

# Macroinvertebrate diversity and abundance in two created wetlands in Ohio

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## Abstract

Benthic macroinvertebrates were sampled in October 2005 in two experimental created wetlands as part of an on-going attempt to monitor ecosystem development over the long term. These wetlands are eleven years old, and during creation one wetland was planted while the other was allowed to colonize naturally. The wetlands were sampled using the Hester-Dendy plate method and D-frame dipnets. The two methods collected 4391 organisms from 22 taxa over a two-week period. The results were analyzed using Shannon-Wiener diversity index and non-parametric Mann-Whitney test, but statistical analysis did not support any significant differences in abundance and diversity between sites. The wetlands have shown an increase in diversity over the past ten years, but variability in sampling methods and rigor make it difficult to confirm a trend. The study of this whole-ecosystem experiment investigates how multiple factors may structure communities.

## Introduction

Wetlands are being created and restored around the world, often in an effort to replace the habitat values lost through destruction of natural wetlands (Gilbert and Anderson, 1998; Mitsch, 1998b; NRC, 2001). In replacing wetlands, it is important to be able to assess whether the created habitat provides the same functions as natural wetlands (Wissinger et al., 2001). It can be difficult to assess the functionality of a wetland, but some studies have evaluated the use of indicators such as plants, animals or water quality (Simenstad and Thom, 1996; Blackwell and Maltby, 1998; Rader et al., 2001). Most created wetlands are built for both habitat and water quality value, but it takes time for habitats to mature and communities to establish themselves (Galatowitsch and van der Valk, 1996; Mitsch and Wilson, 1996).

In many aquatic habitats, macroinvertebrate communities are useful indicators of biotic integrity (Broderson et al., 1998; Weatherhead and James, 2001; Stanczak and Keiper, 2004). Invertebrates are crucially important to the overall functioning of wetland ecosystems due to their central position in food webs and their role in processing detritus (Batzer et al., 1999; Euliss et al., 1999; Merritt et al., 1999). Benthic organisms live in or near the substrate. Macroinvertebrates are organisms large enough to be seen with the naked eye and include worms, crustaceans, mollusks, mites and insects. Most of the aquatic vegetation,

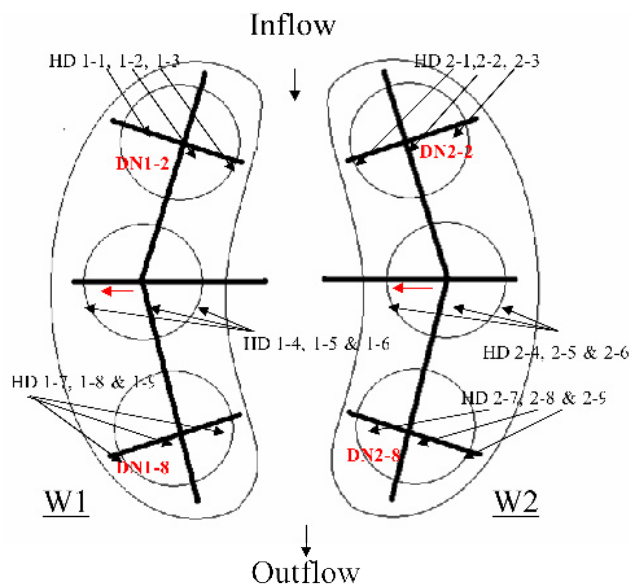


Figure 1. Location of sampling stations in the experimental wetlands with Hester Dendy traps (HD) and Dipnets (DN)

or primary production, in wetlands enters the detritus pathway rather than being grazed by consumers. Bacteria and fungi break down (and enrich) plant litter and these organisms are consumed by macroinvertebrates, which are eaten in turn by fish, birds, amphibians and mammals. Many species of birds (both wetland and non-wetland species) consume vast numbers of the emerging masses of insects that develop in wetland ecosystems (Batt, 2000). The structure of aquatic macroinvertebrate communities is complex and depends on factors such as vegetation structure, hydroperiod, food resources, temperature, or oxygen. This leads to high temporal and spatial variety of insects inhabiting various types of wetlands.

Macroinvertebrates can provide good indicators of community response to environmental change, as some taxa are quite sensitive to pollution or other abiotic factors (Dvorak and Imhof, 1998; Martin and Neely, 2001; Brown and Batzer, 2001). Comparisons between studies can be difficult to perform as the method used in sampling the macroinvertebrate community can affect the results. Invertebrate samples collected using different sampling methods or at another time of year might yield different organisms or numbers collected. An earlier study at the

Olentangy River Wetland Research Park (ORWRP) found that bottle traps were most effective for total number of organisms collected, followed by dip nets and Hester-Dendy (HD) plates (Custer and Johnson, 1998). The objective of this study is to determine if there is a difference in the abundance or diversity of aquatic macroinvertebrates between the experimental wetlands. We will also examine whether the sampling methods used can offer a valid comparison of the macroinvertebrate community from year to year.

## Methods

### Study area

The study was carried out in two created wetlands at the ORWRP, on the Ohio State University campus in Columbus, Ohio. Wetland 1 (W1) was planted with thirteen common wetland plant species, while Wetland 2 (W2) was allowed to colonize naturally when the wetlands were created in 1994 (Mitsch, 1998a). Each adjacent wetland covers 1-ha, receives

Table 1. Summary of benthic macroinvertebrate studies at the ORWRP

Year of study	Author(s)	Dominant taxa	Number of Organisms	Number of Taxa	Methods*	When sampled
1994	Nairn et al. (1995)	Gastropods	772	10	Colonization plates plus surber	Oct 7–28
1994	Dabrowska and Lentz (1995)	Mollusks	NA	9	Ceramic tile colonization plates	Dec
1994	Martin and Armitage (1995)	NA	NA	16	Sieve and forceps	Sep–Nov
1994	Minamyer (1995)	Odonates only	NA	18	Seine (part of fish survey) and aerial nets	Jun
1995	Dabrowska (1996)	Chironomidae	NA	18	Colonization plates	Oct–Nov
1995	Metzker (1996)	Chironomidae	1883	25	Clay colonization plates	Oct–Nov
1996	Hart et al. (1996)	Gastropods	3225	38 in W1 32 in W2	DN, HD and clay colonization	Oct 10–24
1996	Dabrowska (1997)	Gastropods	NA	20	Substrate colonization plates	Oct–Nov
1997	Spieles (1998)	Chironomidae	1557	41	HD, ET	Apr–Oct
1997	Cochran (1998)	Gastropods	1907	9	HD, minnow traps	Oct 6–Nov 3
1998	Lowry (1999)	Gastropods	1355	10	HD, BT, DN	Oct
1998	Custer and Johnson (1999)	Gastropods	1355	19	BT, HD, DN	Oct
1999	Frazier and Mitsch (2000)	Gastropods	NA	8	HD, ET, FT	Oct
1999	Custer et al. (2000)	Chironomidae	264	36	HD, ET	Apr 2–May 6
2000	Acharyya and Mitsch (2001)	Gastropods	572	15	HD, DN, BT	Oct
2001	Webb and Mitsch (2002)	Gastropods	510	12	HD, DN	Oct
2002	Holland and Mitsch (2003)	Collectors in outflow	894	19	HD, BT	Oct
2003	Grubh and Mitsch (2004)	Oligochaetes in HD; Gastropods in DN	22880	26	HD, DN	Oct 9–Nov 6
2004a	Gamble et al. (2005)	Gastropods overall	4895	35	HD, BT, DN	Sep
2004b		Amphipods	1112	19	HD	Oct
2005	Current study	Chironomidae	4391	22	HD, DN	Oct

\* HD = Hester-Dendy colonization plate; BT = Bottle trap, DN = Dipnet, FT = Funnel trap, ET = Emergent traps

the same amount of water, and experiences the same biotic and abiotic conditions. The macroinvertebrate communities have been surveyed each year since the wetlands were created, under a variety of sampling regimes (Table 1). For the past twelve years, HD samplers have been used (along with other sampling methods) as a standard method to facilitate comparisons between years.

### Sampling design

Macroinvertebrates were sampled using HD plates and dip nets. HD samplers were placed at the inflow, middle and outflow area of the wetlands, using established sampling locations (Gamble et al., 2005). Nine sets of HD plates were set in each wetland, three in each of the inflow, middle and outflow areas (Figure 1). Each set of plates was suspended above the sediments and attached to the boardwalk with fishing line. The HD plates were left in place for two weeks, from October 7–21, 2005. The plates were carefully removed at the end of the sampling period, by gently maneuvering each unit into a net before lifting it out of the water and transferring it into a labeled plastic bag. Each sample was rinsed and scraped with a spatula into a bucket, the contents were passed through a #40 sieve (mesh size of 420 microns) and backwashed with 95% ethanol into a labeled collection jar for later identification.

The dipnet samples were taken only at the inflow (W1-2 and W2-2) and outflow (W1-8 and W2-8) areas (Figure 1). For the dipnet sampling, performed on October 25, 2005,

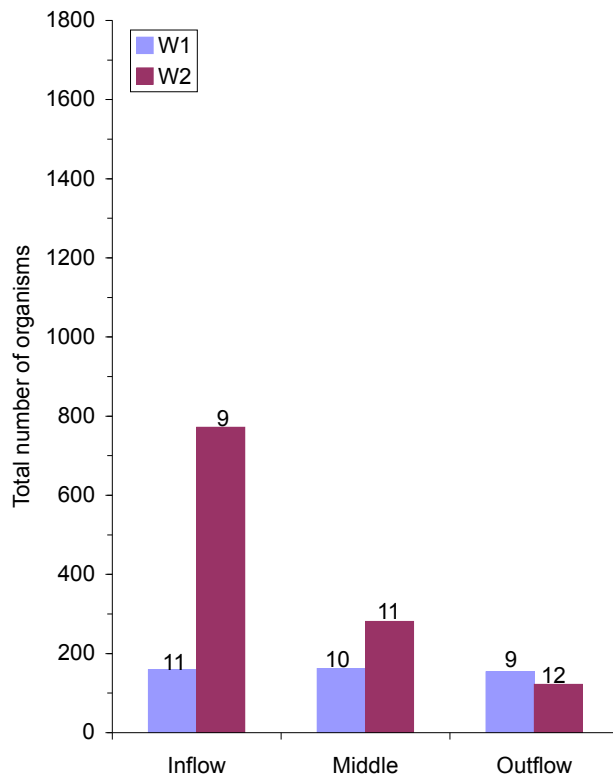


Figure 2. Distribution of organisms collected by HD traps in the inflow, middle and outflow regions of W1 and W2. The numbers indicate number of different taxa collected at that site.

the net was placed on the sediment and pulled through one meter, and then passed back and forth along the same path just above the sediments three more times. The contents of the net were rinsed into a bucket, and sieved and collected as above.

Each sample was sorted under a dissecting microscope and the invertebrates identified to family level if possible, using keys in Merritt and Cummins (1996), Arnett (2000) and Voshell (2002). The Shannon-Wiener index for richness and diversity was performed for comparison to previous years' studies. The two indices that were calculated for W1 and W2 are for species diversity and species evenness. Species diversity,  $H'$ , is calculated by taking the negative mathematical sum of the relative proportion of species to the total number of species,  $p_i$ , multiplied by the natural logarithm of itself.

$$H' = -\sum p_i * \ln p_i$$

Species evenness,  $J$ , is calculated by taking the species diversity and dividing it by the natural logarithm of the species richness. Species richness,  $S$ , is defined as the total number of species.

$$J = H' / \ln S$$

$H'$  values normally range from 0–5 to indicate the diversity of the population.  $J$  values range from 0–1 to one to indicate how evenly the species are distributed. Graphical analysis of the data indicated that the distribution was heavily skewed (Figures 2 and 3), and this was confirmed by performing an Anderson-Darling test for normality

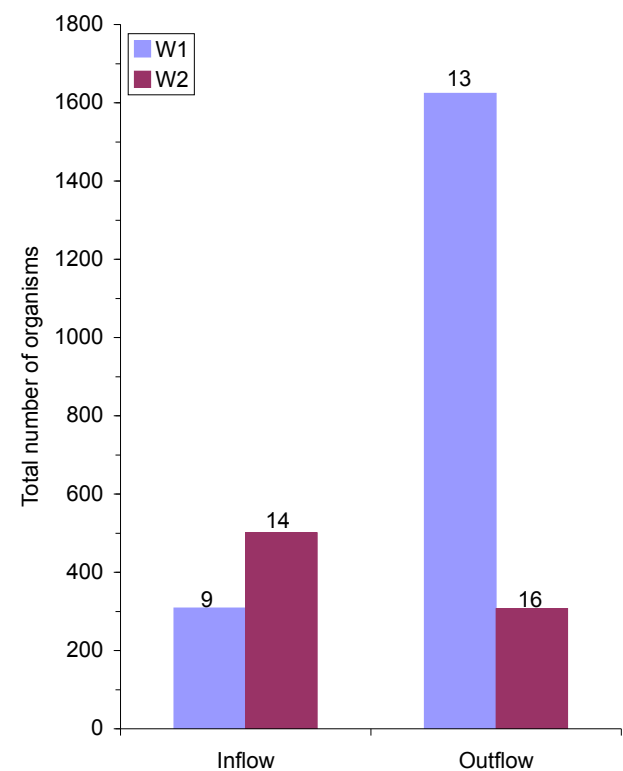


Figure 3. Distribution of organisms collected by dip net in the inflow and outflow regions of W1 and W2. The numbers indicate total number of taxa collected at that site.

Table 2. Macroinvertebrates sampled on Hester-Dendy (HD) plates

Order: Family	W1 inlet	W1 middle	W1 outlet	W2 inlet	W2 middle	W2 outlet	Total
Acariformes: Hydrocaridae	0	0	0	0	0	1	1
Amphipoda: Gammaridae	11	0	1	70	34	3	119
aquatic worm	84	6	8	334	31	27	490
Coleoptera: Haliplidae	0	0	0	1	0	0	1
Corbiculoidea: Sphaeriidae	7	8	1	4	33	3	56
Diptera: Chironomidae	12	21	95	8	6	10	152
Ephemeroptera: Caenidae	0	28	0	0	12	4	44
Hemiptera: Corixidae	3	2	0	0	0	0	5
leech	19	18	5	41	75	37	195
Odonata: Coenagrionidae	4	36	1	3	21	11	76
Odonata: Libellulidae	2	16	5	0	43	3	69
Pulmonata: Lymnaeidae	1	4	0	0	0	0	5
Pulmonata: Physidae	15	23	36	29	8	7	118
Pulmonata: Planorbidae	0	0	2	0	3	1	6
Trichoptera: Hydroptilidae	1	0	0	0	0	0	1
Tricladidia: Planariidae	0	0	0	282	15	15	312
Total	159	162	154	772	281	122	1650

Table 3. Macroinvertebrates sampled with dipnets

Order: Family	DN1 inlet	DN1 outlet	DN2 inlet	DN2 outlet	Total
Acariformes: Hydrocaridae	0	2	1	8	11
Amphipoda: Copepoda	0	0	0	36	36
Amphipoda: Gammaridae	7	3	20	5	35
aquatic worm	134	120	100	95	449
Coleoptera: Haliplidae	0	0	4	0	4
Coleoptera: Hydrophilidae	0	0	1	0	1
Corbiculoidea: Sphaeriidae	18	9	92	26	145
Diptera: Ceratopogonidae	1	21	7	2	31
Diptera: Chironomidae	50	1375	164	89	1678
Ephemeroptera: Baetidae	15	6	0	0	21
Ephemeroptera: Caenidae	0	6	0	10	16
Hemiptera: Corixidae	40	36	10	18	104
leech	7	1	14	5	27
Odonata: Coenagrionidae	0	1	0	2	3
Prosobranchia: Hydrobiidae	0	0	1	0	1
Pulmonata: Lymnaeidae	0	0	6	1	7
Pulmonata: Physidae	37	29	69	5	140
Pulmonata: Planorbidae	0	15	0	3	18
Trichoptera: Polycentropodidae	0	0	0	1	1
Tricladidia: Planariidae	0	0	12	1	13
total	309	1624	501	307	2741

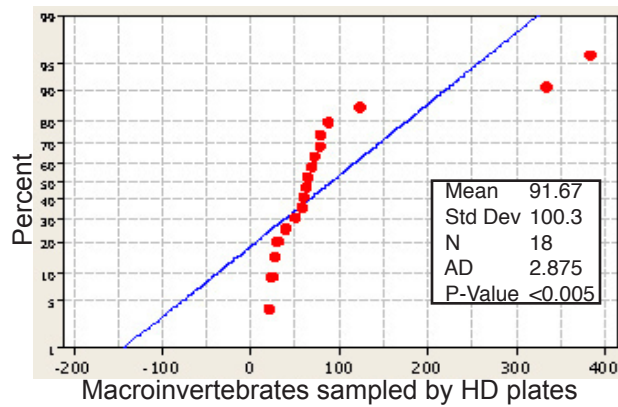


Figure 4. Anderson-Darling test for normality (Normal) in population sampled by HD traps

(Figure 4). Because the standard t-tests are not accurate for non-normally distributed populations, a non-parametric Mann-Whitney test was used to compare the abundance of organisms between W1 and W2.

## Results

The HD plates collected a total of 1650 organisms, with 14 families representing 11 orders plus two classes, Hirudinea and Oligochaeta, that were not classified to family (Table 2). The dipnet sampling collected 2741 organisms from 18 families representing 12 orders, plus the two classes (Table 3). The Shannon-Wiener indices for diversity and evenness were 2.02 and 0.77 in W1 and 1.84 and 0.74 in W2, respectively. The diversity index was compared to previous studies (Figure 5). These results indicate an increase in diversity over time, but due to the fact that sampling and identification was not uniform across studies, a certain amount of variability is apparent. The current indices are comparable to the past three years. The Mann-Whitney test did not support any significant differences between wetlands ( $p = 0.22$ ,  $Z = -24.0$ , with 95% confidence interval of  $-255.0, 8.9$ ). The five most abundant taxa sampled by HD in each wetland are shown according to their distribution within each region of the wetlands (W1 in Figure 6a, W2 in Figure 6b).

## Discussion

The results of this study suggest that macroinvertebrate diversity and abundance continue to converge between the wetlands. The two ORWRP wetlands receive identical amounts of water and solar radiation, and the surrounding upland communities are similar, so any differences should be attributable mainly to the different macrophyte communities. Comparison of the two wetlands have shown that the communities of plants and animals have experienced cycles of divergence and convergence, but after ten growing seasons some differences are diminishing (Mitsch et al., 2005a,b). While overall productivity and percent vegetation cover in the wetlands is approximately

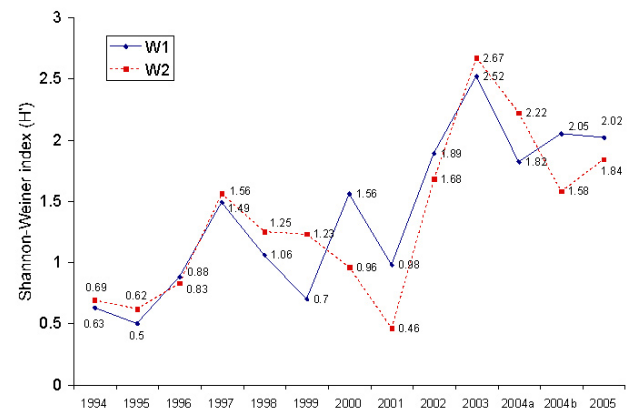


Figure 5. Shannon-Weiner index for diversity in the experimental wetlands

equal, the macrophyte community still shows differences between sites. The planted wetland is more diverse, with four plant species dominant. W2, the naturally colonized site, is dominated by *Schoenoplectus tabernaemontani* and *Typha* spp. (Mitsch et al., 2005b). Apparently the differences in plant community do not structure the macroinvertebrate community in a statistically significant fashion. Our results are in accordance with the hypothesis that over time, the experimental wetlands will converge in structure and function (Mitsch and Wilson, 1996). It is possible that overall productivity is higher in less diverse communities, and that macrophyte community diversity may not necessarily lead to higher macroinvertebrate diversity (Mitsch and Day, 2004). Another possibility is that a combination of factors is at work. A survey of the fish and amphibian communities showed differences between the experimental wetlands (Fink and Mitsch, 2005). Differential predation may have interacted with the effect of the plant community to create an overall similarity in macroinvertebrates, as was observed in prairie wetlands in Minnesota (Zimmer et al., 2001). In this study, one wetland contained fish while the other did not. Production biomass of the invertebrate communities was similar between the wetlands, despite the lack of biomass in larger size classes in the wetland containing fish. The authors hypothesized that a combination of the presence/absence of fish, and differences in the macrophyte communities of the two wetlands played important roles in their results.

Our sampling design was motivated largely by the need to generate data that could be compared to previous years, but the HD sampling method is known to under-represent some macroinvertebrate species. The method does allow for quantitative data analysis, as the area sampled is consistent between sampling stations. A comparison of the different methods for sampling macroinvertebrates in wetlands found that the type of device found to be most effective varied from wetland to wetland (Batzer et al. 2001). Larger corers and sweep nets are the only methods that collect whole communities, and both have several disadvantages (time-consuming to sort through plant matter, dense vegetation inhibits operation, etc.). Although the sweep



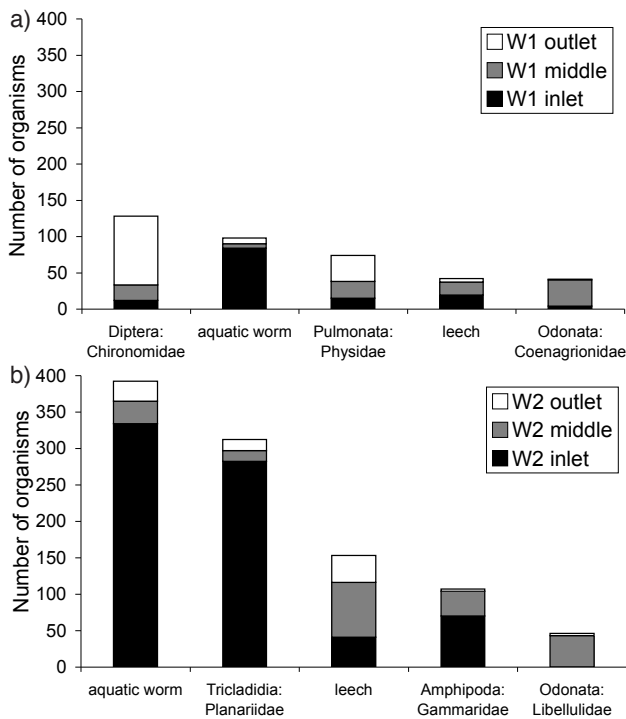


Figure 6. Distribution of the five most abundant taxa in (a) W1 and (b) W2 collected using HD traps

net method is semiquantitative, it tends to be the preferred method for bioassessment of wetlands. Rapid deployment and ease of use are the primary factors influencing this choice. Muzaffar and Colbo (2002) evaluated two sampling techniques for estimating diversity and density of benthic macroinvertebrates. Neither sampling technique captured representatives of all the benthic macroinvertebrate taxa found on coarse rocky substrates within the littoral zone of two ponds in Newfoundland, Canada. Furthermore, density estimates for the abundant taxa collected by the two sampling techniques are not equal, illustrating the different selectivity of each technique even for the taxa collected by both techniques.

The Shannon-Weiner diversity index is designed to be used when species are identified to the species level, but most of the ORWRP studies have only identified organisms to the family or genus. The indices are not as reliable when the organisms are not identified to species, but the degree to which the results may be skewed cannot be determined. Another confounding factor is related to sampling procedures. Aquatic worms tend to fracture easily under normal collecting and sorting procedures. The tendency to count broken sections as individual worms can lead to over-estimation of the number of this specific taxon.

A certain degree of error in collection and sampling of aquatic macroinvertebrates can be seen in other studies. A study in the Lake Ontario and St. Lawrence River plains of New York looked at macroinvertebrates, plants and birds (Brown and Batzer, 2001). They found significant

differences in the abundances of specific macroinvertebrate taxa between restored and reference wetlands. In their study, the authors attempted to minimize the impact of sampling on subsequent community development by processing the samples on site and returning live invertebrates to the habitat. Other studies at the ORWRP have occasionally used less effective sampling and sorting techniques, such as sorting samples on the boardwalk or using bottle traps that allowed predators to consume part of the sample (Custer and Johnson, 1998; Holland and Mitsch, 2002). The processing of organisms on site, instead of in the lab, is very likely to lead to errors in identification or to the exclusion of small or camouflaged species. The macroinvertebrate sampling for each year has been performed by different personnel, with varying levels of expertise in the identification of aquatic insects, and to different levels of intensity. Some studies have collected organisms over a short period, others for longer. The macroinvertebrate study conducted by Gamble (2004) collected 4895 macroinvertebrates with 35 taxa from seven classes and 17 orders, using three methods (H-D plates, bottle traps and dipnets). An intensive study performed during the pulsing experiment collected 22,880 macroinvertebrates with a total of 26 taxa from 13 orders and 26 families, using HD plates and dipnets. The HD plates were left in place for one month, and dipnet sampling was carried out on alternate days for three weeks (Grubh and Mitsch, 2003). Overall the differences in technique, rigor of identification, biases inherent in sampling method and other variables makes drawing comparisons between studies somewhat problematic (Gamble et al., 2004). What these studies at ORWRP over the years do reveal are the general trends that indicate how multiple factors can interact with each other in ways difficult to predict from studies with controlled systems.

The importance of whole-ecosystem studies is more than just large size. For example, a mesocosm experiment may investigate the effects of nutrients on plants but a whole-ecosystem study is needed to investigate the role of nutrients on ecosystem functions, with plants being affected by the nutrients but also at the same time by herbivory, decomposition, sedimentation, and a host of other factors that are not independent of the nutrient flow (Mitsch and Day, 2004). The complexity of interactions between many factors can lead to difficulties in estimating ecosystem functioning by use of one or a few parameters. Despite the limitations for comparisons between studies due to variability in sampling method and rigor, the value of examining whole-ecosystem experiments over time lies in the ability to discover how multiple factors can drive productivity and community development. Subtle interactions may have an incremental or synergistic effect that can only be observed by long-term whole-ecosystem studies. These on-going investigations into the many factors influencing the experimental wetlands at the ORWRP can lead to many surprising and valuable insights into ecosystem ecology.

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