ABSTRACT

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REFERENCE TIDAL FRESHWATER
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This study examined the seed bank and vegetation of a restored tidal freshwater marsh located in Washington, D.C. and compared it to an older restored marsh, a natural urban marsh, and a natural non-urban marsh. A study examining the effects of a beaver impoundment on the vegetation and edaphic factors in the natural non-urban reference site was also conducted. The number of seedlings, vegetation cover, taxa density, evenness, and diversity of vegetation were compared among sites in these studies.

The restored marshes were more similar to the natural urban wetland than to the natural non-urban wetland with regard to the seed bank and vegetation. Duration of flooding from the beaver impoundment was found to be an important factor affecting vegetation composition at the non-urban reference site. Findings included: urban

restoration projects should likely have urban reference sites; and natural disturbances may have similar influences upon both natural and restored systems.

WETLAND RESTORATION IN URBAN SETTINGS: STUDIES OF VEGETATION AND SEED BANKS IN RESTORED AND REFERENCE TIDAL FRESHWATER MARSHES

By

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Thesis submitted to the Faculty of the Graduate School of the University of Maryland, College Park, in partial fulfillment of the requirements for the degree of Master of Science 2005

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Chapter 1: Introduction

Wetlands are dynamic systems (Hobbs and Harris 2001; Hilderbrand et al. 2005), changing in response to anthropogenic and natural stressors (Foote et al. 1996; Coleman et al. 1998; Kennish 2001; Hudon 2004). Humans have ditched, diked, filled, and drained countless wetland acres, while human induced states (e.g., global warming) and natural processes and organisms (e.g., hurricanes, beavers, alligators, and plants) have also altered wetland ecosystems (Mitsch and Gosselink 2000). Approximately 53% of historic wetlands in the conterminous United States have been lost (Dahl 1990). Given the stronger wetland protection laws in existence (e.g., the Clean Water Act), and the goal of "no net loss" of wetlands established under the first Bush Administration, better preservation and restoration of ecosystems has become relatively common.

Although wetland restoration is a relatively widespread activity, it is a young science, with many early papers and books on the topic published beginning in the early 1990s (Kentula et al. 1992; National Research Council 1992). It soon became evident that many restoration or creation projects did not meet all of their respective goals, usually for soil parameters (Craft et al. 1999; Atkinson and Cairns 2001; Morgan and Short 2002; Craft et al. 2003). Some restoration practitioners and scientists would maintain that these projects were a success because the wetland may have resembled its structural historic form. Others would consider the functions provided to be key (Stanturf et al. 2001; Campbell et al. 2002; Warren et al. 2002). Functional equivalency between restored and natural wetlands has not been established (Kentula 2000). Stanturf et al. (2001) question whether equivalency is possible, at least in bottomland hardwood forests.

The fundamental goal of wetland restoration is the return of a disturbed wetland to its preexisting natural condition, including structural and functional characteristics (National Research Council 1992; Mitsch and Gosselink 2000). Although not a functional component of wetlands, restoration of "natural" vegetation is an objective of most wetland restoration projects; however, restored vegetation frequently does not resemble vegetation of natural systems (Galatowitsch and van der Valk 1996). It is often difficult to restore vegetation in complex systems such as tidal freshwater marshes (Baldwin 2004). Vegetation structural parameters can serve as proxies for functional attributes of systems, e.g., biomass as primary production (USEPA 1990; Thursby et al. 2002; Asaeda 2005).

Given that approximate natural hydrologic conditions exist, restoration of wetland plant communities can be improved with knowledge of the life history of species, existing seed bank, sources of propagules and species dispersal, and the means of growth and survival of plant species (Middleton 1999; Budelsky and Galatowitsch 2000). Many species of plants can survive in the seed bank for long periods, but important species may be missing (Middleton 1999), e.g. species with large floating seeds.

For my research, I continued and enhanced a study of vegetation and seed bank development in Kingman Marsh, a tidal freshwater marsh in Washington, D.C., restored in 2000. This work was done as a cooperative project between the U.S. Geological Survey and the University of Maryland. I compared Kingman Marsh (urban restored) to three tidal freshwater wetlands surrounding the metropolitan Washington, D.C. area (Kenilworth Marsh, urban and restored in 1992-93; Dueling Creek, a natural urban wetland; and Patuxent Marsh, a natural non-urban wetland in an adjacent watershed).

Natural, as defined in this study, meant that the wetland developed independently of direct human intervention, while restored sites received sediment input and vegetation plantings. Urban areas included those wetlands within the Anacostia watershed, which is 65% urban, while the Patuxent watershed, at 21% urban, was considered non-urban. Relative to each other, the Anacostia watershed is the more urban watershed.

The seed bank and dispersal routes of plant species were studied previously at Kingman, along with the vegetation, soils and hydrologic regimes of all four marshes (Neff 2002). My work continued the monitoring effort at these sites through the five year mark (2004). The first two years (2000 and 2001) of research revealed that although Kingman Marsh's seed bank contained a diverse assemblage of species, some important annuals, e.g., *Polygonum arifolium* and *Impatiens capensis*, were missing or occurred at low abundance in Kingman Marsh's vegetation cover but were plentiful in Patuxent Marsh (Neff 2002). A tidal freshwater marsh study conducted at Kenilworth Marsh (restored) showed that the vegetation there contained a significantly lower number and relative cover of annual species than reference sites (Baldwin and Derico 1999). Similarly, Leck (2003) found perennial species to be more important than annuals in the vegetation of a created tidal freshwater wetland in New Jersey.

At least two important questions emerge based on the previous studies. First, why does the restored Kingman Marsh not contain a vegetation community or seed bank similar to a natural marsh such as Patuxent Marsh? And second, is Patuxent Marsh, a natural non-urban marsh, an appropriate reference site for the urban Kingman Marsh, or is Dueling Creek, a natural urban wetland, a more appropriate reference? Many authors have cautioned that restoration projects in general will not come to resemble natural

systems (National Research Council 1992; Ehrenfeld 2000a; Warren et al. 2002), especially restorations in urban areas (Brinson and Reinhardt 1996; Grayson et al. 1999). The approach in choosing restoration goals should, perhaps, be altered for systems depending on their level of degradation and landscape position.

Natural system disturbances must also be taken into account during the restoration or management of any wetland system. Kingman Marsh experienced intense goose herbivory shortly after planting (Hammerschlag et al. 2000; 2001; 2002; 2003; Neff 2002). Patuxent Marsh, the natural non-urban wetland, also experienced herbivory, likely by muskrat and/or beaver, both species of which I found evidence of on site (a beaver dam and multiple muskrat mounds) (personal observation). Other herbivores at Patuxent may include resident Canada geese and carp (Baldwin and Pendleton 2003). Patuxent Marsh also experienced a more intense disturbance when the beaver constructed their dam across a creek draining the wetland system and created a large impoundment where there had previously been tidal fluctuations.

Goal and objectives

The goal of this project was to continue long-term monitoring (2000-2004) of the seed bank and vegetation at a recently restored marsh (Kingman Marsh) during years 2002-2004 and to compare its development to an older restored marsh (Kenilworth Marsh) and to two natural reference wetlands (Dueling Creek and Patuxent Marsh). An additional study in this project grew out of the monitoring conducted at the natural, non-urban reference site, Patuxent Marsh. While sampling there in May 2003, I found a beaver dam blocking Mill Creek (a tidal channel of the Patuxent River), the creek that drains this portion of the tidal freshwater wetland. Upon further investigation, I noticed

that the composition of the vegetation was clearly changing, likely in response to the impoundment created by the dam. This relatively rapid change in vegetation fascinated me and I decided to carry out a study to see if the impounded areas possessed significant differences in parameters (e.g., vegetation: cover, biomass, leaf area index; soils: percent moisture, bulk density, organic matter) than the non-impounded areas. This study provided an opportunity to examine a natural disturbance and observe how the marsh responded. It also allowed study of the variability associated with "natural" reference sites and how this variability might affect determinations of success in restored wetlands.

The specific objectives of my research were:

Objective 1. Seed banks

Characterize the seed banks (density of seedlings, taxa density, evenness, and annual and native species composition) of Kingman Marsh, Kenilworth Marsh, Patuxent Marsh, and Dueling Creek in year three (2003) of the monitoring study, compare changes from year one (2000) and year two (2001) collections, and determine the effect of inundation on germination.

Rationale for Objective 1

- Seed bank studies conducted prior to planting detect species that may become established in a system and those not present in vegetation, especially annuals
 - Allows comparison of restored and natural sites with regard to native vs. nonnative seedlings; annual vs. perennial seedlings; taxa density; abundance of seedlings; diversity; and evenness

- May allow for decisions about which species to plant/replant
- Demonstrates how the seed bank develops temporally at a new restoration site
- Early detection of invasive non-native species
- Flooded and non-flooded treatments can show which species can germinate and survive in extended inundation and which cannot

Objective 2. Vegetation

Characterize the vegetation of Kingman Marsh, Kenilworth Marsh, Patuxent Marsh, and Dueling Creek. Use vegetation parameters (cover, taxa density, diversity, evenness, annuals vs. perennials, natives vs. non-natives, and species composition) to examine the suitability of a natural non-urban marsh as a reference site for an urban restored marsh.

Rationale for Objective 2

- Allows comparison of vegetation characteristics in restored and natural marshes based on percent cover; taxa density; evenness; diversity; annuals vs. perennials; and natives vs. non-natives
- By comparing older and younger restored marshes to natural marshes in urban and non-urban landscapes, I can explore the utility of natural urban and non-urban reference sites in evaluating the success of wetland restoration in urban settings

Objective 3. Beaver dam

Describe the vegetation (cover, biomass, height, leaf area index, canopy cover, species composition, taxa density, evenness, and diversity), physical water quality

(conductivity, salinity, temperature, and depth), and soil characteristics (bulk density, percent moisture, organic matter, and redox potential) of the impounded and non-impounded zones of Patuxent Marsh that may have resulted from a beaver dam constructed in the wetland in mid-2002.

Rationale for Objective 3

- Allows us to study natural disturbance in tidal freshwater marshes
- Assists in quantifying the effect of flooding on vegetation
- Quantifies variability of natural non-urban sites due to hydrology and animal disturbance, two common factors controlling vegetation development in urban wetland restoration projects

Chapter 2: Rapid Development of a High Density Seed Bank in a Restored Tidal Freshwater Wetland

This chapter is the update of a manuscript proposed by K.P. Neff and A.H. Baldwin. Neff collected data during 2000 and 2001. Kelly Neff, Andrew Baldwin, and I decided to expand the manuscript to include my 2003 experiment and to show additional trends in seed bank variability (based on comments from external anonymous reviews of the original manuscript). My contribution was to re-analyze all data to include 2003 findings, and to revise, edit, and update the text. This chapter, in a condensed form, will be submitted by all three authors to a peer reviewed journal.

Introduction

The seed bank (i.e., buried viable seeds and propagules) is an important component of wetland plant community dynamics (van der Valk 1981; Leck 1989), particularly in tidal freshwater marshes (Leck and Simpson 1995; Baldwin and Derico 1999). Standing vegetation composition in tidal freshwater marshes, including vegetation reproduction, is strongly influenced by the seed availability at the site (Parker and Leck 1985; Odum 1988; Leck 1989), with different species having different degrees of dependence on seed (Leck and Simpson 1987; 1995). The seed bank is determined by the production of seed from past and current vegetation and the seed longevity in the site (van der Valk and Pederson 1989), and develops over decades (Harper 1977).

Constructed wetlands often start with poor seed banks (Smith and Kadlec 1985; Reinartz and Warne 1993). Seed bank richness in some restored wetland systems has been reported to be lower than in reference wetlands (Galatowitsch and van der Valk

1996). Leck (2003) reported seed density and seed bank richness to be low the first year in a created tidal wetland and to quickly increase, with prolific seed dispersal implicated as a possible cause. After only a short amount of time, approximately three and a half years, seed density and richness were found to be higher in restored sites than reference sites of tidal freshwater wetlands (Baldwin and Derico 1999), which are sites that have high surface water connectivity to local floating seed sources (Middleton 1999). Due to tidal fluctuations, and the seeds carried in water, tidal wetlands have an advantage over other wetland types in seed influx.

The ability of a site to revegetate after a disturbance should be one function evaluated when determining the success of a restored wetland (Middleton 1999). For this reason, comparisons of seed banks of restored wetlands to reference wetlands may be a good measure of restoration success (Baldwin and Derico 1999). In addition, examining the seed bank for presence of invasive or undesirable species allows predictions of the susceptibility of the restored site to an invasion (Pederson and van der Valk 1984; Brown 1998).

To understand the initial development of seed banks after tidal wetland restoration, we examined changes in the seed bank of a recently restored wetland, Kingman Marsh, immediately after placement of sediment and then the first and third years after restoration. We compared these seed banks with those of an older restored wetland, and two reference wetlands. We hypothesized that the seed bank of the recently restored site would develop quickly, similar to that reported for other restoration and creation sites (Baldwin and Derico 1999; Leck 2003). In 2003, we also studied the effect of flooding on seedling emergence from seed banks and hypothesized that inundation

would have a negative effect on a majority of species, based on previous work (Baldwin and Derico 1999; Baldwin et al. 2001; Wetzel et al. 2001; Peterson and Baldwin 2004). Through a quantitative evaluation of seed bank development, we also hoped to explore the utility of seed bank studies in evaluating the success of restored or constructed wetlands for determining the types and quantities of plantings that might be needed.

Methods

Study sites

In early 2000, the U.S. Army Corps of Engineers restored 13 hectares of tidal freshwater wetlands at Kingman Marsh (N38°54'17", W76°57'42"), located in the urbanized Anacostia watershed in Washington, D.C. (Figure 2.1). The restoration was implemented through dredging and filling of containment cells with Anacostia River sediment to elevations sufficient to support emergent macrophytes. After a period of dewatering, this process was followed by planting of native species: *Juncus effusus*, *Nuphar lutea*, *Peltandra virginica*, *Pontederia cordata*, *Sagittaria latifolia*, *Schoenoplectus pungens*, and *Schoenoplectus tabernaemontani*. We collected our first set of seed bank samples after sediment placement but before planting, with the objective of describing the abundance of seeds present in the placed dredge material.

To provide a context for evaluating results from Kingman Marsh, we also studied seed banks at an older restored urban wetland (Kenilworth Marsh) and two natural wetlands, one urban (Dueling Creek) and one non-urban (Patuxent Marsh). Kenilworth Marsh (N38°54′50″, W76°56′40″), located about 0.8 km upstream of Kingman Marsh along the Anacostia River in Washington, D.C., was restored in 1992-1993 (seven years

The Chesapeake Bay and its Major Tributaries



Figure 2.1 Regional map indicating the Anacostia River and Washington, D.C., and the Patuxent River, a tributary of the Chesapeake Bay.

prior to Kingman Marsh) using dredge material to raise elevations to levels suitable for wetland vegetation (Bowers 1995). Kingman and Kenilworth marshes historically were part of a large expanse of tidal freshwater marshes located throughout the Anacostia watershed system (Syphax and Hammerschlag 1995). However, in the 1920s and 1930s they were removed by dredging to create lakes for recreational purposes for city residents, for channelization, and for disease management (A. Baldwin, University of Maryland, personal communication; USEPA 2002). The lakes proved to be poor recreational areas and habitat due to the shallow unvegetated mudflats, apparent at low tide (USEPA 2002; Neff 2002) and both were restored as mentioned.

The urban natural site, Dueling Creek, (N38°55′28″, W76°56′29″) is a remnant 0.41-hectare tidal freshwater marsh located about 0.8 km upstream from Kenilworth Marsh along Dueling Creek, a small tributary to the Anacostia River. It is one of the few remaining (unaltered) sections of the Anacostia marsh system and has been suggested as a potential reference site for nearby tidal restorations (Biohabitats 1990), and has been used as a reference site in past studies (Baldwin and Derico 1999; Hammerschlag et al. 2000; 2001; 2002; 2003; Kassner 2001; Neff 2002). Patuxent Marsh (N38°48′38″, W76°42′37″) is a relatively undisturbed tidal freshwater marsh located in the adjacent Patuxent River watershed 22.5 km southeast of the other sites along the Patuxent River.

Seed bank sampling

As part of a broader monitoring effort, a total of 35 vegetation monitoring transects, each 35 m long, were randomly located within the wetlands (18 at Kingman Marsh; 8 at Kenilworth Marsh; 3 at Dueling Creek; 6 at Patuxent Marsh) (see Chapter 3).

Transects were divided into seven 5 m long sectors, and a 4.8 cm diameter by 5 cm deep core of soil (surface area = 18.1 cm^2 , volume = 90.4 cm^3) was taken for seed bank assay at each of the five middle sectors. Samples were taken from the first third of these 5 m sectors in May 2000, the middle third in March 2001, and the last third in March 2003 to avoid sampling the exact same locations. Samples from each time period were combined for the entire transect to form a composite sample of approximately 450 cm³. The later date in 2000 (May) was due to collection just following the placement of dredge material at Kingman Marsh. All sampling dates occurred after the period of natural cold stratification.

Samples were stored at 4°C until processing. Coarse organic matter and vegetative parts (e.g., living rhizomes) were removed from the soil, and each sample thoroughly mixed by hand. A random sub-sample of one half of each soil sample (by weight) was spread in an approximately 1.3 cm thick layer on top of 3.5 cm of moist vermiculite in aluminum pans (15.6 cm wide by 21.8 cm long by 5.1 cm deep) with perforations on the bottom to allow drainage. These were randomly placed on a greenhouse misting bench at the University of Maryland. Samples from 2000 were put in the greenhouse in mid-May 2000, samples from 2001 were put in the greenhouse in early April 2001, and samples from 2003 were placed in the greenhouse in mid-March 2003. Sediment in the pans remained moist but not inundated. The 2003 experiment included a flooded treatment to determine flooding effects on seedling emergence. In 2003, a random sub-sample of one half (by weight) of each sample was used in non-flooded pans and the other half in flooded pans. Flooded pans were prepared by spreading samples in a 1-1.5 cm thickness along the unperforated bottom of pans identical in size to those used for non-flooded

samples. The pans were kept inundated, but to prevent seeds from washing out of the flooded pans, eight small perforations were punched 5-6 mm from the top edge to allow excess water to purge. Six control pans lacking soil and seed were used in 2003, three for flooded samples and three for non-flooded samples. Non-flooded control pans contained vermiculite. The control pans allowed us to detect if plants were seeding and depositing seed into adjacent pans. An incident occurred in 2003 where greenhouse ceiling glass was broken. This disturbed several pans and may have led to cross-contamination of pans through splashing, or seeds may have entered the greenhouse through the open ventilation system, causing cross-contamination. Several control pans in 2001 and 2003 showed germination of seedlings (Appendix B), but not sufficient amounts to include in analyses. *Rorippa palustris* was a prevalent greenhouse weed found in a couple of control pans. In 2003, control 2 non-flooded and control 3 non-flooded pans were surrounded by pans containing soils from Kingman and Kenilworth marshes and may have been cross-contaminated by them during the previously mentioned greenhouse ceiling glass episode.

Emerging seedlings were identified and counted for seven months in 2000 and 2001, and for nine months in 2003. Unknown species were transplanted and grown until they could be identified. This emergence method gives an acceptable estimate of species composition of viable buried wetland seeds (Poiani and Johnson 1988) and is commonly used in wetland seed bank studies (van der Valk and Davis 1978; Leck and Graveline 1979; Leck and Simpson 1994; 1995; Baldwin et al. 1996; Le Page and Keddy 1998; Baldwin and Derico 1999). The USDA PLANTS Database Version 3.5 (2005) and Brown and Brown (1984; 1992) were consulted for plant taxonomy. For consistency, the USDA PLANTS Database Version 3.5 was used for nomenclature, life history, and

native or non-native classification, realizing that some species may not historically be native to certain geographic regions.

Data analysis

Total seedling emergence (number of seedlings emerging/pan), taxa density (number of species/sample), and Pielou's J (a measure of evenness) were analyzed with a repeated measures ANOVA (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Mixed) to detect differences in these parameters across sites and years (example SAS code presented in Appendix C). A critical level of P = 0.05 was used in assigning significance. We ran the Tukey-Kramer test to make pairwise mean comparisons on these parameters. Total seedling emergence was log transformed $\left[\log_{10} (x + 1)\right]$ to meet the assumptions of ANOVA; all other parameters met the assumptions. Means were detransformed for presentation. Unequal upper and lower standard errors were computed by adding or subtracting, respectively, the transformed standard error to the mean and detransforming. Least squares (LS) means were calculated for comparisons between sites, but arithmetic means were presented in tables because LS means estimate values even when they are zero, i.e., nothing is present in a sample. The calculation of least squares means often results in a small negative number which is impossible for some parameters, e.g., taxa density of a sample.

The evenness index, Pielou's J, was calculated as: J = H'/log S (McCune and Grace 2002). The closer J is to one, the more even the distribution of abundance across species. H' is the Shannon-Wiener diversity measure, calculated as: $H' = -\Sigma (p_i \log p_i)$ where p_i is the percentage importance based on relative density and S is the species

richness per transect (Peet 1974; McCune and Grace 2002). Sørensen's Quotient of Similarity (qs) was used to examine the similarity in seed bank species composition between the four sites: qs=2c/(a+b); c = number of species occurring in common at both sites; a = total number of species found at site a; and b = total number of species occurring at site b (Sørenson 1948). The closer qs is to 1, the more similar the two sites.

The total number of annuals and natives was determined for each site in each year. Phragmites australis was assumed to be the non-native genotype for classification purposes. The Chi-square test of independence (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Freq) was then used to determine if the composition of annuals vs. perennials and natives vs. non-natives found at each site was independent of site. Species classified as annual/biennial or annual/perennial in the USDA PLANTS Database Version 3.5 (2005) were included with the annuals. Fisher's Exact Test was used for 2000 data because the sample size was potentially too small for a valid Chi-square test. Plants identified only to family or genus were not included in Sørensen's Quotient of Similarity or Chi-square calculations. In addition, because additional vegetation transects were added, the number of samples collected in 2001 increased at two sites (i.e., Kingman Marsh increased from 12 to 18 samples and Patuxent Marsh from five to six samples). Dominant species in 2003 were determined based on the arithmetic mean number of seedlings per pan. Unidentified dicots and monocots were not included in the dominant species list. Flooded and non-flooded means from the 2003 samples were compared using two-sample t-tests (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc ttest). Means were presented, arithmetically, along with p-values for plant species, seedling density, germination, evenness, and taxa density.

Results

The number of seedlings, taxa density, and species evenness all varied significantly between sites and years, and site x year interactions were all significant (Table 2.1; Figure 2.2). In 2000, Kingman Marsh had lower seedling emergence (approximately three seedlings/pan) than the other urban sites, with values being significantly lower at Kingman Marsh than at Kenilworth Marsh (Figure 2.2). Between 2000 and 2003, seedling number increased significantly and by more than 44 times at Kingman Marsh to 132 seedlings/pan. In 2003, Kingman Marsh's seedling numbers fell between Kenilworth Marsh (the older restored site) and Dueling Creek (the natural urban site) (Figure 2.2).

In 2000, seed bank taxa density was significantly lower at Kingman Marsh (less than 3 species per sample) than at Kenilworth Marsh (approximately 11 species per sample) (Figure 2.2), while in 2003, 38 species emerged from Kingman Marsh seed bank samples, more than at any other site (Table 2.2). Taxa density at Patuxent Marsh increased significantly from 2000 to 2003, becoming similar to all other sites by 2003. Evenness, the relative abundance of each species in a given area, showed a significant decline from 2000 to 2003 at Kingman Marsh (Figure 2.2). Kingman Marsh's evenness fell from its initial high in 2000 of 0.9, to 0.6 in 2001, and 0.65 in 2003. Overall, Patuxent Marsh maintained the greatest evenness across years, although it experienced a continuous but not significant decline in evenness from 2000 to 2003, which did not occur at the other sites. By 2003, all the sites were similar in their evenness.

Sørensen's similarity index for 2003 suggests that the species composition of Kingman Marsh was most similar to Kenilworth Marsh and least similar to Patuxent

Table 2.1 Results of the analysis of variance examining seedling density (number of seedlings/m²), taxa density (taxa/sample), and evenness (J= H'/log S) among four tidal freshwater marshes for years 2000, 2001, and 2003. Reported are Fvalue (Numerator df, Denominator df). Significance noted as *<0.05; **<0.01; ***<0.001; ****<0.0001.

Parameter	Site	Year	Site x Year
Seedling emergence	24.3(3,86)****	16.2(2,86)****	6.35(6,86)****
Taxa density	8.38(3,86)****	15.9(2,86)****	4.71(6,86)***
Evenness	10.2(3,79)****	3.17(2,79)*	3.55(6,79)**



Figure 2.2 Total number of seedlings, taxa density, and evenness of species emerging from soils collected from freshwater marshes: Kingman Marsh and Kenilworth Marsh (restored in 2000 and 1992-93, respectively), and Dueling Creek and Patuxent Marsh (urban and non-urban natural marshes, respectively). Values are least squares means \pm SE based on the 45.2 cm² surface area of each soil sample. Seedling number means and SEs were log₁₀(x+1) transformed, then detransformed for presentation. Means with different letters are significantly different across sites and years. These data represent non-flooded soil samples only.

Table 2.2 The dominant seed bank species found in 2003 at each of the four tidal freshwater marsh study sites, based on the arithmetic mean number of seedlings per pan \pm SE for the non-flooded treatment only. Unidentified dicots and monocots were not included in this list or in the total number of species; they were included in the total number of seedlings. The total number of taxa is a conservative count because some plants could only be identified to genus. The top 10 dominant species at each site are in boldface type. Dashed lines indicate that a particular species was not found at that site. Nomenclature is based on Brown and Brown (1984; 1992) and the USDA PLANTS Database (USDA 2005).

	Kingman Marsh (restored 2000)	Kenilworth Marsh (restored 1992-1993)	Dueling Creek (urban natural)	Patuxent Marsh (non-urban natural)
Cyperaceae	0.61 ± 0.304	2.9 ± 2.22	5.0 ± 4.51	3.3 ± 2.95
Cyperus spp.	20 ± 8.77	31 ± 21.0	$\textbf{4.3} \pm \textbf{3.33}$	—
Echinochloa spp.	0.17 ± 0.0904	1.9 ± 0.934	0.3 ± 0.333	—
Juncus effusus	$11 \pm 3.05*$	13 ± 5.09	6.0 ± 3.51	2.3 ± 1.54
Leersia oryzoides	0.83 ± 0.487	$14 \pm 5.14*$	15 ± 4.04	0.17 ± 0.167
Lindernia dubia	15 ± 4.38	$\textbf{8.4} \pm \textbf{7.00}$	0.3 ± 0.333	—
Ludwigia palustris	74 ± 22.0	$\textbf{8.0} \pm \textbf{3.57}$	$\textbf{3.0} \pm \textbf{3.00}$	$\textbf{3.8} \pm \textbf{2.39}$
Lythrum salicaria†	70 ± 16.0	85 ± 34.9	56 ± 36.3	1.3 ± 0.803
Mikania scandens	0.11 ± 0.0762	1.1 ± 0.742	$\textbf{3.0} \pm \textbf{1.00}$	5.2 ± 2.17
Peltandra virginica	$2.2 \pm 2.16*$	1.6 ± 1.10	—	—
Penthorum sedoides	$\textbf{2.1} \pm \textbf{0.95}$	0.12 ± 0.125		—
Phragmites australis†	—	3.5 ± 3.36	2.3 ± 2.33	—
Pilea pumila	0.056 ± 0.0556	0.25 ± 0.164	0.3 ± 0.333	10 ± 6.93
Poaceae	—	—	—	$\textbf{4.7} \pm \textbf{4.67}$
Polygonum punctatum	0.17 ± 0.0904	—	2.0 ± 2.00	2.7 ± 1.15
Polygonum sagittatum	—	—	$\textbf{3.0} \pm \textbf{2.08}$	2.7 ± 1.67
Rorippa palustris	1.3 ± 0.676	1.2 ± 0.366	3.3 ± 1.76	3.7 ± 2.17
Schoenoplectus tabernaemontani	$4.3 \pm 4.05*$	—	—	—
<i>Typha</i> spp.	7.3 ± 4.11	86 ± 36.0	2.7 ± 1.76	0.8 ± 0.307
Number of samples	18	8	3	6
Total no. taxa (2003)	38	27	22	32
Total no. seedlings (2003) Parameters:	4719	2759	415	340
Density/m ²	47561 ± 8850.5	58493 ± 12,917	$25125 \pm 11,046$	11089 ± 2967.7
No. seedlings/pan	215 ± 40.0	265 ± 58.4	114 ± 50.0	50.2 ± 13.5
Pielou's J (evenness)	0.7 ± 0.04	0.6 ± 0.05	0.7 ± 0.1	0.8 ± 0.05
Taxa density (species/sample)	10 ± 0.590	12 ± 1.13	12 ± 2.52	10 ± 0.882

Non-native† Planted* Marsh, the same pattern found in 2001 (Table 2.3). Kenilworth Marsh species composition was about as similar to Dueling Creek as Kingman Marsh was to Kenilworth Marsh. Similarity indices increased between 2001 and 2003 at Kingman Marsh and Dueling Creek, while Patuxent Marsh and Kenilworth Marsh remained static. All urban sites were least similar to Patuxent Marsh in both years (Table 2.3).

In 2000, about 5% of emerging seedlings from Kingman Marsh were annual species, which increased about five times by 2001, and then decreased by more than half in 2003 to 12% (Table 2.4). By 2003 Patuxent Marsh had the highest percentage of annual species (57%), while Kenilworth Marsh had approximately 7% annuals and Dueling Creek had about 9%. Fisher's Exact Test (for 2000) and the chi-square test of independence (for 2001 and 2003) were significant all three years (2000: P <0.0001; $2001: \chi^2_{0.05,3}=359.8$, P <0.0001; $2003: \chi^2_{0.05,3}=518.1$, P <0.0001), demonstrating that the percentage of annuals was not randomly distributed across sites.

The percentage of native species at Kingman Marsh was high (>95%) in 2000 and 2001, and decreased to about 70% in 2003 (Table 2.4). Dueling Creek's native species steadily increased across years, while Patuxent Marsh remained fairly static. Patuxent Marsh's seed bank, however, was already largely comprised of native species (>90%). Kenilworth Marsh, which varied between years, was similar to Dueling Creek throughout the study. The chi-square test of independence (for 2000, 2001, and 2003) was significant all three years (2000: $\chi^2_{0.05,3}$ =172.4, P <0.0001; 2001: $\chi^2_{0.05,3}$ = 1507.2, P <0.0001; 2003: $\chi^2_{0.05,3}$ = 434.8, P <0.0001). These findings signify that the distribution of native species is dependent on site.

Table 2.3 Sørensen's Quotient of Similarity for seed bank samples collected at two	
restored and two natural sites in 2001 and 2003.	

2001	Kingman Marsh	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
	(restored 2000)	(restored 1992-93)	(urban natural)	(non-urban natural)
Kingman	1	0.62	0.43	0.38
Kenilworth		1	0.52	0.41
Dueling Creek			1	0.40
Patuxent				1
Patuxent				1

2003	Kingman Marsh	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
Kingman	1	0.70	0.60	0.51
Kenilworth		1	0.65	0.40
Dueling Creek			1	0.57
Patuxent				1

	% Annuals (total seedlings)		% Nati	ives (total see	edlings)	
Area	2000	2001	2003	2000	2001	2003
Kingman Marsh (restored 2000)	5.4	26	12	96	97	69
Kenilworth Marsh (restored 1992-93)	3.4	3.8	6.8	36	48	44
Dueling Creek (urban natural)	0	2.3	8.8	30	45	50
Patuxent Marsh (non-urban natural)	31	20	57	92	97	96

Table 2.4 Percentage of annual and native seedlings that emerged by year at each study site. Seedlings identified to family or genus were not included in the totals.

Species of importance at Kingman Marsh differed between years and from those at other sites. In 2000, the most abundant species emerging from Kingman Marsh seed bank samples (pre-planting) were *Juncus effusus* and *Juncus tenuis*, although even these species occurred at only about two seedlings/sample (Neff 2002). In 2001, the most abundant species were *Lindernia dubia* (28.9 ± 12.32), *Ludwigia palustris* (80.9 ± 25.24), and Cyperaceae species (48.3 ± 12.11). Many other species occurred at levels higher than *Juncus* spp. did in 2000. Dominants in 2003 at Kingman Marsh included *Ludwigia palustris* and *Lythrum salicaria* (\geq 70 seedlings/sample) (Table 2.2). Dominant species at Kenilworth Marsh in 2003 included *Lythrum salicaria*, *Typha* spp. and *Cyperus* spp. (with values of approximately 85, 85, and 30 seedlings/sample respectively). *Lythrum salicaria* was important at all urban sites in 2003, comprising about one-third of the seedlings from Dueling Creek samples. The annual *Pilea pumila* was the most important species at Patuxent Marsh, but occurred in other sites at low densities.

The number of emerging seedlings was significantly about four times higher in non-flooded than in flooded treatments (Table 2.5). Taxa density of emerging seedlings was also significantly negatively affected by flooding, with about four species/sample in flooded samples and >10 species/sample in non-flooded samples (Table 2.5). Species evenness was not significantly affected by the flooded/non-flooded regime. About 10 taxa had significantly higher seedling emergence under non-flooded than flooded conditions, including *Cyperus* spp., *Juncus effusus*, *Juncus tenuis*, *Lythrum salicaria*, *Mikania scandens*, *Panicum dichotomiflorum*, *Panicum* spp., *Penthorum sedoides*, *Polygonum lapathifolium*, and *Rorippa palustris*. No species had significantly higher

Table 2.5 Arithmetic means \pm SEs for seedlings from all sites that germinated in the 2003 flooded versus non-flooded seedling emergence study. Arithmetic means \pm SEs are also presented for density/m², the number of seedlings per pan, Pielou's J and taxa density. Dashed lines indicate species were not found in that water regime. Bold numbers indicate the top five dominant species found in each treatment. Nomenclature and origin are based on the USDA PLANTS Database (USDA 2005).

*P<0.05

	Flooded	Non-flooded	
Species	Mean	Mean	P-value
Amaranthus cannabinus	_	0.057 ± 0.0398	0.1603
Ammannia coccinea	0.028 ± 0.0286	-	0.3244
Asteraceae	_	0.057 ± 0.0398	0.1603
Bidens spp.	0.028 ± 0.0286	0.028 ± 0.0286	1.00
Boehmeria cylindrical	-	0.23 ± 0.201	0.2639
Conyza canadensis	-	0.086 ± 0.0480	0.0831
Cuscuta gronovii	-	0.20 ± 0.141	0.1647
Cyperaceae	0.028 ± 0.0286	2.0 ± 0.802	0.0209*
<i>Cyperus</i> spp.	_	18 ± 6.61	0.0123*
Digitaria sanguinalis	_	0.028 ± 0.0286	0.3244
Draba sp.	_	0.028 ± 0.0286	0.3244
Dulichium arundinaceum	_	0.028 ± 0.0286	0.3244
Echinochloa spp.	0.17 ± 0.0765	0.54 ± 0.244	0.1536
Eleocharis obtusa	0.14 ± 0.0600	0.17 ± 0.145	0.8562
Epilobium coloratum	_	0.086 ± 0.0857	0.3244
Helenium autumnale	_	0.028 ± 0.0286	0.3244
Hydrilla verticillata	0.028 ± 0.0286	_	0.3244
Hypericum mutilum	_	0.14 ± 0.102	0.1688
Impatiens capensis	_	0.057 ± 0.0398	0.1603
Juncus acuminatus	0.28 ± 0.203	0.057 ± 0.0398	0.2769
Juncaceae	0.57 ± 0.0398	_	0.1603
Juncus effusus	0.26 ± 0.132	9.4 ± 2.03	<0.0001*
Juncus spp.	-	0.34 ± 0.235	0.1543
Juncus tenuis	-	0.26 ± 0.103	0.0178
Lamiaceae	-	0.057 ± 0.0398	0.1603
Leersia oryzoides	3.6 ± 1.29	4.9 ± 1.59	0.5241
Lindernia dubia	$\textbf{3.7} \pm \textbf{1.38}$	9.9 ± 2.91	0.0599
Ludwigia alternifolia	0.028 ± 0.0286	-	0.3244
Ludwigia palustris	19 ± 8.24	41 ± 12.6	0.1630
<i>Lycopus</i> sp.	-	0.028 ± 0.0286	0.3244
Lythrum salicaria	2.9 ± 2.77	60 ± 12.4	<0.0001*
Mentha arvensis	-	0.057 ± 0.0571	0.3244
Mikania scandens	-	1.5 ± 0.504	0.0067*
Myosotis laxa	-	0.057 ± 0.0398	0.1603
Nuphar lutea	0.028 ± 0.0286	-	0.3244
Panicum dichotomiflorum	0.028 ± 0.0286	0.71 ± 0.300	0.0292*
Panicum longifolium	—	0.028 ± 0.0286	0.3244
Panicum spp.	_	0.11 ± 0.0546	0.0437*
Peltandra virginica	0.77 ± 0.333	1.5 ± 1.13	0.5333
Penthorum sedoides	0.028 ± 0.0286	1.1 ± 0.511	0.0465*

(continued)

Table 2.5 (continued)

	Flooded	Non-flooded	
Species	Mean	Mean	P-value
Phalaris arundinacea	0.057 ± 0.0571	0.11 ± 0.0896	0.5927
Phragmites australis	0.028 ± 0.0286	1.0 ± 0.791	0.2279
Pilea pumila	0.43 ± 0.324	1.8 ± 1.27	0.2932
Poaceae	-	0.11 ± 0.0546	0.0437*
Poaceae sp.1	-	0.80 ± 0.800	0.3244
Polygonum arifolium	0.086 ± 0.0857	0.11 ± 0.0896	0.8184
Polygonum lapathifolium	0.028 ± 0.0286	0.28 ± 0.105	0.0233*
Polygonum pensylvanicum	-	0.057 ± 0.0571	0.3244
Polygonum punctatum	0.14 ± 0.102	0.71 ± 0.294	0.0736
Polygonum sagitattum	_	0.71 ± 0.368	0.0606
Polygonum spp.	0.17 ± 0.171	0.028 ± 0.0286	0.4165
Rorippa palustris	_	1.9 ± 0.536	0.0013*
Sagittaria latifolia	0.26 ± 0.150	0.028 ± 0.0286	0.1424
Saururus cernuus	_	0.028 ± 0.0286	0.3244
Schoenoplectus tabernaemontani	0.54 ± 0.486	2.2 ± 2.08	0.4436
Sonchus asper	_	0.057 ± 0.0398	0.1603
Symphyotrichum puniceum	_	0.086 ± 0.0857	0.3244
<i>Typha</i> spp.	12 ± 4.14	24 ± 9.96	0.2718
Unknown dicot	0.17 ± 0.119	1.1 ± 0.439	0.0591
Uknown monocot	0.57 ± 0.240	2.0 ± 0.482	0.0106*
Parameters:			
Density/m ²	$10,100 \pm 2410$	$41,900 \pm 6060$	<0.0001*
No. seedlings/pan	46 ± 10.9	190 ± 27.4	<0.0001*
Pielou's J (evenness)	0.68 ± 0.0409	0.67 ± 0.0264	0.7994
Taxa density (species/sample)	4.2 ± 0.358	11 ± 0.460	<0.0001*

means in flooded pans, but *Leersia oryzoides*, *Ludwigia palustris* and *Typha* spp. germinated well in both flooded and non-flooded conditions.

Discussion

Patterns of seed density and diversity

The seed bank at Kingman Marsh developed rapidly, showing large increases in emerging seedling density and taxa density between 2000 and 2001. In 2003, all sites were similar with regard to these parameters (Figure 2.2). In a created tidal wetland in Delaware, Leck (2003) found significantly higher seedling density (means ranging from about 450 to 62,000 seeds/m² in the first year and 55,000 to 301,000 seeds/m² in the second year) and taxa density (increasing from 9 to 22 species/sample) after one year of development. These numbers were higher than at the natural sites, a finding also reported by Baldwin and Derico (1999) in a previous study of Kenilworth Marsh conducted approximately four years after restoration (mean values of eight to 13 species/sample at the restored sites and seven to eight species/sample at the natural sites; mean density of 75,000 to 130,000 seeds/m² at the restored sites and 15,000 to 55,000 seeds/m² at the natural sites).

Sources of buried seeds

There are several possible sources for the high densities of seed found at Kingman Marsh in 2001, including: 1) seeds from dredge material; 2) seed production by the planted species during the 2000 growing season; 3) seed dispersal into the site (air and water) that directly entered the seed bank; and 4) seed production by plants that
colonized, flowered, and reproduced during the 2000 growing season. Regarding dredge material (1), the seed bank from 2000 had low numbers of emerging seedlings and low taxa density, so apparently few seeds were present in the dredge material (or dispersed into the site prior to our 2000 seed bank collection) (Neff 2002). Siegley et al. (1988) found a seed density of 980 seeds/m² in dredge material, similar to our report of about 310 seeds/m² in Kingman Marsh dredge material. The contribution of seeds produced by planted species to the seed bank (2) is a possibility for three of the seven species planted: *Juncus effusus, Peltandra virginica*, and *Schoenoplectus tabernaemontani* (Table 2.2). A low early contribution of planted species to the seed bank was found by Collins and Wein (1995), with planting not affecting richness or species composition and only one species, *Eleocharis quadrangulata*, being present mainly in seed bank samples from planted sections.

Of the remaining potential seed sources, seed dispersal (3) and seed production by colonizing plants (4), other research (Neff and Baldwin 2005) has shown high densities of seeds dispersed into Kingman Marsh, with the majority of species originally entering through water dispersal. Since high densities of seed continued to be dispersed into Kingman Marsh throughout 2000 (Neff and Baldwin 2005), it seems likely that seed dispersal directly contributed to the seed bank. Comparing the 2001 seed bank collected from transects with and without vegetation cover in 2000, allows an estimate of relative quantities of seed entering the seed bank through direct dispersal and seed entering through seed rain of adjacent vegetation. The three Kingman Marsh transects with little or no vegetation in 2000 had low seedling density (approximately 2,200 seeds/m²) and species density (six species/sample) in 2001, values about twice as high as those found

for the seed bank in 2000, but much lower than the overall seedling density and species density in 2001. Since seed rain directly from the vegetation cover contributed little seed to these three transects and increases in seed were dependent on dispersal, these findings suggest that seed rain from established volunteer (i.e., non-planted) vegetation was the source of the majority of seeds found in the 2001 seed bank. It is possible other environmental factors limited seed deposition at these transects, including lower elevations that encouraged seed to continue floating, and lack of vegetation to trap dispersed seed (Griffith and Forseth 2002). Many seeds that were dispersed into Kingman Marsh in 2000 grew, flowered, and seeded during the 2000 growing season, with most species in the 2001 seed bank also being documented in the 2000 vegetation (Neff 2002).

Two-thirds of the 2003 seed bank was due to two species, *Ludwigia palustris* and *Lythrum salicaria*. Different species may develop seed banks by different mechanisms, as illustrated by these two predominant species. *Ludwigia palustris* was abundant in water dispersal samples, while *Lythrum salicaria* was present but at much lower density (Neff and Baldwin 2005). However, both occurred at similar density in 2003. This suggests that *Lythrum salicaria* established from few seeds and then reproduced prolifically, depositing many seeds into the seed bank. Seeds of *Ludwigia palustris*, on the other hand, likely established a high-density seed bank initially.

Effect of seed bank sampling date

Like Kingman Marsh, Patuxent Marsh and Dueling Creek seed and taxa density increased between 2000 and 2001, suggesting that the increases seen at Kingman Marsh during that time might be due to natural temporal fluctuations or late sampling date

(May) in 2000. A late sampling date has been found to affect natural sites more than restored sites, since natural sites tend to have higher densities of transient large-seeded species (e.g., *Impatiens capensis, Polygonum arifolium*) while restored sites are dominated by persistent small-seeded seeds (e.g., Cyperaceae, Juncaceae) (Thompson and Grime 1979; Leck and Brock 2000). We also observed this in our study. Another reason the late sampling date in 2000 may have had little effect on the restored Kingman Marsh samples was because the soil had recently been placed and seed germination, the main source of seed loss from the seed bank during this time, was nearly absent in May 2000 (personal observation).

Of the important seed bank species seen at Patuxent Marsh or Dueling Creek in 2001 but not in 2000, many of them are large-seeded species that may be transient (e.g., *Pilea pumila, Polygonum arifolium*, and *Polygonum sagittatum*), meaning that the seeds germinate early in the growing season or lose viability, and therefore do not persist year-round. Other species in the Dueling Creek and Patuxent Marsh seed banks found in 2001 but not 2000 included known transient species (e.g., *Impatiens capensis, Peltandra virginica*) (Leck and Simpson 1987). We found no significant change in seedling density, taxa density, or evenness at Kenilworth Marsh between 2000, 2001, or 2003, likely because the seed bank there is largely made up of persistent species. The increase in density at Kingman Marsh was due to seed rain dispersal by small-seeded persistent species at Patuxent Marsh and Dueling Creek explain the low number of seedlings and taxa density at these sites in 2000. The May 2000 sampling date probably occurred after many of these species had set seed. This also explains the increases in seedling number and taxa

density we observed at these sites in 2002 and 2003, when sampling occurred in March, before seed set. The dominant vegetation at Kenilworth Marsh is largely comprised of monotypic stands of *Typha* spp. (Neff 2002) and stands of *Lythrum salicaria*, both of which are abundant in the seed bank and appear to persist there. These species contribute to the high number of seedlings seen each year (Table 2.2).

In 2001, taxa and seedling density at Patuxent Marsh were still lower, though not statistically different, than at the other sites, though evenness was higher. Other studies have also found seed bank density and richness to be higher at restored sites than at natural sites (Baldwin and Derico 1999; Leck 2003). The low values at Patuxent Marsh may have been due to dominant vegetation species that produce few or no seeds (e.g., *Acorus calamus*) (Leck and Simpson 1987). The continued upswing in seedling and taxa density at Patuxent Marsh from 2001 to 2003, though not significant, could be due to natural variation in the seed bank. Interestingly, Patuxent Marsh had more species in 2003 than in 2001, but lower evenness because one of its species (*Pilea pumila*) was found in much greater abundance in the seed bank than other species. The decrease in evenness seen at Patuxent Marsh may also be due to a beaver dam that has drastically altered the hydrology at the site. It was constructed in mid-2002 and has begun to change the vegetation across the wetland in response to the alteration of tidal fluctuation (see Chapter 4).

Effect of flooding

Non-flooded experimental pans had significantly higher seedling emergence and taxa density than flooded pans. These findings were analogous to Baldwin and Derico's

(1999) work at Kenilworth Marsh and Dueling Creek in 1997. Seedling emergence has been shown to decrease due to flooding in previous studies (Baldwin et al. 2001; Wetzel et al. 2001; Peterson and Baldwin 2004). Some species will not germinate without a drawdown period (van der Valk 1981). Draw-down does not occur for long in this ecosystem due to semidiurnal tides. These findings illustrate the importance of designing a restored or created wetland with adequate elevation (i.e., high enough) to support seedling emergence. If the elevation is too low, seedlings may not establish, even if seeds are plentiful in the soil. *Lythrum salicaria*, a non-native, often invasive species, was found in significantly higher numbers in non-flooded conditions, indicating that flooding could be a control measure for this species. *Polygonum lapathifolium* was also significantly affected by flooding in a negative manner.

Increases in site similarity

According to Sørensen's Quotient of Similarity, Kingman Marsh and Kenilworth Marsh, the two restored sites, are becoming more similar with time, based on seed (Table 2.3). Kenilworth Marsh is also becoming more similar to Dueling Creek, the natural urban marsh. This finding suggests that, with time, the restored marshes may one day resemble natural urban marshes with regard to their species composition. These sites are likely much more similar to each other than to Patuxent Marsh because they have similar stresses placed upon them due to their location (e.g., low water quality (low dissolved oxygen, high coliform counts), high storm water runoff, intensive surrounding land use). All three urban marshes are surrounded by development (65% urban in Anacostia vs. 21% urban in Patuxent), connected by the same waters, and geographically close. This

may increase the likelihood that they share similar seed bank species. Interestingly, Kingman Marsh and Dueling Creek became much more similar to Patuxent Marsh in 2003 but Kenilworth Marsh did not. Kenilworth Marsh had a fair number of taxa in 2003 and had the highest seedling density (Table 2.2), but neither of these propelled its species makeup towards that of a non-urban natural marsh. *Polygonum* species appear to be absent from Kenilworth Marsh's seed bank, while at Patuxent Marsh they are dominant species.

The percentage of annual species at each site increased between 2000 and 2003 (Table 2.4). This could be due to natural seed bank variation, annual changes in vegetation composition, or more favorable germination conditions for the annual species present at the sites. Parker and Leck (1985) found seven of the 10 most dominant species in their tidal freshwater wetland study to be annuals, but annuals comprised only half of the species found in the seed bank of each of their study zones. Baldwin and Derico (1999) also found higher numbers of annuals at the restored Kenilworth Marsh than at Dueling Creek in 1997.

The percentage of native species decreased between 2000 and 2003 at Kingman Marsh, increased at Kenilworth Marsh and Dueling Creek, and remained relatively stable at Patuxent Marsh (Table 2.4). The natives lost ground at Kingman Marsh in 2003 because *Lythrum salicaria*, a non-native, was the second most dominant species (Table 2.2). This species tends to become invasive and may continue this trend at Kingman Marsh. This species may be locally abundant and therefore show dominance in the seed bank (Leck and Graveline 1979). Kenilworth Marsh and Dueling Creek had 50% or less native species in their seed banks, also due to the large numbers of *Lythrum salicaria*

found in their samples. Kingman Marsh had approximately equal numbers of *Ludwigia palustris* and *Lythrum salicaria* per pan during 2003, likely keeping Kingman Marsh's non-natives in a more stable ratio with its natives. Although *Ludwigia palustris* was found in the seed bank at Patuxent Marsh, it is rarely seen during vegetation surveys. Care was taken to pull seedlings prior to flowering in the greenhouse, but one or two seeds may have contaminated a Patuxent Marsh pan and led to these findings.

Implications for wetland restoration

These studies suggest that a large seed bank will quickly develop in restored or created wetlands with surface water connectivity to other wetlands. Before spending large sums of money in providing supplemental plant material, inexpensive seed bank/seed transport studies may be a worthwhile investment prior to planting. If seed assays reveal adequate densities of seed (Mitsch and Cronk 1992), and the seed composition has low densities of undesirable species (Pederson and van der Valk 1984; Brown 1998), there may be little need to provide additional propagules through planting or seeding (Kentula et al. 1992). However, it may be necessary to supplement these propagules if there are certain desired species missing from the seed bank (Zedler 2000), as some species may take a long time to naturally reach the site (e.g., Acorus calamus, Impatiens capensis, Polygonum arifolium, Polygonum sagittatum). As with vegetation development at a restored wetland, it is still unclear if these restored wetland seed banks will ever resemble those of natural wetlands or if they will develop into something altogether different. After 10 years, the seed bank of Kenilworth Marsh still did not resemble the seed banks of the natural sites. Comparisons of Kenilworth Marsh at 10 years old (this study) versus

Kenilworth Marsh at three and a half years old (Baldwin and Derico 1999) show that seed densities have decreased over time (mean of 75,000-95,000 seeds/m² to 40,000 seeds/m²). However, richness values have increased over time (mean of 8.5-10.5 species/samples to 12 species/sample), suggesting that the seed bank at Kenilworth Marsh continues to change.

Conclusion

In conclusion, seed banks are an important component of vegetation dynamics, providing a history of the site as well as the availability of seeds for regeneration following disturbance, including restoration. Seed banks in some restored wetlands, e.g., tidal, develop rapidly due to their hydrologic connectivity to other areas that may provide seed sources. The rapid establishment of seed banks makes them a parameter that could easily be studied in tidal areas to determine potential vegetation composition of a site. Since the seed bank is a reflection of biotic and abiotic factors (e.g., vegetation, hydrology, disturbance, herbivores), and is relatively easy to study, we suggest that seed banks are a valuable metric for evaluating the success of wetland restoration projects.

Chapter 3: Defining Reference Sites for Urban Restoration: Are Natural Non-urban Wetland Sites Suitable for Wetland Restoration Comparisons? Evidence from Maryland and Washington, D.C.

Introduction

The field of wetland restoration has grown tremendously during the last two decades. During that time, many manuals and descriptive guides were introduced, including: *An Approach to Improving Decision Making in Wetland Restoration and Creation* (Kentula et al. 1992), *Wetland Restoration, Flood Pulsing, and Disturbance Dynamics* (Middleton 1999), and *Handbook for Restoring Tidal Wetlands* (Zedler 2001). Many sources of restoration information promote reference sites to guide restoration, especially when little or no background information is known about the site to be restored (Hobbs and Harris 2001; Zedler 2001; Kentula 2002; Interagency Workgroup on Wetland Restoration 2003; Thayer et al. 2003). Reference sites, in the most general terms, are usually natural sites (as geographically close to a potential restoration site as possible) that: 1) can be referred to when planning a restoration; or 2) used as a comparison for a restored site during its development.

Reference sites present a view of how a degraded site may once have looked and functioned, and provide information (e.g., soils, hydrology, or plant information) about the local area (Interagency Workgroup on Wetland Restoration 2003; Thayer et al. 2003). They are especially helpful if history or background information cannot be found for the degraded site, which is often the case (Interagency Workgroup on Wetland Restoration 2003). Reference sites should, however, be similar to restored sites with regard to size,

tidal range, water quality, landscape position, and surrounding land use (Neckles et al. 2002).

Several authors have proposed caution or questioned the use of natural reference sites with urban restored areas (Kentula et al. 1992; Ehrenfeld 2000b; Baldwin 2004) because the predictability of restoration succession can be difficult and fraught with surprises (Zedler 2001). If chosen references are natural and relatively undisturbed by humans, the restored wetland may never reach those conditions (National Research Council 1992; Ehrenfeld 2000a; Warren et al. 2002; Hilderbrand et al. 2005), particularly in urban areas (Brinson and Reinhardt 1996; Grayson et al. 1999). The surrounding land use and history of a site may preclude it from reaching the desired trajectory. Both Grayson et al. (1999) and Thayer et al. (2005) have defined multiple reference sites that may be used in restoration projects (control sites that are degraded and not restored, and natural reference sites having conditions the restoration can hope to attain) so that developments in a restored site can be credited to the restoration, to nature, or to other outside forces. Though restored sites are often carefully planned, environmental conditions ultimately determine how a system progresses, sometimes even in the face of intense management. Outcomes of a restoration therefore, may not be those planned for or desired (Klötzli and Grootjans 2001). Natural systems too go through variation and disturbances (Middleton 1999) (see Chapter 4) and cannot be expected to follow predictable successional stages (Klötzli and Grootjans 2001).

The original objective of this study was to monitor the vegetation of a recently restored marsh, Kingman Marsh, an older restored marsh, Kenilworth Marsh, a natural urban marsh, Dueling Creek, and a non-urban natural marsh, Patuxent Marsh, during the

years 2002 through 2004. I wanted to explore the differences and similarities at all four sites with regard to cover, diversity, taxa density, and evenness. Both Dueling Creek and Patuxent Marsh served as reference sites for the restoration work at Kingman Marsh. However, as differences between the urban and natural reference sites became clearer, I became interested in the question of whether urban reference sites are more appropriate for urban restorations than non-urban reference sites. Specifically, is Patuxent Marsh, a relatively undisturbed, non-urban, natural marsh, an appropriate reference site for the more urban, long disturbed Kingman Marsh or, is Dueling Creek, the urban reference site, a more appropriate restoration target? To my knowledge, this study is the first to compare urban and non-urban reference sites in evaluating restoration success of wetlands.

Methods

Study sites

Kingman Marsh, Kenilworth Marsh, and Dueling Creek are located in the Anacostia watershed in Maryland and Washington, D.C. This watershed is approximately 375 km² and has the following land uses: 65% urban, 8% agriculture, 27% forest, 0.2% wetland, and 0.4% barren. Patuxent Marsh is located in the Patuxent River watershed in Maryland, which is approximately 269 km², and has the following land uses: 21% urban, 29% agriculture, 47% forest, 2% wetland, and 0.8% barren (MDNR and MDE, http://www.dnr.state.md.us/watersheds/surf/index.html, accessed 10 October 2005). Kenilworth Marsh, located 0.4 kilometers upstream of Kingman Marsh within the Kenilworth Aquatic Gardens National Park, was restored in 1992-93 in a manner similar to Kingman Marsh. In 2001, RODEO herbicide was applied by the National Park Service to large expanses of *Phragmites australis* in Kenilworth Marsh which affected vegetation transects sampled during this study (Hammerschlag et al. 2001; Neff 2002). (See Chapter 2 for more site detail.)

Study set-up

This research was part of a cooperative study between the University of Maryland and the U.S. Geological Survey. Transects were randomly placed in Spring 2000 by these groups in each of the four marshes: 18 at Kingman Marsh; eight in Kenilworth Marsh; three in Dueling Creek; and six in Patuxent Marsh. (See Neff 2002 for full details of transect set-up.) Transects were 35 m long and anchored by (permanent) PVC end pole markers. Each transect also had a start and end point to ensure that the same side of the transect was monitored each year. Each transect was divided into seven 5 m long by 1 m wide sectors.

Vegetation transect monitoring occurred in mid-May, mid-July, and mid-September of 2000-04. This research is focused on data collected in 2002-04 because: 1) I assisted in the collection of this data during my Masters research work, which began in 2002; and 2) waiting a couple of years for Kingman Marsh to establish avoids the initial two years of vegetation development, during which Kingman Marsh experienced rapid initial plant colonization followed by extensive vegetation dieback due to goose herbivory and hydrologic changes (Hammerschlag et al. 2000; 2001; Neff 2002). A team of two people sampled transects by sector, and recorded species and percent cover on field data sheets. Tape measures were strung between the two PVC poles, parallel to the

sectors, with care taken not to walk on vegetation to be sampled. Teams assessed transects from above, sector-by-sector, either averaging individual estimates of percent cover by species, or deciding on a single estimate together. Measures of no cover were also assessed. Cover classes used were: 0.1%, 0.5%, from 1-15% in units of one percent, and from 15-100% in units of five percent. Data for each transect was checked in the field to have at least 100% cover including areas of no cover and detritus, with more than 100% cover possible if there were multiple vegetation canopies. Detritus results are not presented in this paper.

Data analysis

Plant taxonomy nomenclature and life history were based on Brown and Brown (1984; 1992) and the USDA PLANTS Database (USDA 2005). This study includes data only from 2002 through 2004 because Kingman Marsh experienced a rapid colonization immediately after sediment placement in 2000, but vegetation began to disappear the subsequent year (Hammerschlag et al. 2001; Neff 2002). Vegetation was analyzed with a repeated measures ANOVA (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Mixed) to detect differences in vegetation between sites and time (months within years or between years). A critical level of P = 0.05 was used in determining significance. We ran the Tukey-Kramer test to determine pairwise mean comparisons. *Lemna minor* and *Hydrilla verticillata*, aquatic floating and submerged species, respectively, were removed from primary data presentations and calculations, but included in results to show differences that these species caused. Because total vegetation cover met the

assumptions of ANOVA and normality, we assumed normality and homogeneity of variances for all individual species and vegetation parameters for consistency of analysis.

The evenness index, Pielou's J, was calculated similarly to seed bank analysis in Chapter 2. The percentage importance (p_i) in this study was relative cover expressed as a proportion and S was the species richness across sectors within transects (Peet 1974; McCune and Grace 2002). Dominant species were identified as the ten most abundant species based on the least squares means of cover for each species across years. Unidentified dicots and monocots were not included in the dominant species list. *Phragmites australis* was assumed to be the non-native genotype for classification purposes. Sørensen's Quotient of Similarity was used to determine the similarity in species composition at the four sites and was calculated as in Chapter 2.

The total number of annuals and natives was determined for each site in each year. These data were then analyzed with an ANOVA (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Mixed) to detect differences between sites within years. A critical level of P = 0.05 determined significance, and the Tukey-Kramer test determined pairwise mean comparisons. As for other vegetation analyses, *Lemna minor* and *Hydrilla verticillata* were removed from primary data presentations and calculations, but included in the results to illustrate differences due to these species. Species classified as annual/biennial or annual/perennial in the USDA PLANTS Database (2005) were included with the annuals. Vegetation identified only to family or genus was not included in the native species calculations, but was included in the annual/perennial species calculations if the whole genus fell within either the annual or perennial category.

Results

Percent cover and taxa density

Kingman Marsh, the younger restored site, had significantly lower cover (28%) than the other sites (restored: Kenilworth Marsh 97%; natural: Dueling Creek 84%; Patuxent Marsh 86%), which were not significantly different in their percent covers (Figure 3.1). There were no site by time interactions for cover or taxa density (Table 3.1), so main effects could be examined. Cover and taxa density had strong site and time (month within year) effects (site: P<0.0001 for cover and taxa density; time: P<0.0001 for cover, P<0.01 for taxa density) (Table 3.1; Figure 3.1; Figure 3.2). Plotted by month and year, Kingman Marsh consistently had the lowest vegetation cover (Figure 3.3), although it followed the pattern of lowest cover in May, highest cover in July, and intermediate cover in September seen at the other sites and in the plots by time (Figure 3.2; Figure 3.3). July 2002 had the highest overall percent cover (Figure 3.2), although the sites remained fairly consistent in their vegetation cover between 2002 and 2004 (Figure 3.3).

Patuxent Marsh was separated into two areas with one transect on the north side of a highway and the other five transects on the south side (see Chapter 4). The south side transects began to be inundated in mid-2002 when a beaver dam was constructed across the creek which feeds the marsh, creating a large ponded area that enveloped the transects (see Chapter 4). This area of the marsh contained two aquatic species not found at the north side transect: *Hydrilla verticillata* and *Lemna minor*. While these species were removed from the primary analyses, alternative analyses were conducted with them so



Figure 3.1 Site main effects of mean cover, taxa density, Shannon-Weiner diversity, and evenness at four tidal freshwater marshes. Values are least squares means \pm SE. Solid symbols represent data not containing the submerged species, *Hydrilla verticillata*, and the floating species, *Lemna minor*, while open symbols include these species. Means with different letters are significantly different across sites and apply only to the solid symbols (emergent species only).

Table 3.1 Analysis of variance table of vegetation parameters for all sites. Significance is: *<0.05; **<0.01; ***<0.001; ****<0.0001. Expression written Fvalue (Numerator df, Denominator df) significance.

Emergent species only:					
Parameter	Site	Month(Year)	Site*Month(Year)		
Cover	133(3,277)****	4.11(8,277)****	1.53(24,277)		
Taxa density	89.9(3,277)****	2.88(8,277)**	0.882(24,277)		
Shannon-Wiener Diversity	93.8(3,277)****	1.31(8,277)	0.731(24,277)		
Evenness	71.6(3,277)****	0.336(8,277)	1.00(24,277)		
Annual cover	54.6(3,301)****	30.5(2,301)****	13.6(6,301) ****		
Perennial cover	91.6(3,301) ****	0.681(2,301)	2.40(6,301)*		
Native cover	126(3,301) ****	6.92(2,301)**	4.66(6,301)***		
Non-native cover	20.8(3,301) ****	0.0653(2,301)	0.294(6,301)		

With aquatic species, *Hydrilla verticillata* and *Lemna minor*:

Parameter	Site	Year	Site*Year
Cover	167(3,277)****	4.94(8,277)****	0.946(24,277)
Taxa density	104(3,277) ****	2.66(8,277)**	0.657(24,277)
Shannon-Wiener Diversity	113(3,277)****	1.33(8,277)	0.775(24,277)
Evenness	79.7(3,277)****	0.318(8,277)	0.957(24,277)
Annual cover	54.6(3,301) ****	30.5(2,301)****	13.6(6,301) ****
Perennial cover	94.7(3,301) ****	0.822(2,301)	2.71(6,301)*
Native cover	141(3,301) ****	4.84(2,301)**	2.93(6,301)**
Non-native cover	10.7(3,301) ****	0.0761(2,301)	0.227(6,301)



Figure 3.2 Time main effects (month(year)) of mean cover, taxa density, Shannon-Weiner diversity, and evenness across 2002, 2003, and 2004 at four tidal freshwater marshes. Values are least squares means \pm SE. Solid symbols represent data not containing the aquatic species *Hydrilla verticillata* and *Lemna minor*, while open symbols include these species. Means with different letters are significantly different across sample dates and apply only to the solid symbols (emergent species only). Means with no letters indicate that no significant differences were found.

that their effects on measured parameters could be studied. Patuxent Marsh would have possessed the highest percent cover if these species were included with the emergents (Figure 3.1).

Kingman Marsh possessed significantly lower taxa density (2 species) than Kenilworth Marsh (6 species), Dueling Creek (8 species), or Patuxent Marsh (7 species) (Figure 3.1). The same pattern described above for cover across time can be discerned at all sites in 2002, but in 2003 and 2004, there was not an obvious pattern among the sites (Figure 3.3). Only Kenilworth Marsh increased in species between May and September 2004; the other sites decreased between these same months (Figure 3.3). The largest amount of species were found in 2002 (approximately 7), with 2003 being a low year (5 species), and 2004 an intermediate year (5-6 species) (Figure 3.2).

Shannon-Weiner diversity and evenness

Kingman Marsh had significantly lower diversity than the other sites (Figure 3.1). Kenilworth Marsh had two and half times greater diversity than Kingman Marsh, while Dueling Creek and Patuxent Marsh had approximately three times greater diversity (Figure 3.1). Main effects of site (P<0.0001) were examined when no site by time interactions or significant time effects for diversity or evenness were found (Table 3.1). Similar to taxa density, diversity was highest across all sites in 2002, lowest in 2003, and intermediate between the two in 2004 (Figure 3.2). Diversity, percent cover, and taxa density were similar between Kenilworth Marsh, Dueling Creek, and Patuxent Marsh in September 2004 (Figure 3.3, Figure 3.4). Kingman Marsh had significantly lower evenness (approximately two times lower) than Kenilworth Marsh, Dueling Creek, or



Figure 3.3 Plots of site by month(year) interaction means for percent cover and taxa density of species measured in vegetation transects from freshwater marshes: Kingman Marsh and Kenilworth Marsh (restored in 2000 and 1992-93, respectively), and Dueling Creek and Patuxent Marsh (urban and non-urban natural marshes, respectively). Values are least squares means \pm SE.



Figure 3.4 Plots of site by month(year) interaction means for evenness and Shannon-Weiner diversity of species measurements taken in vegetation transects from freshwater marshes: Kingman Marsh and Kenilworth Marsh (restored in 2000 and 1992-93, respectively), and Dueling Creek and Patuxent Marsh (urban and non-urban natural marshes, respectively). Values are least squares means \pm SE.

Patuxent Marsh (Figure 3.1). With the exception of a drop in evenness across all sites in July 2003, evenness was fairly consistent over time (approximately 0.5-0.6) (Figure 3.2).

Annuals vs. perennials and natives vs. non-natives

There was a general trend among natural sites to have a higher cover of annual species in 2002 and lower cover in 2003 and 2004 (Figure 3.5). Kingman Marsh remained fairly consistent across time at approximately 2% coverage of annual species, and Kenilworth Marsh increased annual species cover between 2003 and 2004 (Figure 3.5). Cover of annuals at Patuxent Marsh decreased significantly between 2002 and 2003 (approximately 55% to 15%), resulting in a significant site by year interaction (Table 3.1; Figure 3.5). Kenilworth Marsh was not significantly different than Kingman Marsh in its cover of annual species during 2002 and 2003, while in 2004 Dueling Creek and Patuxent Marsh were not significantly different than Kingman Marsh (Figure 3.5). Between 2002 and 2004, Dueling Creek, Kenilworth Marsh, and Patuxent Marsh converged to approximately 15% annual cover (Figure 3.5).

Kenilworth Marsh consistently had the highest perennial cover across years, between 80% and 90% (Figure 3.5). Patuxent Marsh, including the two aquatic species, had the next highest coverage of perennials. Patuxent Marsh (minus the two aquatic species) and Dueling Creek had between 50% and 60% coverage of perennials between 2002 and 2004 (Figure 3.5). Kingman Marsh had the lowest coverage of perennial species across all years (Figure 3.5). In 2003 and 2004, Kingman had significantly lower coverage of perennials than all other sites (Figure 3.5). This pattern of variation in cover between sites over time resulted in a significant site by year interaction (Table 3.1).



Figure 3.5 Cover of annual and perennial species measured in 2002, 2003, and 2004 at four tidal freshwater marshes. Values are least squares means \pm SE. Means with different letters are significantly different across sites and years. Samples identified to family were not included in the calculations for annual and perennial species, but samples identified to genus were included in these calculations if the genus contained only one life history type. Calculations for Patuxent Marsh were made with (solid squares) and without (open triangles) the perennials *Hydrilla verticillata* and *Lemna minor* to show differences associated with these aquatic species. Mean comparisons were not run on Patuxent Marsh samples containing all species.

Kingman Marsh had significantly lower percent cover of native species than all the sites, resulting in an approximately four to five times lower percent cover of natives than Patuxent Marsh, the site with the highest coverage of natives in 2002 and 2003 (Figure 3.6). Both Dueling Creek and Patuxent Marsh followed a downward trend in their coverage of native species between 2002 and 2004, while Kenilworth Marsh had an upswing in this parameter during those same years, resulting in a significant site by year interaction (Table 3.1; Figure 3.6). *Lemna minor*, a native aquatic species, did not affect cover at Patuxent Marsh until 2004, when it increased Patuxent's native cover by approximately 15% (Figure 3.6).

There was a clear and strongly significant site effect on percent cover of nonnative species, with all sites remaining fairly static in their percent cover of non-natives across years (Table 3.1; Figure 3.6). Kenilworth Marsh had approximately 20% cover of non-natives in 2002 and 2003, the highest cover at any site, and approximately 16% cover in 2004 (Figure 3.6). Including *Hydrilla verticillata* in calculations gave Patuxent the second highest percent cover of non-native species (approximately 10%) (Figure 3.6). By removing this invasive, which began to expand throughout the marsh following construction of the beaver dam, Patuxent had the lowest percent cover of non-natives (approximately 0.5%) (Figure 3.6). Kingman Marsh and Dueling Creek remained intermediate to the other two sites, with about 5% non-native cover (Figure 3.6).



Figure 3.6 Cover of native and non-native species measured in 2002, 2003, and 2004 at four tidal freshwater marshes. Values are least squares means \pm SE. Means with different letters are significantly different across sites and years. Samples identified to family or genus were not included in the calculations for native and non-native species. Calculations for Patuxent Marsh were made with (solid squares) and without (open triangles) the non-native species, *Hydrilla verticillata*, and the native species, *Lemna minor*, to show differences associated with these aquatic species. Mean comparisons were not run on Patuxent Marsh samples containing all species.

Site similarities

In 2002, Kingman Marsh, the younger restored site, was most similar to Kenilworth Marsh (qs=0.69), the older restored site, and least similar to Patuxent Marsh (qs=0.43), the non-urban natural site (Table 3.2). Kenilworth Marsh was most similar to Kingman Marsh, and least similar to Patuxent Marsh (qs=0.46). Dueling Creek, the urban natural site, was most similar to Kenilworth Marsh (qs=0.59) and least similar to Kingman Marsh (qs=0.47), while Patuxent Marsh was most similar to Dueling Creek (qs=0.55) and least similar to Kingman Marsh.

In 2004, Kingman Marsh was again most similar to Kenilworth Marsh (qs=0.57), but at a lower similarity than in 2002 (Table 3.2). Kenilworth Marsh was most similar to Dueling Creek (qs=0.59), and likewise Dueling Creek was again most similar to Kenilworth Marsh. Patuxent Marsh was most similar to Kenilworth Marsh (qs=0.52) and equally similar to Kingman Marsh and Dueling Creek (qs=0.47). Patuxent Marsh became less similar to Dueling Creek, the other reference site, in 2004. Kingman Marsh became more similar to Patuxent Marsh and Dueling Creek, the natural sites, in 2004, while Kenilworth Marsh became more similar to Patuxent Marsh and remained the same similarity to Dueling Creek. In summary, both restored marshes (Kingman and Kenilworth) were more similar to the urban reference site, Dueling Creek, than they were to the non-urban reference, Patuxent Marsh in 2002 and 2004.

Peltandra virginica and *Typha* spp. were within the top ten dominant species at all four sites (Table 3.3). *Peltandra virginica* was the most dominant species in Kingman Marsh (approximately 8%), and was the second most dominant species at each of the

Table 3.2 Sørensen's Quotient of Similarity for transect monitoring at a young restored site, Kingman Marsh, an older restored site, Kenilworth Marsh, an urban natural site, Dueling Creek, and a non-urban natural site, Patuxent Marsh, in 2002 and 2004.

2002	Kingman Marsh	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
Kingman Marsh	1	0.69	0.47	0.43
Kenilworth Marsh		1	0.59	0.46
Dueling Creek			1	0.55
Patuxent Marsh				1

2004	Kingman Marsh	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
Kingman Marsh	1	0.57	0.51	0.47
Kenilworth Marsh		1	0.59	0.52
Dueling Creek			1	0.47
Patuxent Marsh				1

other three sites. *Lythrum salicaria*, a non-native species, was the second most dominant plant at Kingman Marsh. It was also a dominant at Kenilworth Marsh and Dueling Creek. Three of the seven species planted at Kingman Marsh, *Juncus effusus*, *Peltandra virginica*, and *Schoenoplectus tabernaemontani*, were within its top ten dominant species (Table 3.3). Kenilworth Marsh's most dominant species was *Typha* spp. and its third most dominant was *Phragmites australis*, an invasive plant, also found among the top ten dominants of Kingman Marsh. Dueling Creek also contained *Phragmites australis*, but outside the area of transect sampling (personal observation).

The most dominant species at Patuxent Marsh, *Nuphar lutea*, only also occurred at Kingman Marsh (Table 3.3), although it was also found at Kenilworth Marsh outside of the transects. Both *Hydrilla verticillata* (non-native) and *Lemna minor*, the aquatic species, fall within the top five dominant species at Patuxent Marsh. The most dominant species at Dueling Creek was *Leersia oryzoides*, also among the top five dominants of Kenilworth Marsh. *Sparganium eurycarpum*, a dominant at both natural sites, was absent at the two restored marshes. Kingman Marsh had the highest amount of no cover, defined as unvegetated mudflats or open water, among the four sites (Table 3.3). Dueling Creek had the next highest amount, but it was approximately one third the amount in Kingman Marsh. Kenilworth Marsh had the smallest amount of open area (6%).

The only dominant annual species occurring at Kingman Marsh was *Polygonum punctatum* (Table 3.3). Kenilworth Marsh contained the annuals *Impatiens capensis* and *Zizania aquatica* within its dominant species, while the natural marshes had *Impatiens capensis*, *Polygonum arifolium*, and *Polygonum sagittatum* occurring as dominants.

Table 3.3 The dominant cover species found across all years (2002, 2003, 2004) at each of the four tidal freshwater marsh study sites, based on least squares means of cover \pm SE. Within species, means with different letters indicate significant differences between sites. If no letters are shown, but a species was present in more than one site, there were no significant differences between sites. Means for the top 10 dominant species at each site are in boldface type. Dashed lines indicate that a particular species was not found at that site. Nomenclature is based on Brown and Brown (1984; 1992) and the USDA PLANTS Database (USDA 2005).

Species	Kingman Marsh	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
Acorus calamus	_	_	_	$\textbf{5.8} \pm \textbf{0.703}$
Hydrilla verticillata†	_	_	_	9.6 ± 1.38
Impatiens capensis•	0.012 ± 0.479^{b}	6.1 ± 0.722^{a}	$3.7 \pm \mathbf{1.17^a}$	$6.3\pm0.827^{\rm a}$
Juncus effusus*	1.8 ± 0.386	0.062 ± 0.581	_	0.42 ± 0.666
Leersia oryzoides	$0.22\pm0.665^{\text{c}}$	13 ± 1.00^{b}	$30 \pm \mathbf{1.62^a}$	$0.18\pm1.15^{\rm c}$
Lemna minor	—	_	_	$\boldsymbol{6.0\pm0.737}$
Ludwigia palustris	1.7 ± 0.414	0.0082 ± 0.624	0.058 ± 1.01	0.16 ± 0.716
Ludwigia peploides	0.79 ± 0.195	_	_	0.0079 ± 0.336
Lythrum salicaria†	3.9 ± 0.433^{a}	1.8 ± 0.653^{b}	2.7 ± 1.06^{ab}	_
Nuphar lutea*	0.38 ± 0.794^{b}	_	_	17 ± 1.37^{a}
Orontium aquaticum	_	_	_	$\textbf{3.2} \pm \textbf{0.339}$
Peltandra virginica*	7.7 ± 0.946^{b}	17 ± 1.42^{a}	11 ± 2.31^{ab}	14 ± 1.63^{a}
Phalaris arundinacea	_	$\textbf{3.8} \pm \textbf{0.456}$	$\textbf{3.8} \pm \textbf{0.739}$	_
Phragmites australis†	1.2 ± 1.12^{b}	17 ± 1.69^{a}	_	_
Pilea pumila●	0.00018 ± 0.246^{b}	0.16 ± 0.370^b	0.32 ± 0.600^{b}	$3.3\pm\mathbf{0.424^{a}}$
Polygonum arifolium $ullet$	_	0.83 ± 0.765^{b}	5.6 ± 1.24^{a}	9.2 ± 0.876^{a}
Polygonum punctatum•	0.59 ± 0.479^{b}	0.96 ± 0.722^{b}	9.9 ± 1.17^{a}	1.8 ± 0.827^{b}
Polygonum sagitattum•	0.00044 ± 0.290^{b}	0.26 ± 0.438^{b}	3.4 ± 0.709^{a}	2.7 ± 0.501^{a}
Salix nigra	3.2 ± 0.494^{a}	1.3 ± 0.744^{ab}	_	0.19 ± 0.852^{b}
Schoenoplectus fluviatilis	_	8.8 ± 0.915^{a}	_	0.74 ± 1.05^{b}

(continued)

Tab	le 3.3	(continued)
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Species	Kingman Lake	Kenilworth Marsh	Dueling Creek	Patuxent Marsh
Schoenoplectus tabernaemontani*	$\boldsymbol{0.89 \pm 0.247}$	0.032 ± 0.372	0.037 ± 0.603	0.31 ± 0.426
Sparganium eurycarpum	_	_	$\textbf{3.2} \pm \textbf{0.485}$	$\textbf{3.4} \pm \textbf{0.343}$
<i>Typha</i> spp.	$3.4 \pm \mathbf{0.852^b}$	18 ± 1.28^{a}	$3.7 \pm \mathbf{2.08^{b}}$	$3.4 \pm \mathbf{1.47^{b}}$
Zizania aquatica●	0.48 ± 0.355	1.6 ± 0.535	_	0.27 ± 0.612
No cover	73 ± 1.72^{a}	$6.3 \pm 2.59^{\circ}$	22 ± 4.19^{b}	15 ± 2.96^{bc}

Non-native species[†]

Planted at Kingman Marsh*

Annual species•

Discussion

The suitability of urban vs. non-urban reference sites

The vegetation parameters we measured indicate that the restored sites are more similar to Dueling Creek than to Patuxent Marsh. According to Sørensen's Quotient of Similarity, Kingman Marsh and Kenilworth Marsh were more similar to Dueling Creek (urban natural) than Patuxent Marsh (non-urban natural) in 2002 and 2004 (Table 3.2). The dominant species composition of the restored marshes was more similar to Dueling Creek than Patuxent Marsh (Table 3.3). Kingman and Kenilworth marshes had half as many dominant species in common with Patuxent Marsh as they did with Dueling Creek. Patuxent Marsh also did not contain *Lythrum salicaria* or *Phragmites australis*, both of which were found at Dueling Creek and the restored wetlands.

Ultimately the question becomes whether an appropriate reference site has been chosen for evaluating the success of a restoration project. Restoration projects will mature, but may never reach the desired full functionality of pristine natural wetlands (National Research Council 1992; Middleton 1999; Ehrenfeld 2000a; Kentula 2002). Warren et al. (2002) considered pre-disturbance conditions and some reference sites to be unattainable for their tidal marsh restoration program in Connecticut, but the vegetation functional goals they had set were accomplished given enough of a recovery period. This finding demonstrates that functions such as standing crop and species diversity may be returned to a system, but the system may not necessarily "look" like a natural wetland (Thom 2000). This finding supports what has happened at Kenilworth Marsh over the past ten years. The more degraded an ecosystem is, the less likely it is that all flora or

fauna will be restored (National Research Council 1992), that a restored urban ecosystem's flora and fauna will resemble a non-urban area's (Ehrenfeld 2000b), or that non-native species will be excluded. According to the theory of self-design, the environmental conditions at Kenilworth Marsh, which has a large influx of propagules due to its tidal regime, supported the species and individual seeds that were most likely to survive given the intensive surrounding land use, elevation, and degraded waters (Odum 1989; Mitsch et al. 1998). Some of these species happened to be non-natives.

It may be more appropriate to turn to older restored sites (e.g., Kenilworth Marsh), or natural urban sites (e.g., Dueling Creek), rather than natural non-urban references, to show the potential of young urban restoration projects (Baldwin 2004). Urban restorations often have to overcome many constraints to become established: contaminants, competing land uses, invasive species (National Research Council 1992), decreased water quality due to increases in storm water runoff, the presence of trash, and alteration of sediments and land, including habitat fragmentation (Ehrenfeld 2000b; Leck and Leck 2005). Things such as herbivory and hydrologic alteration can also affect wetland vegetation composition to the extent that urban wetlands may exhibit different vegetation types than wetlands in a non-urban setting (Baldwin 2004). Due to these factors acting alone or in combination, natural, non-urban wetland vegetation may be unattainable in urban wetlands; however, because urban wetlands experience similar urbanization pressures, natural urban wetland vegetation may be attained in restored urban wetlands (Baldwin 2004). Therefore, I am suggesting that urban restorations be evaluated against urban reference sites which represent the type of "natural" ecosystem that develops under the constraints imposed by the urban environment.

Performance curves can be used to compare natural marshes with restored sites to show their level of function over time (Kentula et al. 1992). Figure 3.7 is a hypothetical curve for this study's wetlands, which suggests that the urban marshes will never attain the species composition of Patuxent Marsh, the non-urban reference site with regard to vegetation structure.



Figure 3.7 Performance curve adapted from Kentula et al. (1992) for reference and restored marshes in this study.

Vegetation parameters of restored versus reference sites

Kingman Marsh was restored in 2000, with monitoring beginning immediately

after restoration. Neff (2002) found, as have others (Parikh and Gale 1998; Kellogg and

Bridgham 2002), that recently restored or created sites may attain cover, taxa density, and diversity similar to reference areas. Taxa density has even been found to be significantly higher at mitigation wetlands than natural wetlands (Magee et al. 1999). These gains may be lost however, as the restoration site matures. Kingman Marsh experienced decreases in cover, richness, and diversity after the first year following restoration (Neff 2002). By 2002, Kingman Marsh had significantly lower percent cover, taxa density, diversity, and evenness than the other sites. Enough time may not have been allotted for the analysis of Kingman's restoration. Sites are often not monitored long enough to gauge success (Kellogg and Bridgham 2002). Kingman Marsh had five years to develop and become similar to the reference sites, Dueling Creek and Patuxent Marsh, but it still had four times less cover and taxa, and about three times less diversity than the reference sites (Figure 3.1).

There are also large expanses of mud flats at Kingman Marsh (Table 3.3), which may be a result of too low of an elevation to support germination and growth (Hammerschlag et al. 2001; 2002; A. Baldwin, University of Maryland, personal communication). Flooding has been found to decrease seed germination and species diversity (Baldwin et al. 2001; Leck 2003; Chapters 2; 4). Sediment consolidation or erosion may have occurred after placement, resulting in a slightly lower elevation than was intended (Hammerschlag et al. 2001; 2002; A. Baldwin, University of Maryland, personal communication). Cornu and Sadro (2002) showed that consolidation accounted for much of the elevation loss in a marsh enhancement in Oregon.

Kenilworth Marsh, the ten-year-old restored marsh, was significantly different from the two reference sites in its taxa density, averaging about two species less than both

Dueling Creek and Patuxent Marsh, but was similar to the reference sites in cover, diversity, and evenness (Figure 3.1). In some ways, this suggests that Kenilworth Marsh is similar in structure and function (i.e., diversity and cover) to the reference marshes. Dueling Creek and Patuxent Marsh were not significantly different in cover, taxa density, diversity, or evenness, though they are located in separate watersheds with very different surrounding land uses (Figure 3.1).

Variation in species composition

All four sites shared *Peltandra virginica* and *Typha* spp. as dominant species, albeit in widely varying amounts (Table 3.3). *Peltandra virginica* was planted at Kingman Marsh during the restoration. It did not germinate widely in the seed bank study (Chapter 2), so it may need to be planted if it is a desired species (Neff and Baldwin 2005). Other species planted at Kingman Marsh which did not appear as dominant species in either reference site were *Juncus effusus* and *Schoenoplectus tabernaemontani*. *Lythrum salicaria*, an invasive species found at Dueling Creek, but not Patuxent Marsh, demonstrates that urban areas experience different conditions and that restoration sites in urban areas cannot be expected to mirror conditions of non-urban sites. Magee et al. (1999) found that wetlands located in developed areas had, on average, three more non-native species than wetlands in undeveloped areas.

Both Kingman Marsh and Kenilworth Marsh contained the invasive *Phragmites australis*. This species may, therefore, require management in restored areas. However, non-native species do contribute to the diversity and functioning of wetland systems (Kangas 2004). Kingman Marsh had only one dominant annual species, *Polygonum*

punctatum, while Dueling Creek and Patuxent Marsh contained *Impatiens capensis*, *Pilea pumila*, *Polygonum arifolium*, *Polygonum punctatum*, and *Polygonum sagittatum*. With the exception of *Polygonum arifolium*, Kingman Marsh contained each of these species, although at a low cover. If higher cover of these annual species is desired at levels found in reference marshes, seeding or an increase in sediment elevation to promote seedling germination may be necessary.

The percent cover of annual species was significantly different between restored and reference sites in 2002, but by 2004 there were no significant differences (Figure 3.5). This may be due to natural variation (Thom et al. 2002). Leck (1995) found that vegetation experienced significant variation year-to-year in a ten year study of a tidal freshwater marsh. The significant drop in annual species seen at Patuxent Marsh between 2002 and 2003 was likely due to inundation over a large part of the marsh (Chapter 4). A beaver dam was constructed on the site in mid-2002, which altered the hydrology of the system and the vegetation of the site. The presence of *Hydrilla verticillata* and *Lemna minor* at Patuxent Marsh increased the perennial, native, and non-native cover (Figures 3.5; 3.6).

Kingman Marsh had significantly less native cover than the other sites. It is unlikely that competitive displacement by *Lythrum salicaria* ($3.9\% \pm 0.433$) contributed greatly to the low native species cover in Kingman Marsh, because Dueling Creek ($2.7\% \pm 1.06$) had a comparable amount of this species, yet had significantly higher amounts of native cover than Kingman Marsh. Kenilworth Marsh, which had a high cover of *Phragmites australis*, had the highest cover of non-native species. With *Hydrilla verticillata* present, Patuxent Marsh also had high cover of non-natives.
Conclusion

If appropriate reference sites are not used, a determination of success is unlikely (Grayson et al. 1999). Urban systems like Kenilworth Marsh and Dueling Creek have similar cover, richness, and diversity to the non-urban reference site, Patuxent Marsh, yet they contain non-native species. The surrounding land use in intensive use areas may contribute to the supply of non-native species in wetlands. Magee et al. (1999) found significantly more non-native/invasive species associated with intense land use than with wetlands surrounded by undeveloped land. This may be due to alteration of dispersal patterns, or altered hydrology, geomorphology, or disturbance regime (Ehrenfeld 2000b; Baldwin 2004). Simply because an urban restoration site contains non-natives should not preclude it as a failure or as a restoration site. These conditions originate from surrounding land uses that are not likely to diminish. The restored wetlands will be functional, but not exact replicas of the original (Ehrenfeld 2000a; Thom et al. 2002; Warren et al. 2002). Which is more important, to have a functioning system, or one which requires constant upkeep in terms of plantings, dredging, herbicide applications, etc.? The National Research Council (1992) says that a goal of restoration should be a self-sustaining system, clearly functioning on its own, which Kenilworth Marsh has achieved and Kingman Marsh may achieve with time.

Chapter 4: Vegetation and edaphic responses to a beaver dam impoundment in a tidal freshwater marsh

Introduction

Prolonged flooding in wetlands has been shown to decrease seed germination (Baldwin and DeRico 1999; Peterson and Baldwin 2004) and alter vegetation structure (Baldwin et al. 2001). Beaver activity, which is often associated with flooding, has increased in the United States over the past several decades (Syphard and Garcia 2001; Foster et al. 2002; Handwerk 2004), possibly leading to an increase in impounded areas. Flooding caused by structurally altered watercourses may alter vegetation at these sites. Several vegetation studies have been conducted in areas impacted by beaver activity, mostly occurring along streams and rivers (Johnston and Naiman 1990; Syphard and Garcia 2001; Perkins and Wilson 2005).

Wetland systems have not been extensively studied for beaver impacts, particularly with regard to flooding effects on vegetation. No studies dealing with tidal freshwater wetlands were found during my literature review. Studies have been conducted correlating flooding with lower germination rates and/or altered vegetation composition (Gough and Grace 1998; Cornu and Sadro 2002). Lower sediment elevation (essentially, equivalent to higher water level) was found to significantly reduce stem density and biomass of *Panicum hemitomon* in a subsidence simulation study by McKee and Mendelssohn (1989). My study was conducted post-dam construction, but an analysis of adjacent transect data collected during another study (Chapter 3) in this wetland during pre-construction, construction, and post-construction of the dam revealed

that the dam had an influence on the vegetation in the wetland overall. Cover of *Impatiens capensis* and *Polygonum arifolium*, richness, and total percent cover was similar across the wetland (with the exception of transect 5) before the dam was built in early 2002, but diverged after the dam began impounding the wetland system behind it (late 2002) (Chapter 3).

The objective of this study was to examine the effect of flooding on a new impoundment created by a beaver dam on tidal freshwater marsh vegetation, and on soil and water parameters through monitoring of plots in impounded and non-impounded areas. Because of the inhibitory effects of flooding on vegetation growth (Gough and Grace 1998; Baldwin and DeRico 1999; Baldwin et al. 2001; Cornu and Sadro 2002; Peterson and Baldwin 2004), I hypothesized that impounded areas would have a lower percent cover of emergent vegetation, lower taxa density, different species composition, and lower biomass than the non-impounded area. I also expected that between years, the two areas would grow more dissimilar with regard to vegetation structure as long as the dam was not compromised and that impounded areas would be flooded 100% of the time. The dissimilarity would occur as a result of the constant water in the impoundment selecting certain species for germination and reducing growth rates of those species due to water stress. The expectation that the impoundment would be flooded 100% of the time arose out of current field conditions when this study was designed.

Methods

Study site

Patuxent Wetland Park contains a tidal freshwater marsh located southeast of Washington, D.C. along a small tidal channel of the Patuxent River (Neff, 2002) (Figures 2.1; 4.1). This marsh served as a reference site for seed bank studies and vegetation monitoring conducted at restored marshes in Washington, D.C. (Chapters 2; 3). During mid-2002, beavers (*Castor canadensis*) began building a dam in the wetland (Figure 4.1). The dam, made of sticks and logs, extended approximately seven meters across a small tidal channel of the Patuxent River named Mill Creek. It began at the upland forested edge of the wetland before it crossed the creek and continued into the middle of the marsh, where it was made of mud, vegetative stalks, and rhizomes. While I realize that the road (MD Rte 4) bisecting Patuxent Marsh may have an influence on the hydrology of the site, it was there a sufficient amount of time prior to the construction of the beaver dam (years) for me to evaluate and analyze the effect of the dam on the marsh.

The area north (upstream) of the dam became impounded, creating a pond behind the dam. The vegetation was sparse here and the area was continuously flooded throughout the study (personal observation). The area south of the dam remained tidally influenced and bordered the creek that fed the wetland. The south area had small rivulets running from the impoundment to the creek and many annual seedlings, such as *Impatiens capensis, Polygonum arifolium* and *Polygonum punctatum*, which were sprouting in May 2003. The dam, where it extended into the wetland, ranged from 75 cm to 100 cm wide and 23 cm tall relative to the adjacent marsh. The beavers continued to



Figure 4.1 Study set-up in Patuxent Wetland Park occurred above and below the beaver dam along Mill Creek, a small tidal channel off of the Patuxent River.

extend the dam throughout this study and after, when the dam was examined during subsequent visits.

Study set-up

Twelve plots, six in the impounded area and six in the non-flooded area, were established in early August 2003. They were oriented in lines parallel to the dam, and at least 10 meters from it. They were placed outside of the direct physical influence of the beavers, i.e., where plants had obviously been influenced by herbivory or digging. The plots were more than six meters apart along the dam and at least four meters apart in 2-D space. Placement of plots in the non-flooded area avoided rivulets running through this area. Each plot had one tall marker pole and six shorter poles that delineated the outline of the plot, which was 1 m x 2 m. The shorter side of the plots faced the dam. Sampling of vegetation, soils, leaf area index (LAI), light, and water quality was conducted from 12-14 August 2003 and from 27-28 July 2004. Biomass was collected at its approximate peak on 02 September 2003 and 07 September 2004.

Vegetation sampling was conducted within a 1 m x 2 m quadrat made of PVC which was laid on the top of the six poles marking each plot. The quadrat had wooden dowels making up 32 cells, each 25 cm x 25 cm. This was done to determine frequency of vegetation and to make estimating cover more consistent, as each cell was approximately 3%. Two people monitored vegetation, each looking at half the plot. A species was identified and its frequency (presence/absence in a cell) determined. Two methods for cover were used. First, cover of each species was decided by both people according to the following cover classes: <1%, 1-5%, 5-25%, 25-50%, 50-75%, 75-90%

and > 90%. The upper end of the cover class was recorded. Second, the actual cover of each species was determined by each person on their half of the plot, then averaged to get the percentage for the whole plot using the following cover classes: <1%, 1-15% in increments of one, and >15% in increments of five. The cover had to equal or exceed 100%. Two cover methods were used to explore the effectiveness of the different methods; in this chapter, only the actual cover was used in analyses. The average canopy height of each species was estimated, as was the typical canopy height of the entire plot. Cover and frequency of open water or mudflats and detritus were also recorded.

For biomass harvesting, a 1 m² quadrat was placed adjacent to the plots and all standing aboveground biomass was cut or broken off at the surface and placed in separate plastic bags for each plot. Care was taken not to collect biomass from rivulets in non-flooded areas. The plastic bags were transported back to the University of Maryland and placed in a growth chamber at 4°C until they could be transferred to paper bags and put in a 26.7°C chamber (to prevent rotting). Samples were then dried in a drying oven for 48 hours at 80°C and allowed to cool for approximately 20 minutes. They were then placed in a tared plastic weigh pan and weighed on a Mettler PMK34-K scale (0.1 g resolution).

Two soil cores were collected in each plot using a bulb corer (440 cm³ each) during vegetation sampling. These two cores were combined, placed in a labeled plastic bag, and samples were transported back to the University of Maryland. Aluminum pans were labeled with plot number, weighed, tared, and each sample was placed in a pan and they were re-weighed. The plastic bags were scraped out to ensure the full sample was obtained and large organic matter (e.g., sticks, rhizomes) was removed. After drying the samples at 80°C until a constant mass was achieved, they were again weighed. The

following calculation was used to determine soil moisture content: $M_d = (W_{ws} - W_{ds})/W_{ds}$ x 100%, where M_d is the percent moisture, W_{ws} is the mass of wet soil (g) and W_{ds} is the mass of dry soil (g). Bulk density was determined by: $\rho = W_{ds}/V$, where ρ is the bulk density (g/cm³), W_{ds} is the mass of dry soil (g) and V is the total sample volume (cm³).

Organic matter was determined using the Loss-On-Ignition Method (LOI) (Sparks 1996). Oven-dried soils were ground into a fine powder with a mortar and pestle and kept in a dessicator until final processing. Crucibles were labeled in pencil on the bottom, heated at 400°C for two hours in a muffle furnace, and allowed to cool in a dessicator. Their weight was determined to the nearest 0.1 mg. Each crucible was then tared, one to three grams of soil was added, and the mass recorded. The samples were heated for 24 hours at 105° C, cooled in a dessicator, and their mass determined. The weight of the oven dried sample was calculated by subtraction. The samples were then ignited in a 400°C muffle furnace for 16 hours to burn off organic matter, cooled in a dessicator, and reweighed. The weight of the ignited sample was determined by subtraction. The percent of LOI was calculated by: $(mass_{105} - mass_{400})/mass_{105} \times 100$.

The water depth in each plot was recorded during vegetation sampling, and could change depending on the time of measurement relative to the tide. These measurements did correspond closely with water levels measured by datalogging water level recorders. Conductivity (μ S), temperature, and salinity were measured with a YSI 30 portable meter. Leaf area index (LAI) was measured with the LAI-2000 Plant Canopy Analyzer (LI-COR, Inc.). One reading was taken above the canopy and two below. Light levels were measured with the LAI 250 Light Meter (LI-COR, Inc.). Readings were taken above and below the canopy and recorded, as were the time of day and sky conditions.

Redox potential was measured with an Orion 250A meter and a RadioShack Digital Multimeter. Six platinum tipped electrodes, to have replicate samples, were randomly placed in the plots at a depth of 10 cm. The meter's probe was vented, placed in the soil, and the mV measurements were taken and recorded. The six measurements for each plot were averaged and 244 mV were added to each to determine E_h (redox potential). The E_H was not adjusted for pH; however, previous measurements across all sites were found to fall within 1 pH unit in 2000 and 2001 (pH range 6.23-7.06) (Neff 2002).

Datalogging water level recorders

Ecotone water level recorders (Remote Data Systems, Inc., Whiteville, North Carolina) were used in this study to record water levels and flooding duration above and below the dam at set intervals throughout the day. A water level recorder was installed in the impounded area in March 2001 during a related study and will hereafter be referred to as the impounded well. The water level recorder installed south (downstream) of the dam was placed on 09 April 2004 and will hereafter be referred to as the non-impounded well. The elevation of plots relative to wells was surveyed on 25 March 2004 with a Wild NA30 Automatic Level (Leica). Both wells were placed at lower elevations than the plots.

The water level recorders were installed in the wetland in holes dug approximately 120 cm deep. The water level probe sat inside a 7.6 cm diameter slotted PVC pipe screened to about 1m above the soil surface. The recorders' capacitance probe had a data logger attached to the top that recorded water levels at 15 minute intervals.

The top of the water level recorder containing the data logger was approximately 92 cm above the sediment. Data were downloaded with a Palm portable operating system using Ecotone software, the manufacturer of the water level recorders. Due to well malfunctioning, usable data were downloaded when both wells were in operation from 09 April – 23 July 2004.

Data analysis

Vegetation, water quality, and soil characteristics were analyzed with an ANOVA (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Mixed) to detect differences in these parameters across years and treatments. A critical level of P = 0.05 assigned significance. *Lemna minor*, a floating aquatic species, covered many impounded areas at approximately 100% cover, so I calculated vegetation parameters including and excluding it. All parameters met the assumptions of ANOVA and normality, with the exception of biomass which was log transformed then detransformed for presentation. Significance determinations for plot elevation, flooding duration, and water level differences between the wells were made with two sample t-tests.

In order to determine if soil and water parameters were correlated, a correlation analysis was carried out (SAS version 8.2 for Windows, SAS Institute, Cary, NC, Proc Corr). This analysis determined the amount of association (Sokal and Rohlf 1987) between the measured parameters: percent moisture, bulk density, organic matter, E_h , conductivity, temperature, salinity, and water depth.

The species evenness index, Pielou's J, was calculated as in Chapters 2 and 3 for each plot. The percentage importance (p_i) was based on the relative cover and S was the

species richness per plot (Peet 1974; McCune and Grace 2002). Dominant species were determined based on least squares means of cover for each species in each year. Unidentified dicots and monocots were not included for consideration in the dominant species list.

Results

Vegetation response

In the non-impounded zone, biomass was significantly higher and percent open canopy significantly lower than in the impounded zone (Table 4.1; Figure 4.2). Biomass was approximately 145 g in the non-impounded zone in 2003, compared to 40 g in the impounded zone, while the percent open canopy in the impounded area was 65% versus 23% in the non-impounded area in 2004 (Figure 4.2). The average height of vegetation in the non-impounded area increased from 2003 to 2004 (approximately 0.8 m to 1.1 m), and decreased in the impounded area (approximately 1.2 m to 0.9 m) during this time, resulting in a significant treatment by year interaction (Table 4.1; Figure 4.2). Leaf area index, the area of foliage (i.e., stems and leaves) per unit area of ground, was similar between the two sites in 2003 (Figure 4.2). In the non-impounded zone, leaf area increased significantly by 2004 to approximately 2.7 versus 1.3 in the impounded zone, again resulting in a significant treatment by year interaction (Table 4.1; Figure 4.2).

Lemna minor comprised much of the total percent cover found in the impounded area, especially in 2004 (75%) (Figure 4.3). Including it in calculations of percent cover did not result in significant findings (Table 4.1). However, when this floating aquatic

Table 4.1 Results of the analysis of variance examining vegetation, soil, and water characteristics of two treatment regimes, impounded and non-impounded, in a tidal freshwater marsh for years 2003 and 2004. The results for vegetative formulas with and without *Lemna minor*, a floating species, are included to show differences that this non-emergent causes in vegetative composition. The Fvalue is reported. The numerator df was 1, while the denominator df was 20 in each case, with the exception of vegetation height, which was 1 and 19 respectively.

Significance noted as ⁺0.05-0.10; *<0.05; **<0.01; ***<0.001; ***<0.0001.

Parameter	Location	Year	Location x Year
Vegetation:			
Vegetation height	3.84^{+}	0.146	16.6***
LAI	13.9**	12.0**	5.20*
Percent open canopy	12.0**	2.68	2.22
Biomass	30.5****	0.395	2.14
With Lemna minor included			
Total Cover	0 373	0.0761	1.01
Taxa Density	10.8**	4.57*	0.564
Pielou's J (evenness)	3.37^{+}	1.62	6.45*
Shannon-Weiner Diversity	3.95 ⁺	0.251	3.40^{+}
Without Lemna minor			
Total Cover	23.3****	0.442	4.98*
Taxa Density	13.2**	3.51 ⁺	0.219
Pielou's J (evenness)	6.45*	0.0300	2.07
Shannon-Weiner Diversity	3.38 ⁺	1.08	0.507
Soils:			
Bulk density	4.91*	16.4***	0.374
Percent moisture	2.43	11.8**	0.343
Percent organic matter	3.33^{+}	3.14^{+}	0.0217
Redox potential (E _h)	3.14	8.30**	2.91
Water:			
<u>Conductivity</u>	0.258	76 2****	1 26+
Solinity	0.230	20.5 ^{****}	4.20
	1.23	11.2^{++}	1.23
Iemperature	1/.3***	2.68	4.19
Water depth	35.5****	1.54	0.117



Figure 4.2 Vegetation height, leaf area index, canopy openness, and biomass measured in impounded and non-impounded areas of a beaver dam-impacted tidal freshwater marsh on the Patuxent River. Figures represent vegetation parameters not including *Lemna minor*.

species is removed from calculations of percent cover, the two areas are found to be significantly different across years, resulting in a significant treatment by year interaction (Table 4.1; Figure 4.3). Due to these findings, and because *Lemna minor* is not an emergent species, I will mainly concentrate on the figures without *Lemna minor*. Taxa density was significantly higher, by about two times, in the non-impounded area (Table 4.1; Figure 4.3). Evenness was significantly higher in the impounded zone in 2003 (0.80 versus 0.60), but the two areas converged to equal evenness in 2004 (0.70) (Table 4.1; Figure 4.3). Shannon-Weiner diversity did not differ significantly between impounded areas (P = < 0.10) (Table 4.1; Figure 4.3).

Lemna minor and *Peltandra virginica* were found to have significantly higher cover in the impoundment than the non-impoundment, while *Polygonum arifolium*, *Impatiens capensis*, and *Leersia oryzoides* had significantly greater cover in the nonimpounded area (Table 4.2; Table 4.3). Dominant species found in impounded and nonimpounded areas did not differ greatly between 2003 and 2004, with *Lemna minor* and *Polygonum arifolium* remaining dominant in their respective zones (Table 4.2). *Sparganium eurycarpum* was a dominant throughout the wetland in both years, ranging from approximately 4% cover to 17% cover. *Typha latifolia* cover was significantly lower in 2004 than 2003, moving from a dominant in both zones to barely present in either in 2004 (Table 4.2; Table 4.3). Dominant plant species calculated based on frequency varied little when compared with the dominant species based on percent cover (Figure 4.4). The exception was the 2004 non-impounded area, where *Lemna minor* was found in each cell sampled for frequency, but did not have 100% cover overall. In the



Figure 4.3 Vegetation cover, taxa density, species evenness, and Shannon-Weiner diversity measured in impounded and non-impounded areas of a beaver dam-impacted tidal freshwater marsh on the Patuxent River. Graphs on the left represent least squares means calculated without *Lemna minor*, while those on the right were calculated with *Lemna minor*.

Table 4.2 All species found in impounded and non-impounded areas of Patuxent, a tidal freshwater marsh. Numbers reported are percent cover least squares means \pm SE. Numbers in bold represent the top five dominant species each year in the impounded and non-impounded areas. "–" means a species was not found at this location; SEs for these locations are the same as for least squares means whose species were found in that location. Flooded plots were flooded 100% \pm 0 during 2003 and 2004, while non-flooded plots were flooded 42.4% \pm 3.82 of the time.

	2003			2004				
	Impor	unded	Non-im	pounded	Impo	unded	Non-im	pounded
Species	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Acorus calamus	_		_		1.2	0.583	_	
Amaranthus cannabinus	-		_		-		1.1	0.400
Bidens spp.	-		_		0.17	0.242	1.4	0.242
Boehmeria cylindrica	-		_		0.083	0.0417	-	
Cuscuta gronovii	-		_		0.17	0.118	0.17	0.118
Cyperaceae	-		_		_		0.17	0.0833
Cyperus spp.	0.33	0.167	_		_		_	
Galium tinctorium	-		_		0.083	0.156	0.58	0.156
Hypericum boreale	-		0.17	0.0833	_		_	
Impatiens capensis	-		7.8	1.86	0.50	1.86	4.2	1.86
Leersia oryzoides	0.50	6.29	8.3	6.29	1.2	6.29	22	6.29
Lemna minor	52	7.03	0.083	7.03	75	7.03	0.50	7.03
Ludwigia palustris	-		_		0.33	0.0833	_	
Mikania scandens	-		6.7	3.34	0.17	3.34	1.3	3.34
Murdannia keisak	-		_		0.17	0.119	0.25	0.119
Nuphar lutea	6.7	2.46	_		5.5	2.46	-	
Peltandra virginica	27	4.02	2.3	4.02	22	4.02	4.3	4.02
Pilea pumila	-		0.92	2.27	0.33	2.27	8.5	2.27
Polygonum arifolium	-		44	8.17	1.0	8.17	26	8.17
Polygonum punctatum	-		0.17	5.63	-		18	5.63
Polygonum sagitattum	_		_		_		0.083	0.0417
Polygonum spp.	_		6.5	2.87	_		0.50	2.87

	2	003			20	04	
Impor	unded	Non-im	pounded	Impo	unded	Non-im	pounded
Mean	SE	Mean	SE	Mean	SE	Mean	SE
0.83	0.646	_		1.1	0.646	_	
7.0	3.93	4.5	3.93	0.17	3.93	7.7	3.93
_		1.6	1.13	_		3.4	1.13
0.83	0.449	_		0.33	0.449	_	
17	7.03	8.3	7.03	3.8	7.03	17	7.03
11	3.64	8.9	3.64	_		1.4	3.64
_		_		0.083	0.0589	0.083	0.0589
_		0.17	0.0932	_		0.083	0.0932
	Impor Mean 0.83 7.0 - 0.83 17 11 - -	2 Impounded Mean SE 0.83 0.646 7.0 3.93 - 0.83 0.449 17 7.03 11 3.64 - - - - -	2003 Impounded Non-im Mean SE Mean 0.83 0.646 - 7.0 3.93 4.5 - 1.6 0.83 0.449 17 7.03 8.3 11 3.64 8.9 - - 0.17	2003 Impounded Non-impounded Mean SE Mean SE 0.83 0.646 - - 7.0 3.93 4.5 3.93 - 1.6 1.13 0.83 0.449 - 17 7.03 8.3 7.03 11 3.64 8.9 3.64 - - - - - 0.17 0.0932 -	$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$	$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$	$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$

Table 4.2 (continued)

Table 4.3 Results of the analysis of variance for percent cover of individual species found in a tidal freshwater marsh for years 2003 and 2004. The Fvalue is reported. The numerator df was 1, while the denominator df was 20 in each case. Significance noted as $^+0.05$ -0.10; *<0.05; **<0.01; ***<0.001; ***<0.0001.

Species	Treatment	Year	Treatment x Year
Acorus calamus	1.00	1.00	1.00
Amaranthus cannabinus	1.83	1.83	1.83
Bidens spp.	6.66*	10.7**	6.66*
Boehmeria cylindrica	1.00	1.00	1.00
Cuscuta gronovii	0.00	2.00	0.00
Cyperaceae	1.00	1.00	1.00
Cyperus spp.	1.00	1.00	1.00
Galium tinctorium	2.57	4.57*	2.57
Hypericum boreale	1.00	1.00	1.00
Impatiens capensis	9.51**	0.721	1.25
Leersia oryzoides	5.34*	1.40	1.16
Lemna minor	81.4****	2.65	2.46
Ludwigia palustris	4.00^{+}	4.00^{+}	4.00^{+}
Mikania scandens	1.34	0.617	0.698
Murdannia keisak	0.122	3.05^{+}	0.122
Nuphar lutea	6.13*	0.0563	0.0563
Peltandra virginica	27.7****	0.172	0.832
Pilea pumila	3.99^{+}	3.03^{+}	2.54
Polygonum arifolium	17.6***	1.17	1.45
Polygonum punctatum	2.58	2.49	2.49
Polygonum sagittatum	1.00	1.00	1.00
Polygonum spp.	1.48	1.09	1.09
Pontederia cordata	2.20	0.0375	0.0375
Sagittaria latifolia	0.405	0.218	1.62
Schoenoplectus fluviatilis	4.92*	0.662	0.662
Schoenoplectus tabernaemontani	1.69	0.310	0.310
Sparganium eurycarpum	0.126	0.0809	2.37
Typha latifolia	0.00837	6.45*	0.231
Unidentified dicot	0.00	2.00	0.00
Zizania aquatica	1.80	0.200	0.200



Figure 4.4 Frequency of occurrence of species found in impounded and non-impounded areas of Patuxent, a tidal freshwater marsh. Species acronyms are listed in Appendix A. Numbers reported are percent frequency least squares means \pm SE. The greatest percent frequency that a species could occur in any one plot was 100%, based on a species occurring in 32 out of 32 cells. Flooded plots were flooded 100% \pm 0 during 2003 and 2004, while non-flooded plots were flooded 42.4% \pm 3.82 of the time.

2004 non-impoundment, *Leersia oryzoides* was the second most dominant based on frequency and *Polygonum arifolium* was third most dominant, but these roles were reversed in cover findings.

Environmental variables

Water depth was significantly lower in non-impounded zones than impounded zones across years (Tables 4.1; 4.4). The impounded areas' average water depth did not significantly change between 2003 and 2004, while non-impounded areas increased depth by two and a half times (Table 4.4). Conductivity significantly decreased from 2003 to 2004 in both areas (Table 4.4). Salinity decreased significantly in the impounded area between years, but all measurements were well within freshwater limits (Tables 4.1; 4.4). Water temperature was significantly higher in non-impounded than impounded zones, but like salinity, there was only a small difference in measurements. Elevation of the impounded area, relative to the well, was $0.5 \text{ m} \pm 0.02$, while the non-impoundment was not significantly different at an elevation of $0.4 \text{ m} \pm 0.001$ (P=0.0546).

Bulk density differed significantly (but very little in absolute terms) between impounded and non-impounded areas and also differed significantly between years (Table 4.1). Percent moisture was significantly higher in 2003 than 2004 for both impounded and non-impounded zones. Percent organic matter was greater each year in the non-impounded area, but not to a significant extent. E_h differed significantly between years, with 2003 having a higher soil redox potential.

Water depth was negatively associated with percent organic matter and redox potential (E_h) (Table 4.5). As water depth increased from 2003 to 2004, organic matter

Table 4.4 Soil and water quality parameters collected at both flooded and non-flooded areas of Patuxent Marsh with significant effects based on an alpha level of P<0.05. Numbers presented are least squares means \pm SE. Flooded plots were flooded 100% \pm 0 during 2003 and 2004, while non-flooded plots were flooded 42.4% \pm 3.82 of the time.

Parameter	Non-impounded 2003	Impounded 2003	Non-impounded 2004	Impounded 2004
<u>Soils:</u> Bulk density (g/cm ³)	0.12 ± 0.0218	0.15 ± 0.0218	0.19 ± 0.0218	0.26 ± 0.0218
Percent moisture	601 ± 51.12	551 ± 51.12	455 ± 51.12	346 ± 51.12
Percent organic matter	41 ± 3.80	34 ± 3.80	34 ± 3.80	27 ± 3.80
Redox (E _h)	134 ± 19.36	132 ± 19.36	111 ± 19.36	43.5 ± 19.36
<u>Water:</u> Conductivity (μS)	264 ± 26.76	332 ± 26.76	182 ± 26.76	140 ± 26.76
Salinity (ppt)	0.13 ± 0.0149	0.17 ± 0.0149	0.10 ± 0.0149	0.10 ± 0.0149
Temperature (°C)	25 ± 0.244	24 ± 0.244	25 ± 0.244	24 ± 0.244
Water depth (m)	0.058 ± 0.0537	0.40 ± 0.0537	0.14 ± 0.0537	0.44 ± 0.0537

	Percent moisture	Bulk density	Organic matter	Redox potential	Conductivity	Temperature	Salinity	Water depth
Percent moisture	1	-0.92****	0.90****	0.47*	0.50*	-0.03	0.28	-0.61**
Bulk density		1	-0.87****	-0.49*	-0.50*	-0.01	-0.27	0.60**
Organic matter			1	0.38 ⁺	0.28	0.19	0.07	-0.71****
Redox potential				1	0.39 ⁺	0.04	0.26	-0.58**
Conductivity					1	-0.30	0.84****	-0.04
Temperature						1	-0.15	-0.55**
Salinity							1	0.12
Water depth								1

Table 4.5 Correlation matrix for Patuxent Marsh soil and water measurements. Values are Pearson's r correlation coefficients. Significance noted as ⁺0.05-0.10; *<0.05; **<0.01; ***<0.001; ****<0.0001.

and redox potential decreased (Table 4.4). Water depth was also negatively associated with percent moisture and temperature (Table 4.5). Water depth was positively associated with bulk density; both increased between years (Tables 4.4; 4.5). Other strong negative associations included bulk density with percent moisture, organic matter, conductivity, and E_h (Table 4.5). Percent moisture had strong positive associations with organic matter, redox potential, and conductivity (Table 4.5). Conductivity and salinity also had a strong positive association.

Discussion

Flooding effects on vegetation

The significantly lower biomass and taxa density found in impounded areas supports studies linking flooding with increased stress on plants (McKee and Mendelssohn 1989; Gough and Grace 1998; Casanova and Brock 2000; Baldwin et al. 2001; McHugh and Dighton 2004), which leads to lower germination rates, species diversity (Baldwin et al. 2001; Leck 2003) and primary productivity (Mitsch and Gosselink 1993; Larmola et al. 2004). Percent cover with *Lemna minor* did not vary significantly between impounded and non-impounded areas. Without it, however, both the year and treatment interacted to produce a significant effect. The impounded area had lower total cover than the non-impounded area in both years, but the percent cover in the impoundment dropped approximately 30% between 2003 and 2004. This decrease may have been due to a number of factors: the disappearance of *Typha latifolia* in 2004, which was a dominant in 2003; increased muskrat herbivory (I observed more muskrat mounds in 2004 than 2003); and observed continued inhibition of vegetation growth and seedling

recruitment due to permanent flooding. Casanova and Brock (2000) and Baldwin et al. (2001) found continuous flooding produced the least amount of biomass as compared with no and intermittent flooding. The impoundment in my study was flooded 100% of the time.

Lemna minor provided the largest percent cover of any species in the impounded area in 2003 and 2004. The most likely cause for this was that *Lemna minor*, a floating aquatic, likely took advantage of the continuously flooded conditions. This species rapidly spreads seed and is shade intolerant (USDA PLANTS 2005); therefore, the impoundment likely provided perfect growing conditions with large expanses of open canopy, the low leaf area of emergent plants, and lack of tidal action. *Lemna minor* also affected the vegetation evenness index in the impoundment by its sheer density. Evenness decreased sharply in the impoundment between 2003 and 2004 when this species was included in calculations because it was found in high abundance in these areas. Without *Lemna minor* in calculations, the non-impoundment increased in evenness between years.

Polygonum arifolium, dominant in non-impounded areas, was found in small amounts in the impounded zone. My findings support those of Baldwin (2004) and Baldwin et al. (2001), who suggest that the lack of *Polygonum arifolium* and other annual species in the impoundment were due to higher than normal water levels. *Typha latifolia*, a dominant species in both zones in 2003, was not found in the impounded zone and was not a dominant in the non-impounded area in 2004. *Typha latifolia*'s decline may be due to the presence of rather large numbers of muskrat, *Ondatra zibethicus* (personal observation based on their mounds), which often feed on this species and do not range far from their homes (Barnes 1991), potentially causing eat-outs. Connors et al. (2000) found

that muskrat decreased biomass of *Typha angustifolia* in tidal freshwater marshes along the Hudson River, New York.

Height of vegetation in the non-impounded zone increased between years, while vegetation in the impoundment decreased in height. This difference may have been due to the continuous flooding present in the impounded zone, which caused a hypoxic root zone that resulted in reduced growth rate (Faulkner et al. 1989). My finding is in contrast to McKee and Mendelsshon (1989), Kirkman and Sharitz (1993), and Howard and Mendelssohn (1995), who found higher leaf and plant heights when plants were subjected to inundation (13-15 cm). *Typha latifolia*, a dominant in the impoundment in 2003, but absent in 2004, likely affected the height disparity expressed between the two zones. It is normally one of the taller species in this wetland, so its absence decreased overall height measurements per plot. The large cover of *Polygonum arifolium* in the non-impounded area grew on top of other species and itself to create the height measured in 2004 (personal observation).

Leaf area index increased greatly between years in the non-impounded area, possibly due to lower flooding stress than was found in the impounded zone, where leaf area increased a small amount. Shifts in the plant structure between years (e.g., less *Polygonum arifolium* cover in 2004 than in 2003) may also have altered leaf area index. Pezeshki et al. (1998) found leaf area to be significantly lower in *Salix nigra* cuttings in continuously flooded conditions than leaf area measurements in controls receiving adequate water and drainage. Also, the non-impounded area had higher biomass measurements so that plants in this zone would need to grow larger, taller, and potentially faster in order to compete for light.

Flooding effects on soil parameters

Bulk density increased in the impoundment from 2003 to 2004, but remained steady in the non-impounded zone. Alternatively, Bruland and Richardson (2004) found bulk density increased from wet to dry zones in their study of created wetlands in Virginia. The strong positive association between bulk density and water depth could be a result of increased water volume placing more weight on the sediments, compacting them (A. Baldwin, University of Maryland, personal communication). This may potentially occur if water cannot enter the substrate and instead surrounds it, placing pressure on the sediments, causing them to compress (A. Baldwin, University of Maryland, personal communication). A prolonged duration of standing water suggests that soil compaction may have occurred (USEPA 2003; Schwab et al. 2004). The lower bulk densities found in the non-impounded zone each year may be due to the continuous tidal flushing bringing in sediments and moving them around so that they cannot become compacted; likewise, I would expect an increase in the bulk density in the impoundment due to the lack of tidal flushing in the sediments (J. Perry, III, Virginia Institute of Marine Science, personal communication) due to the muted hydrologic regime. In addition, hydrology may have altered (reduced) the conditions to an unsuitable level for burrowing organisms. Compaction may also explain the negative association of water depth with percent moisture; the less pore spaces there are to hold water, the lower the percent moisture of the soil and the higher the bulk density. On the other hand, organic matter is positively associated with percent moisture due to the large number of pore spaces organic soils hold.

Redox potential has been shown to decrease with increasing soil moisture (van Oorschot et al. 2000; Pezeshki 2001). I actually found a positive association between these two variables (Table 4.5); as soil moisture decreased, redox potential also decreased (Table 4.4). Faulkner et al. (1989) report that soil oxygen levels and redox potential decrease with increasing water table levels. My findings support this statement; as the redox potential decreased, our water depth increased (Table 4.4), and the two had a significant negative association (Table 4.5). Pezeshki (2001) predicts that low soil redox potential may lead to a lower photosynthetic rate; this may also contribute to the lower leaf area measured in the impoundment and discussed above. Species must develop mechanisms to deal with saturated or inundated conditions; therefore, reducing soils produce characteristic vegetation for these conditions (Faulkner et al. 1989).

Water quality parameters and effects of the impoundment

Conductivity and salinity both significantly decreased between 2003 and 2004, but these probably did not have a biologically significant effect because they decreased very little in absolute terms. The strong positive association these parameters shared was not surprising since salinity values were derived from conductivity and temperature measurements. Salinity levels were well within freshwater limits of < 0.5 ppt, and were not expected to have effects on plant community structure. Temperature and water depth were significantly affected by the impoundment; impounded areas had lower temperatures possibly due to deeper waters receiving less sunlight and not being able to heat the water. Margolis et al. (2001) found similar temperature results during summer stream measurements above and below beaver dam impoundments in two Mid-Atlantic

streams. Depth was less important than frequency and duration of flooding in determining plant community composition in Australian temporary wetland seed bank studies (Casanova and Brock 2000). The constant flooding that occurred in the impounded area likely had the largest effect on plant community structure in this wetland.

Conclusion

The beaver dam constructed in Patuxent Marsh during early 2002 began having an effect on vegetation almost immediately, with apparent changes observed in the vegetation transects monitored during a concurrent study (Chapter 3). When beaver flooded this wetland, they created a "wetter wetland" (Johnston 2001) that caused significant rapid changes in vegetation and hydroperiod. The vegetation in the impounded area produced less biomass, and had less percent cover and taxa density than vegetation in the non-impounded area. Impounded areas were flooded 100% of the time during this study, likely causing the vegetation changes I observed and altering the plant community structure behind the dam. The beaver dam may have influenced the non-impounded area by the small channels and rivulets I observed running through that zone. By cutting off the daily tidal action to sections of this marsh, the beaver, through their dam, had a large effect on their environs. As they continue to build the dam out into the wetland, they will continue to alter the plant community structure and hydrology of this site, potentially permanently flooding a larger area of the wetland.

Chapter 5: Conclusions

The restoration of wetland ecosystems often will not be successful by following a series of "how-to" steps (Hilderbrand et al. 2005). Monitoring the seed bank before and after restoration will assist in learning which species may become established at a site, may warn of the possibility of invasion by non-natives, and will give an indication of species missing from the seed bank that may need to be planted. Kingman Marsh, the young restored site, quickly developed a seed bank that was comparable to natural sites in abundance of seedlings, taxa density, and evenness. A species of first or second importance at the urban sites was Lythrum salicaria. While this species was also found as a dominant in the seed bank at Patuxent Marsh, I feel this may have been due to cross contamination of pans, as this species was not found in the vegetation there. Three control pans used during the seed bank study were found to contain Lythrum salicaria, showing that it was able to cross contaminate pans, either through splashing or seeding in the greenhouse. Also, the number of seeds germinating from Patuxent Marsh samples was lower than from the other sites, so that dominant species in this marsh were found at relatively low abundance. Lastly, Patuxent Marsh's seed bank may contain Lythrum salicaria seeds, but not express them in the vegetation. Natural marshes may be more resistant to non-native species than restored or constructed marshes. Leck and Leck (2005) found Lythrum salicaria, Phalaris arundinacea, and Phragmites australis to be more prevalent in constructed wetland soils and vegetation than in reference marshes. Both restored wetlands were missing or had low abundance of the annuals *Pilea pumila*, Polygonum punctatum, and Polygonum sagittatum.

The vegetation study also highlighted the importance of *Lythrum salicaria* at the urban marshes, but it was not present in the vegetation at Patuxent Marsh. The older restored site, Kenilworth Marsh, was not significantly different from the reference sites with regards to percent cover, diversity, or evenness in the vegetation; with time, Kingman Marsh may also achieve this status.

Comparing the seed bank and vegetation species composition, it appears that the seed bank, with favorable greenhouse conditions, may have allowed more seedlings and species to germinate than was possible in the vegetation, given environmental conditions. In particular, *Cyperus* spp. and *Lindernia dubia* were more prolific in the seed bank than the vegetation, especially at Kingman Marsh. *Peltandra virginica* was the first or second most dominant species in the vegetation at all sites, but it has transient seeds, and so is not often seen in the seed bank, whereas *Lythrum salicaria*, *Typha* spp., and *Leersia oryzoides* are species often seen in both locations.

The seed bank tends to overestimate the percent of annuals present in the vegetation, likely because the seed bank in general is comprised of many annual species. It also does not take into account the open water or mudflat areas (potentially too low in elevation to support vegetation) that may not allow seeds to germinate; e.g., the seed bank supported approximately 70% natives at Kingman Marsh in 2003, but the vegetation only supported about 15%. The discrepancy may have been due to the large open water and mudflat areas present at Kingman Marsh (personal observation). The seed bank can also potentially underestimate the vegetation makeup of native species; e.g., Dueling Creek's seed bank contained about 50% natives versus the 75% found in the vegetation. Interestingly, Kenilworth Marsh's percentage of annuals and natives, and the

species composition, corresponded closely between the seed bank and vegetation, whereas other sites did not correspond as closely in these parameters.

The mudflat and open water areas at Kingman Marsh also may have contributed to the low evenness and taxa density in the vegetation. Kenilworth Marsh had the lowest evenness of the four sites in the seed bank, likely due to the abundance of *Typha* spp. and *Lythrum salicaria* in the seed bank. Patuxent Marsh and Dueling Creek, the two natural marshes, had similar evenness indices between the vegetation and seed bank, indicating that with time, the restored sites may possess higher evenness in both the vegetation and seed bank.

Sørensen's Quotient of Similarity in the seed bank study showed that Kingman Marsh and Kenilworth Marsh were becoming more similar to the reference sites with time (2001 to 2003), especially Dueling Creek, the urban natural marsh. In the vegetation study, Kingman and Kenilworth marshes were also more similar to Dueling Creek than they were to Patuxent Marsh, the natural non-urban site. The soils at the restored marshes were also found to be more similar to Dueling Creek than to Patuxent Marsh in terms of organic matter content and chemical composition (Neff 2002). It could be said that the three urban wetlands were more similar to each other because they all lie within the Anacostia watershed. I think that if we had looked at other urban sites outside of this watershed though, we would observe the same pattern; urban areas are more similar to urban areas likely because the urban environment alters wetlands lying within it, e.g., Dueling Creek. Similarly, the Patuxent watershed is more similar to other non-urban areas, such as the Nanticoke watershed in Eastern Maryland (personal observation). The

main characteristic differentiating these areas is the urban nature of the surrounding land use.

The findings from this project strongly point towards the greater suitability of Dueling Creek as a reference site than Patuxent Marsh. They also show that we should refer to older projects when restoring ecosystems, i.e., Kenilworth Marsh, so that we may learn what to expect as a restoration progresses. Of course, ecosystem development does not occur in a linear fashion; there is bound to be variability which is not often taken into account during restoration (Hilderbrand et al. 2005). Zedler and Callaway (2000) call for the evaluation of ecosystem responses to variability and unplanned disturbance events that occur in systems, e.g., herbivory and invasion of non-native species, so that we can learn how to make wetlands more resilient.

Kingman Marsh experienced intense goose herbivory shortly after planting (Hammerschlag et al. 2000; 2001; 2002; 2003; Neff 2002). In the face of this disturbance, stronger goose exclosures were installed. The exclosures may allow vegetation to develop to a point where it can withstand the herbivory pressure of non-migratory Canada geese. My project also explored disturbance, in this case brought about by a beaver dam. The dam created an impoundment that permanently flooded portions of Patuxent Marsh and altered the vegetation such that biomass (primary production) and taxa density were significantly higher in the non-impounded area. These findings allow us to study the effects of flooding in a field setting, as opposed to the seed bank greenhouse setting. Water depth and duration of inundation were significantly greater in the impounded area than the non-impounded area at Patuxent Marsh, increasing the likelihood that flooding was the major factor in the vegetation composition changes I observed over two years.

Kingman Marsh also experienced vegetation changes in response to flooding; many areas of marsh sediments subsided and/or eroded after dredge material placement and could not sustain seedlings or plant re-establishment due to the resulting higher water levels, especially when coupled with goose grazing pressure (Hammerschlag et al. 2002). Vegetation changes may also be the result of variation in stream flow occurring within and between years.

The disturbance induced by beavers at Patuxent Marsh was a natural process, as was the foraging and mound building by muskrats. These same processes, i.e., flooding due to low elevation and goose herbivory, are viewed as negative unnatural processes at Kingman Marsh. Just as non-native species occur in urban ecosystems due to surrounding land use changes and anthropogenic alteration of systems, natural processes are bound to cause different results in urban systems than they do in natural systems.

The lessons learned from this project are several: 1) the seed bank is a source of seeds for regeneration and also an indicator of the presence of non-native species; 2) seed bank studies can be used to compare natural and restored systems; 3) urban restoration sites should rely on urban reference sites in order to more realistically evaluate the potential of restored systems; and 4) impacts from animal disturbance and altered hydrology are important in both natural and restored systems, and should be considered, as well as incorporated into, tidal marsh restoration planning.

Appendix A. List of Plant Species

The comprehensive list of plant species (Latin names and life history characteristics) found during the 2000, 2001, and 2003 seed bank studies, 2002-2004 vegetation monitoring study, and 2003-2004 beaver dam study. A/P indicates annual or perennial species. If a species was indicated as biennial, it was included with the annuals. N/I indicates the species as native or introduced. Nomenclature, duration, origin, and wetland indicator status are based on the USDA PLANTS Database (2005).

Species	Family	A/P	N/I	Acronym
Acer saccharinum L.	Aceraceae	Р	Ν	ACESAC
Acer spp.	Aceraceae	-	_	ACESP
Acorus calamus L.	Acoraceae	Р	Ν	ACOCAL
Alisma subcordatum Raf.	Alismataceae	Р	Ν	ALISUB
Amaranthus blitum L.	Amaranthaceae	А	Ι	AMABLI
Amaranthus cannabinus (L.) Sauer	Amaranthaceae	Р	Ν	AMACAN
Ammannia coccinea Rottb.	Lythraceae	А	Ν	AMMCOC
Ampelopsis brevipedunculata (Maxim.) Trautv.	Vitaceae	Р	Ι	AMPBRE
Apios americana Medik.	Fabaceae	Р	Ν	APIAME
Arthraxon hispidus (Thunb.) Makino	Poaceae	А	Ι	ARTHIS
Artemisia vulgaris L.	Asteraceae	Р	Ι	ARTVUL
Asclepias incarnata L.	Asclepiadaceae	Р	Ν	ASCINC
Asteraceae	Asteraceae	_	_	—
Bidens cernua L.	Asteraceae	А	Ν	BIDCER
Bidens connata Muhl. Ex Willd.	Asteraceae	А	Ν	BIDCON
Bidens frondosa L.	Asteraceae	А	Ν	BIDFRO
Bidens laevis (L.) B.S.P.	Asteraceae	A/P	Ν	BIDLAE

Appendix A (continued)

Species	Family	A/P	N/I	Acronym
Bidens spp.	Asteraceae	_	_	BIDSP
Bidens tripartita L.	Asteraceae	А	Ν	BIDTRI
Boehmeria cylindrica (L.) Sw.	Urticaceae	Р	Ν	BOECYL
Brassica L. sp.	Brassicaceae	_	_	BRASSP
Calamagrostis coarctata (Torr.) Eat.	Poaceae	Р	Ν	CALCOA
Carex comosa Boott	Cyperaceae	Р	Ν	CARCOM
Carex lacustris Willd.	Cyperaceae	Р	Ν	CARLAC
Carex lurida Wahlenb.	Cyperaceae	Р	Ν	CARLUR
Cardamine pensylvanica Muhl. Ex Willd.	Brassicaceae	A/P	Ν	CARPEN
Carex L. spp.	Cyperaceae	_	_	CARSP
Carex stricta Lam.	Cyperaceae	Р	Ν	CARSTR
Carex tribuloides Wahlenb.	Cyperaceae	Р	Ν	CARTRI
Carex vulpinoidea Michx.	Cyperaceae	Р	Ν	CARVUL
Cephalanthus occidentalis L.	Rubiaceae	Р	Ν	CEPOCC
Chamaesyce maculata (L.) Small	Euphorbiaceae	А	Ν	CHAMAC
Chelone glabra L.	Scrophulariaceae	Р	Ν	CHEGLA
Cicuta maculata L.	Apiaceae	A/P	Ν	CICMAC
Conyza canadensis (L.) Cronq.	Asteraceae	А	Ν	CONCAN
Cornus amomum P. Mill.	Cornaceae	Р	Ν	CORAMO
Coronilla varia L.	Fabaceae	Р	Ι	CORVAR
Cuscuta gronovii Willd. ex J. A. Schultes	Cuscutaceae	Р	Ν	CUSGRO
Cynodon dactylon (L.) Pers.	Poaceae	Р	Ι	CYNDAC
Cyperaceae	Cyperaceae	_	_	_

Append	lix A	(continued)	1
rippene		(continued)	

Species	Family	A/P	N/I	Acronym
Cyperus erythrorhizos Muhl.	Cyperaceae	A/P	N	CYPERY
Cyperus flavescens L.	Cyperaceae	А	Ν	CYPFLA
Cyperus odoratus L.	Cyperaceae	A/P	Ν	CYPODO
Cyperus L. spp.	Cyperaceae	—	_	CYPSP
Cyperus strigosus L.	Cyperaceae	Р	Ν	CYPSTR
Dichanthelium clandestinum (L.) Gould	Poaceae	Р	Ν	DICCLA
Dichanthelium spp.	Poaceae	—	—	DICSP
Digitaria ischaemum (Schreb.) Schreb. ex Muhl.	Poaceae	А	Ι	DIGISC
Digitaria sanguinalis (L.) Scop.	Poaceae	А	Ν	DIGSAN
Draba L. spp.	Brassicaceae	_	_	DRABASP
Dulichium arundinaceum (L.) Britt.	Cyperaceae	Р	Ν	DULARU
Echinochloa crus-galli (L.) Beauv.	Poaceae	А	Ι	ECHCRU
Echinochloa muricata (Beauv.) Fern.	Poaceae	А	N/I	ECHMUR
Echinochloa Beauv. spp.	Poaceae	А	_	ECHSP
Echinochloa walteri (Pursh) Heller	Poaceae	А	Ν	ECHWAL
<i>Eclipta prostrata</i> (L.) L.	Asteraceae	A/P	Ν	ECLPRO
Eleocharis obtusa (Willd.) J.A. Schultes	Cyperaceae	A/P	Ν	ELEOBT
Eleocharis ovata (Roth) Roemer & J.A. Schultes	Cyperaceae	А	Ν	ELEOVA
Eleocharis R. Br. spp.	Cyperaceae	—	—	ELESP
Eleusine indica (L.) Gaertn.	Poaceae	А	Ι	ELEIND
<i>Epilobium ciliatum</i> Raf.	Onagraceae	Р	Ν	EPICIL
Epilobium coloratum Biehler	Onagraceae	Р	Ν	EPICOL
Appendix A (continued)

Species	Family	A/P	N/I	Acronym
Eragrostis pectinacea (Michx.) Nees ex Steud.	Poaceae	A/P	Ν	ERAPEC
Eragrostis pilosa (L.) Beauv.	Poaceae	А	Ν	ERAPIL
Eupatorium perfoliatum L.	Asteraceae	Р	Ν	EUPPER
Eupatorium serotinum Michx.	Asteraceae	Р	Ν	EUPSER
Eupatorium spp.	Asteraceae	_	_	EUPSP
Fraxinus pennsylvanica Marsh.	Oleaceae	Р	Ν	FRAPEN
Galium tinctorium L.	Rubiaceae	Р	Ν	GALTIN
Glyceria striata (Lam.) A.S. Hitchc.	Poaceae	Р	Ν	GLYSTR
Helenium autumnale L.	Asteraceae	Р	Ν	HELAUT
Heteranthera reniformis Ruiz & Pavón	Pontederiaceae	Р	Ν	HETREN
Hibiscus moscheutos L.	Malvaceae	A/P	Ν	HIBMOS
Hydrilla verticillata (L.f.) Royle	Hydrocharitaceae	Р	Ι	HYDVER
Hypericum boreale (Britt.) Bickn.	Clusiaceae	Р	Ν	HYPBOR
Hypericum mutilum L.	Clusiaceae	A/P	Ν	HYPMUT
Impatiens capensis Meerb.	Balsaminaceae	А	Ν	IMPCAP
Iris pseudacorus L.	Iridaceae	Р	Ι	IRIPSE
Juncaceae	Juncaceae	_	_	—
Juncus acuminatus Michx.	Juncaceae	Р	Ν	JUNACU
Juncus diffusissimus Buckl.	Juncaceae	Р	Ν	JUNDIF
Juncus effusus L.	Juncaceae	Р	Ν	JUNEFF
Juncus gerardii Loisel.	Juncaceae	Р	Ν	JUNGER
Juncus pelocarpus E. Mey.	Juncaceae	Р	Ν	JUNPEL
Juncus L. spp.	Juncaceae	_	_	JUNSP

(continued)

Appendix A (continued)

Appendix A (continued)				
Species	Family	A/P	N/I	Acronym
Juncus tenuis Willd.	Juncaceae	Р	Ν	JUNTEN
Lamiaceae	Lamiaceae	—	_	_
Leersia oryzoides (L.) Sw.	Poaceae	Р	Ν	LEEORY
Lemna minor L.	Lemnaceae	Р	Ν	LEMMIN
Lindernia dubia (L.) Pennell	Scrophulariaceae	А	Ν	LINDUB
Ludwigia alternifolia L.	Onagraceae	Р	Ν	LUDALT
Ludwigia leptocarpa (Nutt.) Hara	Onagraceae	A/P	Ν	LUDLEP
Ludwigia palustris (L.) Ell.	Onagraceae	Р	Ν	LUDPAL
Ludwigia peploides (Kunth) Raven	Onagraceae	Р	Ν	LUDPEP
Ludwigia L. spp.	Onagraceae	_	_	LUDSP
Lycopus americanus Muhl. ex W. Bart.	Lamiaceae	Р	Ν	LYCAME
Lycopus rubellus Moench	Lamiaceae	Р	Ν	LYCRUB
Lycopus L. spp.	Lamiaceae	_	_	LYCSP
Lycopus virginicus L.	Lamiaceae	Р	Ν	LYCVIR
<i>Lythrum salicaria</i> L.	Lythraceae	Р	Ι	LYTSAL
Mentha arvensis L.	Lamiaceae	Р	Ν	MENARV
Microstegium vimineum (Trin.) A. Camus	Poaceae	А	Ι	MICVIM
Mikania scandens (L.) Willd.	Asteraceae	Р	Ν	MIKSCA
Mimulus ringens L.	Scrophulariaceae	Р	Ν	MIMRIN
Mimulus L. spp.	Scrophulariaceae	_	_	MIMSP
Murdannia keisak (Hassk.) HandMaz.	Commelinaceae	Р	Ι	MURKEI
Myosotis laxa Lehm.	Boraginaceae	A/P	Ν	MYOLAX
Myriophyllum spicatum L.	Haloragaceae	Р	Ι	MYRSPI

(continued)

Appendix A (continued)

Appendix A (continued)				
Species	Family	A/P	N/I	Acronym
Nuphar lutea (L.) Sm.	Nymphaeaceae	Р	Ν	NUPLUT
Oenothera biennis L.	Onagraceae	А	Ν	OENBIE
Onoclea sensibilis L.	Dryopteridaceae	Р	Ν	ONOSEN
Orontium aquaticum L.	Araceae	Р	Ν	OROAQU
Panicum dichotomiflorum Michx.	Poaceae	А	Ν	PANDIC
Panicum rigidulum Bosc ex Nees	Poaceae	Р	Ν	PANRIG
Panicum miliaceum L.	Poaceae	А	Ι	PANMIL
Panicum spp.	Poaceae	_	_	PANSP
Peltandra virginica (L.) Schott	Araceae	Р	Ν	PELVIR
Penthorum sedoides L.	Crassulaceae	Р	Ν	PENSED
Phalaris arundinacea L.	Poaceae	Р	Ν	PHAARU
Phalaris L. spp.	Poaceae	_	_	PHASP
Phragmites australis (Cav.) Trin. ex Steud.	Poaceae	Р	N/I	PHRAUS
Pilea pumila (L.) Gray	Urticaceae	А	Ν	PILPUM
Pluchea odorata (L.) Cass.	Asteraceae	A/P	Ν	PLUODO
Poaceae	Poaceae	_	_	_
Poaceae sp. 1	Poaceae	_	_	_
Polygonum arifolium L.	Polygonaceae	А	Ν	POLARI
Polygonum caespitosum Blume	Polygonaceae	А	N/I	POLCAE
Polygonum hydropiper L.	Polygonaceae	А	Ι	POLHYD
Polygonum lapathifolium L.	Polygonaceae	А	Ν	POLLAP
Polygonum pensylvanicum L.	Polygonaceae	А	Ν	POLPEN
Polygonum persicaria L.	Polygonaceae	A/P	Ι	POLPER
(continued)				

Appendix A (continued)

Species	Family	A/P	N/I	Acronym
Polygonum punctatum Ell.	Polygonaceae	A/P	N	POLPUN
Polygonum sagittatum L.	Polygonaceae	A/P	Ν	POLSAG
Polygonum L. spp.	Polygonaceae	_	_	POLSP
Pontederia cordata L.	Pontederiaceae	Р	Ν	PONCOR
Populus deltoides Bartr. ex Marsh.	Salicaceae	Р	Ν	POPDEL
Ptilimnium capillaceum (Michx.) Raf.	Apiaceae	А	Ν	PTICAP
Ranunculus sceleratus L.	Ranunculaceae	A/P	Ν	RANSCE
Robinia pseudoacacia L.	Fabaceae	Р	Ν	ROBPSE
Rorippa palustris (L.) Bess	Brassicaceae	A/P	Ν	RORPAL
Rubus L. sp.	Rosaceae	_	_	RUBSP
<i>Rumex</i> L. sp.	Polygonaceae	_	_	RUMSP
Sagittaria latifolia Willd.	Alismataceae	Р	Ν	SAGLAT
Salix nigra Marsh.	Salicaceae	Р	Ν	SALNIG
Salix sp.	Salicaceae	_	_	SALSP
Saururus cernuus L.	Saururaceae	Р	Ν	SAUCER
Schoenoplectus fluviatilis (Torr.) M.T. Strong	Cyperaceae	Р	Ν	SCHFLU
Schoenoplectus pungens (Vahl) Palla	Cyperaceae	Р	Ν	SCHPUN
Schoenoplectus tabernaemontani (K.C. Gmel.) Palla	Cyperaceae	Р	Ν	SCHTAB
Scirpus cyperinus (L.) Kunth	Cyperaceae	Р	Ν	SCICYP
Scirpus polyphyllus Vahl	Cyperaceae	Р	Ν	SCIPOL
Scirpus L. spp.	Cyperaceae	_	_	SCISP
Scutellaria lateriflora L.	Lamiaceae	Р	Ν	SCULAT
Sium suave Walt.	Apiaceae	Р	Ν	SIUSUA
(continued)				

Appendix A (continued)

Species	Family	A/P	N/I	Acronym
Sparganium eurycarpum Engelm. ex Gray	Sparganiaceae	Р	Ν	SPAEUR
Sonchus asper (L.) Hill	Asteraceae	А	Ι	SONASP
Symphyotrichum dumosum (L.) Nesom	Asteraceae	Р	Ν	SYMDUM
Symphyotrichum lanceolatum (Willd.) Nesom	Asteraceae	Р	Ν	SYMLAN
Symphyotrichum puniceum (L.) A. & D. Löve	Asteraceae	Р	Ν	SYMPUN
Toxicodendron radicans (L.) Kuntze	Anacardiaceae	Р	Ν	TOXRAD
Trifolium L. spp.	Fabaceae	_	_	TRISP
<i>Typha latifolia</i> L.	Typhaceae	Р	Ν	TYPLAT
<i>Typha</i> L. spp.	Typhaceae	_	_	TYPSP
Unknown dicot	_	_	_	UDI
Unknown monocot	_	_	_	UMO
Vernonia noveboracensis (L.) Michx.	Asteraceae	Р	Ν	VERNOV
Zizania aquatica L.	Poaceae	А	Ν	ZIZAQU

Appendix B. Control Pans

Control pan data collected during the seed bank study. Control pans were placed in the greenhouse during the 2001 and 2003 seed bank studies to determine if cross-pan seedling establishment was occurring or if environmental factors in the greenhouse were affecting the study.

Year	Pan	Species	No.
2001	C1	Conyza canadensis	11
	C2	Lythrum salicaria	1
		Sonchus asper	1
2003	C2 non-flooded	Lythrum salicaria	3
		Panicum dichotomiflorum	1
		Unidentified dicot	5
	C3 non-flooded	Lythrum salicaria	5
		Rorippa palustris	1
		<i>Typha</i> sp.	1
		Unidentified dicot	1
	C4 flooded	Ludwigia palustris	1
	C4 non-flooded	Ludwigia palustris	1
		Rorippa palustris	4
	C5 non-flooded	Unidentified dicot	1

Appendix C. Examples of code used for statistical analysis

C.1 Example of SAS code to estimate lsmeans for vegetation species.

*vegetation transect analyses with SW diversity and pielou's j; **data** COVER;

input Year\$ Month\$ Site\$ Area\$ Planted\$ Trans\$ Sector\$ NOCOV TOTVEG ACESAC ACESP ACOCAL ALISUB AMACAN AMPBRE APIAME ARTHIS ARTVUL ASCINC ASTSP BIDCER BIDCON BIDFRO BIDLAE BIDTRI BIDSP BOECYLBRASSSP CALCOA CARCOM CARLAC CARLUR CARPEN CARSTR CARTRI CARVUL CARSP CEPOCC CHEGLA CICMAC CORAMO CUSGRO CYPERY CYPFLA CYPODO CYPSTR CYPSP CYPERSP DICCLA DICSP DULARU ECHCRU ECHMUR ECHWAL ECHSP ECLPRO ELEOBT ELESP EPICIL EUPPER EUPSER EUPSP FRAPEN GALTIN GLYSTR HELAUT HETREN HIBMOS HYDVER HYPMUT IMPCAP IRIPSE JUNACU JUNEFF JUNSP LAMIASP LEEORY LEMMIN LINDUB LUDPAL LUDPEP LUDSP LYCAME LYCRUB LYCVIR LYCSP LYTSAL MENARV MIKSCA MICVIM MIMRIN MIMSP MURKEI MYOLAX MYRSPI NUPLUT ONOSEN OROAQU PANDIC PELVIR PENSED PHAARU PHRAUS PILPUM PLUODO POASP POACESP POLARI POLCAE POLHYD1 POLHYD2 POLLAP POLPER POLPUN POLSAG POLSP PONCOR POPDEL PTICAP RANSCE ROBPSE RORPAL RUBSP RUMSP SAGLAT SALNIG SALSP SAUCER SCHFLU SCHPUN SCHTAB SCICYP SCIPOL SCISP SCULAT SIUSUA SONASP SPAEUR SYMDUM SYMPUN TOXRAD TRISP TYPSPP UNK UNKDIC UNKMON VERNOV ZIZAQU Swdive RICHNESS pielou

· ;

datalines;

<data>;

proc transpose data=cover out=transposed name=species; by year month site area trans sector notsorted; run;

*proc print data=transposed; *run;

proc sort data=transposed; by Site Area Trans year month species; quit;

proc means mean n stderr data=transposed noprint; by Site Area Trans year month species;

var col1; output out=meanstra mean=mmean stderr=error; quit;

proc sort data=meanstra; by species site; quit; proc mixed data=meanstra; by species; class Site Trans Year Month species; model mmean = site|month(year) / ddfm=satterth outp=resids; repeated month(year); lsmeans site|month(year)/diff=all adjust=tukey; ods output lsmeans=lsmean1; ods listing exclude diffs; ods output diffs=diff1; ods output tests3=stat1; run; %include 'f:\pdmix800.sas'; %*pdmix800*(diff1,lsmean1,alpha=.05,sort=yes); quit: **proc sort** data=resids; by species; quit; proc plot data=resids vpercent=50; plot resid*pred/vref=0; by species; quit; Proc univariate data=resids plot normal; var resid; by species; quit; proc print data=lsmean1; quit; /*proc print data=diff1; format estimate stderr 6.2; var site year month site year month Estimate StdErr Adjp; quit;*/ proc print data=stat1; quit; title ""; quit;

C.2 Example of SAS code to estimate the Chi-square test of independence for annual

versus perennial species or native versus non-native species.

```
data ann2003;
input year$ site$ durat$ count;
lines;
<data>;
data ann2003;
*if year='2000' then delete;
*if year='2001' then delete;
*if year='2003' then delete;
if durat='nat' then delete;
if durat='nonnat' then delete;
*if durat='per' then delete;
run;
quit;
```

```
proc sort data=ann2003;
by site;
quit;
```

```
proc freq data=ann2003;
weight count;
tables durat*year / nopercent norow nocol expected chisq;
by site;
quit;
```

C.3 Example of SAS code to compare means between two samples using a t-test.

Title 1 t-test for percent flooded at Patuxent; data flood; input trt\$ rep percent; datalines; <data>; run; PROC TTEST; CLASS trt; VAR percent; quit; C.4 Example of SAS code to compute a correlation matrix using Pearson's r correlation

coefficient.

data soil; input PCTMOI BULKD OM eH CONDuS TEMP SAL watd; datalines; <data>; proc corr data =soil; var PCTMOI BULKD OM eH CONDuS TEMP SAL watd; run;

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