significant differences based on Tukey's Family Error Rate. In B, water table elevation was based on bi-weekly water table readings in an adjacent monitoring well relative to average plot surface elevation data. Horizontal dashed and dotted reference lines represent average soil surface elevations for the four plots in each loading rate (LR 1 = Loading Rate 1, 0 Mg ha⁻¹; LR 2 = Loading Rate 2, 56 Mg ha⁻¹; etc.).

data are reported as means \pm standard error and statistical significance set at the $\alpha = 0.05$ level.

RESULTS

Elevation

There was a positive relationship between loading rate (LR) and soil surface elevation ($F = 158.77$, $df = 113$, $r^2 = 0.58$, $p \leq 0.001$) (Figure 2A) with a maximum elevation difference of approxi-

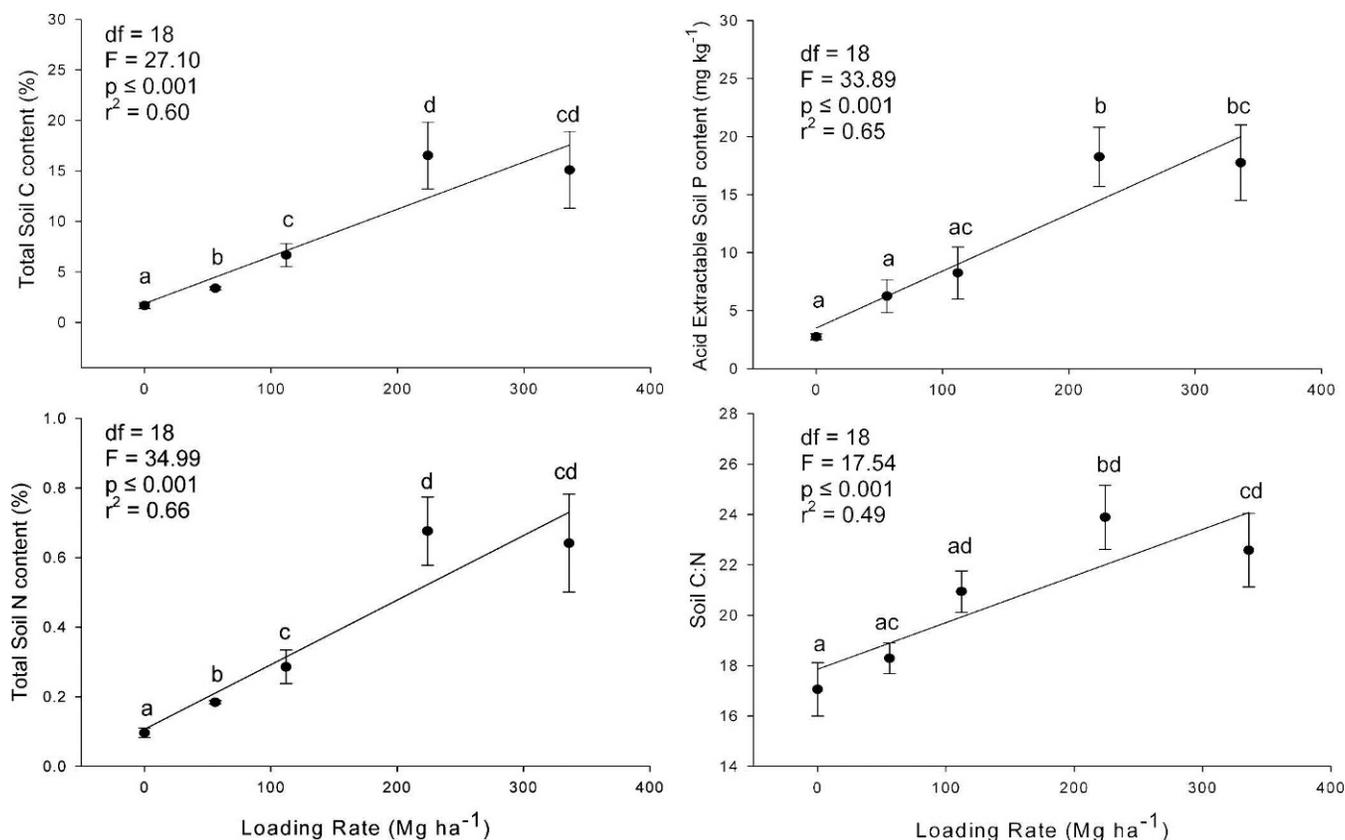


Figure 3. Linear regressions of mean (± 1 SE) soil total-Carbon (C), total-Nitrogen (N), acid extractable Phosphorus (P) content, and C:N ratio vs. Loading Rate in CCW, Charles City County, Virginia, USA, on August 22, 2005. Different letters above the point means denote significant differences based on Wilcoxon Rank Sum Test (C and N) or Tukey's Family Error Rate (P and C:N).

mately 11 cm between LR1 and 5 (Table 1). Pairwise comparison showed that plots in LR1–3 were not significantly different in elevation, while LR4 and 5 were significantly higher than LR1–3 and LR5 was higher than LR4. Differences in plot elevation among loading rates were the result of incomplete incorporation of the Organic Amendment Material (OAM) as noted in Daniels *et al.* (2005), particularly in the higher LR4 (and 5) where conventional tillage (disking and rototilling) could not incorporate the large volumes of mulch applied into the soil. Water table elevation during the 2005 growing season relative to the average surface elevation of plots in each loading rate is presented in Figure 2B. These data suggest that LR1–3 experienced inundated/ponded conditions for at least part of the growing season, whereas LR4 and 5 were not ponded at all during this period, and likely were only saturated following precipitation events. LR1, 2, and 3 were ponded for approximately 25%, 24%, and 17% of the growing season, respectively, with the majority

of inundation events occurring in spring and early summer (April to June).

Soil Nutrients

Soil C, N, and P also increased with loading rate (Figure 3), indicating that soil chemical parameters were affected by and increased with increasing OM loadings. C, N, P and C:N increased with increasing OM from 0–224 Mg ha⁻¹. These chemical concentrations were statistically similar between the highest two OM treatments (224–336 Mg ha⁻¹), likely due to the incomplete incorporation of the OAM at these levels; LR1–3, on the other hand, consisted of relatively uniform mixtures of the OAM and mineral soil.

Plant Assemblage

Sixty-four vascular plant species representing 27 families were collected from the experimental block during the study (Appendix 1). Forty-five species were perennial, 12 annual, six perennial/annual, and

Table 3. Mean (± 1 SE) per quadrat species richness (SR), evenness (J'), Shannon Diversity Index (H'), weighted average (WA), and standing crop biomass among different organic matter loadings over the 2005 growing season (April to October) in CCW, Charles City County, Virginia, USA. Bare ground or standing dead material were not considered. Treatment means indicated by different letters denote significant differences based on Wilcoxon Rank Sum Tests.

Vegetation Variable	Loading Rate (Mg ha^{-1})					n
	0	56	112	224	336	
Species Richness (SR)	7.39 ± 0.45^a	7.75 ± 0.37^a	6.71 ± 0.49^{ab}	7.30 ± 0.40^a	5.29 ± 0.26^b	56
Evenness (J')	0.89 ± 0.01^{ab}	0.92 ± 0.01^a	0.87 ± 0.01^b	0.89 ± 0.01^b	0.86 ± 0.01^b	56
Shannon Index (H')	1.71 ± 0.07^{ab}	1.82 ± 0.05^a	1.54 ± 0.08^{bc}	1.70 ± 0.05^b	1.38 ± 0.05^c	56
Weighted Averages (WA)	1.63 ± 0.03^a	1.57 ± 0.03^a	1.76 ± 0.04^b	2.02 ± 0.05^c	2.12 ± 0.06^c	56
Standing Crop Biomass (g m^{-2})	603.5 ± 179.8^{ab}	580.0 ± 96.9^{ab}	601.5 ± 111.0^{ab}	623.0 ± 193.5^a	789.5 ± 60.2^b	8

one biennial. *Scirpus cyperinus* (FACW+) was the dominant or co-dominant species in each plot (IV range from 18.2–33.3), with standing dead vegetation co-dominant (IV range from 13.8–19.5). Other co-dominants included open unvegetated ground (bare ground) in LRs 1–4 (IV range from 7.8–12.8) and *Eleocharis obtusa* (OBL) in LRs 1 and 2 (IV range from 9.8–10.8). Among LRs 1–3, common subdominant species included *Typha latifolia* (IV range from 3.1–6.0) and *Juncus acuminatus* (IV range from 2.5–4.7). Common subdominant species in LRs 4 and 5 included *Juncus effusus* (IV range from 4.6–7.6) and *Andropogon virginicus* (IV range from 3.3–4.5).

Diversity Measurements. Species richness (SR), Evenness (J'), and Shannon index (H') values were highest in LR 2 and lowest in LR 5 (Table 3), generally decreasing with increasing loading rate. Although regressions were significant ($p \leq 0.01$) and indicated negative relationships of SR, J' , and H' with loading rate, little variation was explained ($r^2 \leq 0.05$).

Community Similarity. The Ellenberg Community Coefficient Similarity Indices (SI_E) were high (>0.5) among all loading rates, ranging from 0.76–0.91. These indices, combined with weak correlations of SR, J' , and H' with loading rate, suggested that species composition of plant assemblages in the experimental treatments were all similar.

Weighted Averages (WA). The WA values ranged from 1.6 ± 0.03 in LR 2 to 2.1 ± 0.1 in LR 5 (Table 3), and showed a positive linear relationship with both loading rate ($F = 100.55$, $df = 278$, $r^2 = 0.27$, $p \leq 0.001$) and surface elevation ($F = 150.17$, $df = 278$, $r^2 = 0.35$, $p \leq 0.001$). The range in WA was narrow, and contained primarily wetland (OBL and FACW) species in all plots (Figure 4). However, the abundance of upland species (FAC through UPL) relative to wetland species (FAC+ through OBL) was influenced by loading rate, as ratios of wetland vs. upland species abundance changed from LR 1 (4.9) to LR 5 (1.2).

Standing Crop Biomass

Peak season biomass estimates ranged from $580 \pm 97 \text{ g m}^{-2}$ in LR 2 to $790 \pm 60 \text{ g m}^{-2}$ in LR 5 (Table 3), but showed neither a significant correlation with loading rate ($F = 1.19$, $df = 38$, $p = 0.283$) nor significant differences among loading rates (Kruskal-Wallis Test, $p = 0.205$). However, a quadratic regression model revealed a significant relationship ($F = 5.64$, $df = 37$, $r^2 = 0.23$, $p = 0.007$) between biomass and soil surface elevation (Figure 5A). These results indicate that peak biomass occurred at an optimum elevation, while values decreased at both higher and lower elevations. Linear regressions were not significant between biomass and any of the soil nutrients measured [N ($F = 0.37$, $df = 38$, $p = 0.547$), P ($F = 0.38$, $df = 38$, $p = 0.539$), C:N ($F = 0.04$, $df = 38$, $p = 0.845$)].

Woody Vegetation Development

Principal Components Analysis revealed similar loadings on the first principal component for total

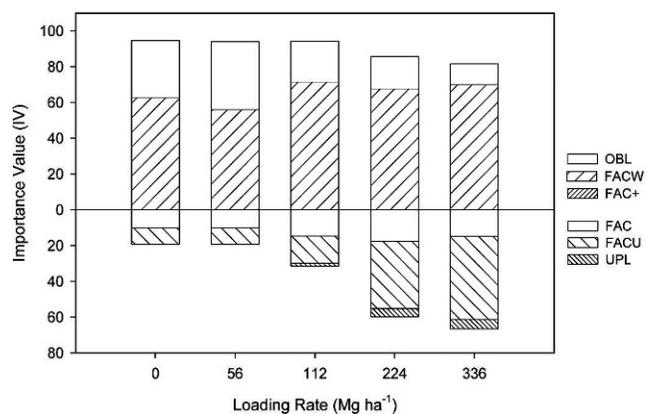


Figure 4. Total importance values of general wetland indicator categories for each loading rate in CCW, Charles City County, Virginia, USA, April to October 2005. FACW and FACU categories include +/- modifiers, while the FAC category includes the - modifier.

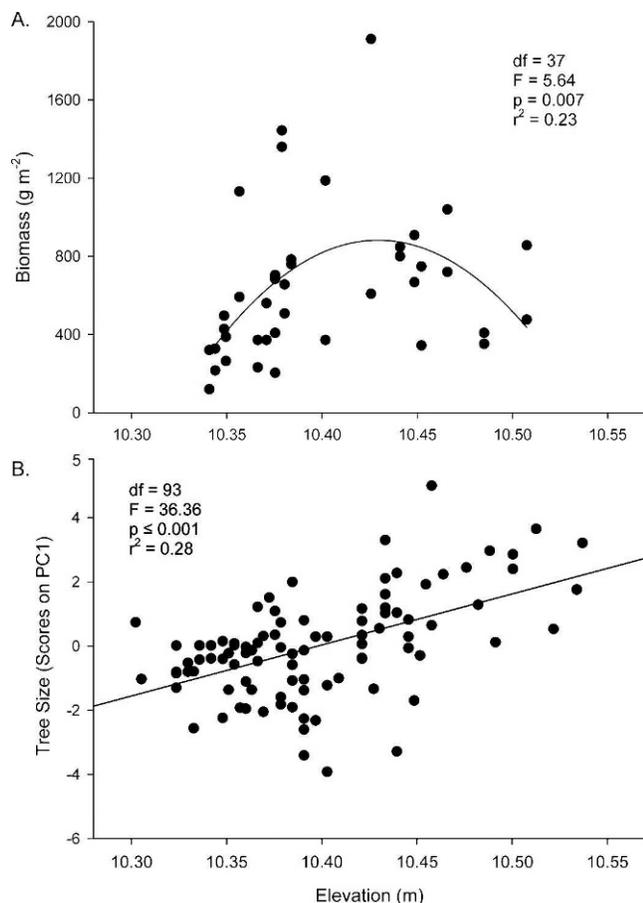


Figure 5. A) Biomass values and B) PC1 Scores (Tree Development) vs. Elevation in CCW, Charles City County, Virginia, USA. Biomass and tree morphometrics were collected on August 22, and June 21, 2005, respectively.

height, crown diameter, and main stem diameter. Due to the similar magnitude of these loadings, as well as the common metric of size (length or width) among these morphometrics, the scores on the first principal component were used as an index of tree "size." Pair-wise comparisons showed significant increases in tree size between LR groups 1–3 and 4–5. Linear regression showed a significant positive relationship of tree size versus loading rate ($F = 24.97$, $df = 93$, $r^2 = 0.21$, $p \leq 0.001$), surface elevation ($F = 36.36$, $df = 93$, $r^2 = 0.28$, $p \leq 0.001$) (Figure 5B), and soil P ($F = 16.77$, $df = 93$, $r^2 = 0.15$, $p \leq 0.001$).

DISCUSSION

Loading Rate Effects on Elevation

Differences in elevation among loading rates reflected the incomplete incorporation of OAM, particularly the higher loading rates (LRs 4 and 5),

into the mineral soil during site preparation, despite three passes with a rototiller (W.L. Daniels pers. com.). This elevation effect was noted in an earlier study of the same experimental block by Daniels *et al.* (2005), suggesting that the OAM in LRs 4 and 5 had not settled to a significant degree in the three years between studies.

Elevation can be used as a proxy for a soil moisture/water saturation gradient, driven by inundation frequency and duration. This assertion is justified by 1) the relatively small aerial extent (680 m²) of the experimental block, 2) the dominance of precipitation-driven hydrology, 3) the determination of CCW as a groundwater recharge site (Despres 2004), and 4) redox potential (E_H) readings that showed higher values in LRs 4–5 than in LRs 1–3 (Daniels *et al.* 2005). These factors lead us to assume that the CCW experimental block as a whole experienced a uniform hydrologic input, and, as such, generally triggered hydrologic inundation more frequently and for longer duration in lower elevation plots than in higher elevation plots; these assumptions were further supported by the water table elevation relative to average plot elevations for each loading rate (Figure 2B). Changes in physical and chemical conditions along hydrologic gradients are well known, and can affect soil O₂ (oxygen) availability (Gambrell and W. H. Patrick 1978, Nedwell 1984, Craft 2001), soil redox potential (E_H) (Ponnamperuma 1972, Vepraskas and Faulkner 2001), and nutrient availability (Bailey *et al.* 1985, Richardson 1985, Bedford *et al.* 1999).

Soil Nutrients

Changes in soil chemical concentrations were expected to be a major driver of the potential differences in plant response among loading rates, especially in relation to standing crop biomass and woody vegetation development. As expected, soil C content, as well as N and P (Figure 3), generally increased with increasing loading rate. Increases in these parameters simply reflected the addition of more OAM, and associated N and P into the soil (Table 2).

The range of soil C found in this study (1%–17%), corresponding to 2.9%–28.4% organic matter (OM = soil C \times 1.72), exceeded the range reported for surface horizons of southern forested wetlands on mineral soils (2.8%–18%) (Lockaby and Walbridge 1998). However, the average OM value of LR 1 (2.9%) was only slightly above the soil surface organic matter concentration of 2% that Baker and Broadfoot (1979) maintained was indicative of

nutritional limitation for many deciduous floodplain trees. Soil total-N levels in our study ranged from 0.2%–0.7% for LRs 2–5, values that were within or slightly higher than those reported for reference wetlands in the Virginia Coastal Plain (0.2%–0.4%) (Whittecar and Daniels 1999). The total-N content of LR 1 (0.1%) was lower than in natural wetlands, and was similar to levels in unamended created wetlands of Virginia (0.03%–0.2%) (Whittecar and Daniels 1999). Available soil P levels (2.8–18 mg kg⁻¹) were within ranges commonly reported for southeastern USA floodplain forest soils (2–18 mg kg⁻¹) (Lockaby and Walbridge 1998). However, LR 1 (2.8 mg kg⁻¹) and LR 2 (6.3 mg kg⁻¹) were below the “crude deficiency level” for available soil P (7 mg kg⁻¹) for hardwood floodplain species as reported by Lockaby and Walbridge (1998).

The low levels of N and P in LR 1 were consistent with the general nutrient deficiency found in other created wetlands (Bischel-Machung et al. 1996, Whittecar and Daniels 1999, Stolt et al. 2000). The increases in nutrient levels associated with increasing loading rates, as well as the impacts of OM loadings on bulk water content and redox potential (Daniels et al. 2005), both suggest that incorporating OM into created wetland soils is important when trying to establish soil biogeochemical environments that are similar to natural wetland systems. However, given the elevation effect observed in this study, it was clear that some of the coarser OAM had not fully decomposed three years following the initial amendments. The confounding factor of hydrological change at higher loading rates should be a consideration when designing such systems.

Plant Assemblage

Vegetation composition in the CCW experimental block was similar to that of other created wetlands in Virginia (Atkinson et al. 1993, Stolt et al. 2000, DeBerry and Perry 2004, Atkinson et al. 2005). We had anticipated that vegetation composition would vary among different organic matter loadings; however, differences were not evident three years after OM amendments were applied. The relatively homogenous vegetative assemblages across the experiment was likely caused by an influx of seed from *Scirpus cyperinus*-dominated plant assemblages surrounding the experimental block (DeBerry 2006).

Scirpus cyperinus dominates many wetlands in Virginia (Yu et al. 1998, Atkinson and Cairns 2001, Atkinson et al. 2005). This species is a biennial that produces large numbers of small (≤ 1 mm), lightweight (1×10^{-5} g) seeds (Shipley and Parent 1991) that are readily transported by wind, water, or

animals (Larson 1999). In addition, this plant can tolerate a wide range of environmental and hydrologic conditions (Kadlec 1958, 1961, Wilcox et al. 1985, Larson 1999).

While *S. cyperinus* dominated all loading rates, *Eleocharis obtusa* cover declined with increasing OM loadings. *Eleocharis obtusa* is a cespitose, obligate wetland annual that occurs commonly throughout the eastern U.S. in “muddy places” (Strausbaugh and Core 1977). However, little ecological information is available for this species, and reasons for its response are unknown. Its high cover in LRs 1 and 2 and decline in abundance in LRs 3–5 (Appendix 1) suggests that relatively large areas (high IV) of bare ground, which also decreased with increasing loading rate, could have encouraged its establishment. It is also possible that *E. obtusa* was able to withstand greater average surface water depths and longer duration of inundation corresponding with the lower elevations of LRs 1–2 (Figure 2B). The evaluation of several growing seasons would be required to differentiate the mechanism behind high *E. obtusa* cover in the lower loading rates.

We hypothesized that plant community diversity would vary with loading rate, and to some extent this occurred. Weak but significant negative relationships with loading rates were observed for species richness (SR), evenness (J'), and diversity (Shannon Index, H') (Table 3). Anderson and Cowell (2004) also reported lower SR and H' values in mulched relative to non-mulched wetlands in Florida. Perhaps nutrient availability at higher loading rates benefits exclusionary, cosmopolitan species, and increased substrate availability at lower loading rates may provide opportunities for species with specialized niches (*sensu* Moore et al. 1989). However, the narrow ranges of diversity values observed over all loading rates and similarity in plant community metrics (i.e., SI_E) indicate that any relationship between loading rate and diversity was minor.

Weighted Average values likely reflected changes in elevation and hydroperiod among plots, rather than differences in loading rate per se. Upland species (FAC through UPL) increased in abundance (IV ranged from 19–67) with increasing loading rate, while importance values of wetland species (hydrophytes, FAC+ through OBL) stayed relatively constant (IV from 82–95). Change in vegetation composition along hydrologic gradients has been routinely studied (Beatty 1984, Messina and Conner 1998, Wall and Darwin 1999, Waddington et al. 2001, Burke et al. 2003, Nicol et al. 2003, Fraser and Karnezis 2005). Vivian-Smith (1997) experimentally demonstrated that elevational differences as minor as 1–3 cm can still affect plant community structure

in wetlands. The 11 cm difference in average elevation between LR1 and LR5 plots likely affected plant species colonization and survival. Over time, however, residual organic matter will break down and elevation differences among plots should decrease. This has the potential to further homogenize plant species.

Standing Crop Biomass

Biomass in study plots (580–790 g m⁻²) was on the low side of the range for natural inland freshwater marshes (500–5500 g m⁻²) (Mitsch and Gosselink 2000) and created wetlands in central Pennsylvania (520–1697 g m⁻²) (Cole *et al.* 2001). However, biomass was generally higher than levels in mulched created wetlands in west-central Florida (349 g m⁻²) (Anderson and Cowell 2004), and for an earlier study at the CCW (146–896 g m⁻²) (DeBerry and Perry 2004).

We expected aboveground standing crop biomass to vary positively with OM loadings, but this was not the case. Our results were similar to others (Cole *et al.* 2001, Anderson and Cowell 2004), who failed to detect differences in biomass between amended and unamended wetlands. We also failed to detect an explicit relationship between biomass and soil nutrients. One reason to add OM to created wetlands is to increase soil fertility, especially by adding N and P (Nair *et al.* 2001) that often limit vegetative productivity (Lockaby and Walbridge 1998). The lack of any biomass-nutrient response in our study was a surprise.

We found that soil surface elevation was a useful predictor of biomass (Figure 5A), and the curvilinear relationship suggests that at an intermediary elevation (~10.45 m) the hydrologic regime produced environmental conditions (i.e., nutrient availability, plant available water, redox potential) optimal for macrophyte production. As for community structure, associations between plant biomass and growth and hydrologic gradients are common (Mitsch and Ewel 1979, Brinson *et al.* 1981, Craft 2001, Craft *et al.* 2002, Fraser and Karnezis 2005). Bayley *et al.* (1985) maintained that sequences of drought and flooding provide a substantial nutrient source to vegetation that is not available in continuously flooded marshes, leading to higher plant production. The higher elevation and high biomass plots in our study may have received sufficient moisture and nutrients from rain and run-off, yet conditions remained aerobic and optimal for growth.

Although the incorporation of OM into created wetland soils certainly has the potential to improve the plant rooting environment, either through

decreasing soil bulk density (Daniels *et al.* 2005) or providing a nutrient source, in the early stages of succession in our created wetlands it appears that soil surface elevation and presumably saturation and inundation were more important. Perhaps, longer periods of time are necessary for coarse OAM to decompose and mineralize nutrients such as N and P into plant available forms. Possibly using a more labile form of OAM could produce a more rapid plant response.

Woody Vegetation Development

Both loading rate and elevation were significant predictors of tree size. The importance of OM amendments and their potential enhancement to soil fertility was supported by the significant positive relationship between tree size and soil P. However, the tree size vs. loading rate relationship may itself highlight the importance of elevation. Pair-wise analysis of tree size vs. loading rate showed a clear and significant separation in tree size between LR groups 1–3 and 4–5 (an elevation difference of 8 cm) but similar size within the two groups. Water table elevations (Figure 2B) suggest that plots in LRs 1–3 occasionally experienced ponded conditions during the 2005 growing season, whereas LRs 4–5 did not. This result may indicate that an elevational transition exists, below which tree growth rates were stunted. *Betula nigra* is a flood-tolerant species and can withstand soil inundation for one to three months during the growing season (Norby and Kozlowski 1983). However, Norby and Kozlowski (1983) showed that dry weights of roots, stems, and leaves of flooded *B. nigra* are reduced compared to unflooded individuals. Many woody wetland species have slower growth rates in flooded bottomland hardwood forests than in wetlands with shorter hydroperiods (Malecki *et al.* 1983, Megonigal *et al.* 1997). In our study, early sapling development probably did not respond to soil OM alone, but instead a combination of elevation-related hydrology and OM-related nutrient gradients better explained tree size differences.

As succession proceeds, organic matter in the higher loading rates may settle due to physical breakdown and decomposition and hydrology will become more similar among the treatments. When this occurs, *B. nigra* sizes may begin to more closely track OM loading rate.

Recommendations

The lack of correlation between plant biomass and organic matter loading rate indicates that

biomass may not always be useful as a functional indicator of created wetland success. Other studies have also reported that soil OM and aboveground biomass were not related (Cole et al. 2001, Anderson and Cowell 2004) or that created wetland aboveground biomass equivalency with adjacent natural wetlands can be achieved even in the early stages of ecosystem development (Whigham et al. 2002, DeBerry and Perry 2004). Organic matter amendments should enhance wetland functionality through increases in soil fertility and moisture holding capacity and decreases in soil bulk density and temperature fluctuation, but plant aboveground biomass may not reflect this enhancement.

Adding an abundance of coarse OAM into created wetlands may inadvertently raise soil surface elevations and decrease the frequency of soil inundation. Elevation change could be minimized by using a less bulky and more labile OAM, although N-overloading could become a concern. We recommend that OM loadings be incorporated into the original soil profile as much as possible, and that loading rates used should not be of a magnitude that alters soil surface elevation. If sites require larger loading rates, it may be necessary to excavate the soil to a greater depth before the amendment is applied.

Because plant response is confounded by elevation, we believe that any recommendation for OM loading in created wetland systems should instead be based on soil nutrient levels. The 112 Mg ha⁻¹ OM loading (in LR 3) is probably appropriate for soils of the Virginia Coastal Plain because average nutrient levels (C = 6.7%, N = 0.3%, P = 8.3 mg kg⁻¹) were within range of natural systems and this rate also minimized change in elevation (+2 cm) from the added material.

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Appendix 1. Plant species and average Importance Values in each loading rate during the 2005 growing season (April to October) in CCW, Charles City County, Virginia, USA. Species names are accompanied by wetland indicator status (Indicator), plant life strategy (Duration), and are separated by loading rate (LR). Nomenclature follows NRCS (2006). Dominant species abundances (by the 50:20 rule) are presented in bold text. Planted species are denoted by *.

Species	Indicator	Duration	LR 1	LR 2	LR 3	LR 4	LR 5
Aceraceae							
<i>Acer rubrum</i> L.	FACW+	Perennial			0.30	0.94	
Alismataceae							
<i>Alisma plantago-aquatica</i> L.	OBL	Perennial				0.17	
Apocynaceae							
<i>Apocynum cannabinum</i> L.	FACU	Perennial					0.39
Asteraceae							
<i>Baccharis halimifolia</i> L.	FACW	Perennial				0.10	
<i>Bidens aristosa</i> (Michx.) Britt.	FACW-	Annual/Biennial	1.18	1.81	2.29	0.52	0.26
<i>Bidens frondosa</i> L.	FACW	Annual	0.83	1.12	0.75	0.06	
<i>Eclipta prostrata</i> (L.) L.	FAC	Annual/Perennial	0.28	0.36	0.63	0.69	0.94
<i>Eupatorium perfoliatum</i> L.	FACW+	Perennial	0.18				
<i>Euthamia graminifolia</i> (L.) Nutt.	FAC	Perennial	0.45	0.07	0.53	0.46	0.41
<i>Solidago canadensis</i> L.	FACU	Perennial	0.09	0.45	0.80	1.51	0.65
<i>Symphotrichum lateriflorum</i> (L.) A.& D. Löve	FAC	Perennial			1.20	0.25	
Betulaceae							
* <i>Betula alleghaniensis</i> Britt.	FAC	Perennial					0.13
* <i>Betula nigra</i> L.	FACW	Perennial	3.10	2.86	3.06	4.28	4.77
Bignoniaceae							
<i>Campsis radicans</i> (L.) Seem. ex Bureau	FAC	Perennial	0.07	0.09	0.09		0.32
Clusiaceae							
<i>Hypericum crux-andreae</i> (L.) Crantz	FACU	Perennial				0.21	
<i>Hypericum hypericoides</i> (L.) Crantz	FACU	Perennial				0.12	
<i>Hypericum mutilum</i> L.	FACW	Annual/Perennial	0.63	0.38		0.42	0.24
Commelinaceae							
<i>Murdannia keisak</i> (Hassk.) Hand.-Maz.	OBL	Perennial	0.99	0.40	0.25		
Cyperaceae							
<i>Carex lurida</i> Wahlenb.	OBL	Perennial		0.10	0.33	0.25	
<i>Carex vulpinoidea</i> Michx.	OBL	Perennial	0.00		0.30	0.47	0.20
<i>Cyperus pseudovegetus</i> Steud.	FACW	Perennial	1.20	0.47	0.59	0.32	
<i>Cyperus strigosus</i> L.	FACW	Perennial	0.19				
<i>Eleocharis obtusa</i> (Willd.) J.A. Schultes	OBL	Annual	9.89	10.79	4.24	1.24	0.65
<i>Rhynchospora capitellata</i> (Michx.) Vahl	OBL	Perennial				0.09	
<i>Rhynchospora corniculata</i> (Lam.) Gray	OBL	Perennial	0.22				
<i>Scirpus cyperinus</i> (L.) Kunth	FACW+	Perennial	20.73	18.17	26.89	27.31	33.30
Euphorbiaceae							
<i>Acalypha rhomboidea</i> Raf.	FACU-	Annual	0.61	0.10	1.33	2.04	3.81
<i>Chamaesyce maculata</i> (L.) Small	FACU-	Annual		0.14	0.11	0.32	0.34
<i>Phyllanthus caroliniensis</i> Walt.	FAC+	Annual			0.10		
Fabaceae							
<i>Lespedeza cuneata</i> (Dum.-Cours.) G. Don	NI	Perennial			0.30	0.12	0.55
Fagaceae							
* <i>Quercus palustris</i> Muenchh.	FACW	Perennial	1.27	1.46	1.41	1.70	2.26
Hamamelidaceae							
<i>Liquidambar styraciflua</i> L.	FAC	Perennial					0.24
Juncaceae							
<i>Juncus acuminatus</i> Michx.	OBL	Perennial	3.78	4.65	2.45	0.79	0.48
<i>Juncus effusus</i> L.	OBL	Perennial	3.54	6.25	6.12	7.57	4.60
<i>Juncus tenuis</i> Willd.	FAC-	Perennial	0.87	0.94	0.76	1.44	0.52
Lamiaceae							
<i>Prunella vulgaris</i> L.	FACU+	Perennial	0.39	0.31	0.46	0.09	
Lythraceae							
<i>Rotala ramosior</i> (L.) Koehne	OBL	Annual	0.20				

Appendix 1. Continued.

Species	Indicator	Duration	LR 1	LR 2	LR 3	LR 4	LR 5
Onagraceae							
<i>Epilobium coloratum</i> Biehler	OBL	Perennial	1.67	1.35	1.46	1.37	1.48
<i>Ludwigia alternifolia</i> L.	FACW+	Perennial	0.53	0.59	1.28	0.80	0.66
<i>Ludwigia glandulosa</i> Walt.	OBL	Perennial	0.63	0.29	0.26	0.19	
<i>Ludwigia palustris</i> (L.) Ell.	OBL	Perennial	2.79	3.90	2.43	1.29	0.57
Oxalidaceae							
<i>Oxalis stricta</i> L.	UPL	Perennial				0.11	0.22
Platanaceae							
<i>Platanus occidentalis</i> L.	FACW-	Perennial					0.30
Poaceae							
<i>Andropogon virginicus</i> L.	FACU	Perennial	0.75	0.87	0.38	3.27	4.45
<i>Arthraxon hispidus</i> (Thunb.) Makino	None	Annual			0.14		
<i>Dichanthelium scoparium</i> (Lam.) Gould	FACW	Perennial	0.20	0.08	0.46	0.00	
<i>Digitaria ischaemum</i> (Schreb.) Schreb. ex Muhl.	UPL	Annual				0.67	0.30
<i>Echinochloa muricata</i> (Beauv.) Fern.	FACW+	Annual	3.23	2.61	0.99	0.65	0.17
<i>Panicum dichotomiflorum</i> Michx.	FACW-	Annual	0.46	0.66	1.04	0.07	0.22
<i>Saccharum giganteum</i> (Walt.) Pers.	FACW+	Perennial	0.09	0.07	0.21	0.69	0.39
<i>Setaria parviflora</i> (Poir.) Kerguelen	FAC	Perennial	1.62	1.79	1.62	2.73	2.29
Polygonaceae							
<i>Polygonum hydropiperoides</i> Michx.	OBL	Perennial	3.76	4.21	1.86	2.03	1.61
<i>Polygonum lapathifolium</i> L.	FACU+	Annual					0.35
<i>Polygonum pensylvanicum</i> L.	FACW	Annual					0.09
<i>Polygonum persicaria</i> L.	FACW	Annual/Perennial	1.19	0.92	0.43	0.26	0.69
<i>Polygonum punctatum</i> Ell.	OBL	Annual/Perennial	0.15				
<i>Rumex crispus</i> L.	FACU	Perennial	0.41	0.44	0.50	0.40	0.99
Rosaceae							
<i>Rubus argutus</i> Link	FACU	Perennial				1.17	0.12
Rubiaceae							
<i>Diodia virginiana</i> L.	FACW	Annual/Perennial				0.25	
Salicaceae							
<i>Salix nigra</i> Marsh.	FACW+	Perennial	0.32	0.03	0.13	0.52	0.15
Scrophulariaceae							
<i>Agalinis purpurea</i> (L.) Pennell	FACW-	Annual			0.17		
Typhaceae							
<i>Typha latifolia</i> L.	OBL	Perennial	4.24	5.99	3.14	2.48	2.01
Vitaceae							
<i>Parthenocissus quinquefolia</i> (L.) Planch.	FACU	Perennial				0.10	
<i>Vitis rotundifolia</i> Michx.	FAC-	Perennial				0.12	
Non-Categorical							
Standing Dead			14.53	13.75	17.85	19.53	19.25
Bare Ground			12.75	11.53	10.38	7.80	8.58