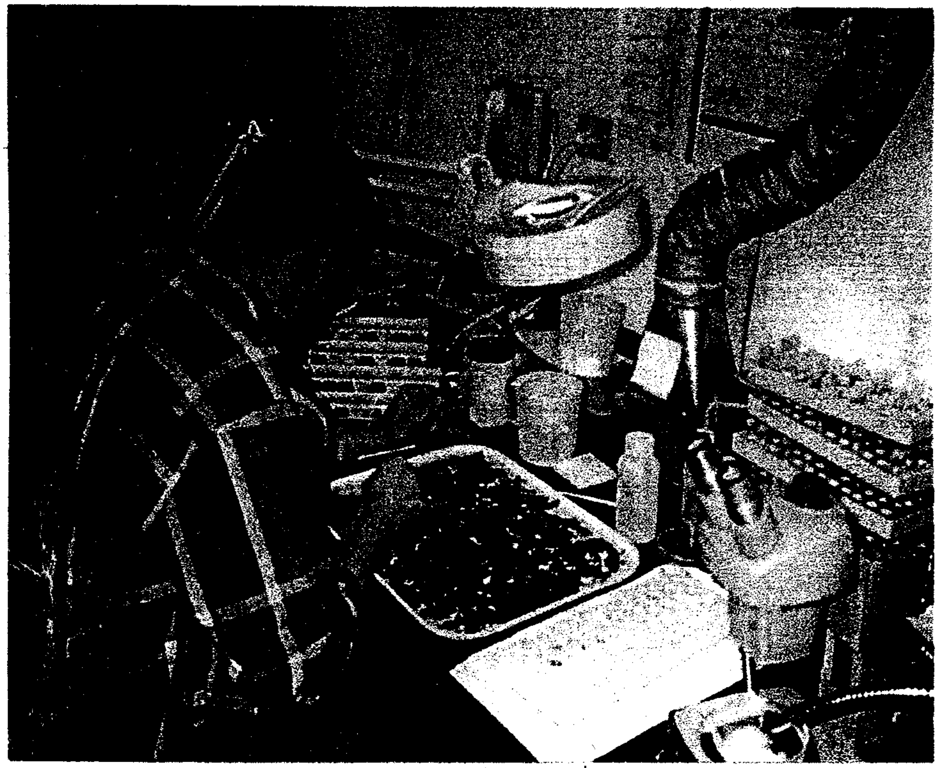
# DEVELOPMENT OF A BIOLOGICAL INDEX AND CLASSIFICATION SYSTEM FOR WISCONSIN WETLANDS USING MACROINVERTEBRATES AND PLANTS

Final Report to USEPA Region V, Wetland Grant #CD985491-01-0

**January 2000**

Prepared by:
Richard A. Lillie, Wisconsin Department of Natural Resources
Bureau of Integrated Science Services, Fish & Habitat Section
1350 Femrite Drive, Monona, Wisconsin 53716



**Funded by a grant from the U.S. Environmental Protection Agency - Region 5**
**Conducted in cooperation with the Bureau of Fisheries Management & Habitat Protection, Lakes & Wetlands Section**

WISCONSIN
DEPT. OF NATURAL RESOURCES

This report was prepared by the Wisconsin Department of Natural Resources under Grant No. CD985491-01-0 from the U.S. Environmental Protection Agency, Region 5. Points of view expressed in this report do not necessarily reflect the views or policies of the U.S. Environmental Protection Agency. Mention of trade names and commercial products does not constitute endorsement of their use.

# TABLE OF CONTENTS

## APPENDICES:
**(Note: printed under separate cover)***



* Copies available upon request from author at Wisconsin Department of Natural Resources, 1350 Femrite Drive, Monona, WI 53716 (phone 608-221-6338; e-mail at lillir@dnr.mail.state.wi.us).

## EXECUTIVE SUMMARY


*We developed two macroinvertebrate-based indices for measuring the biological integrity of Wisconsin palustrine depression wetlands by collecting and processing nearly 100,000 macroinvertebrates from a total of 104 wetland basins distributed across the major ecoregions of Wisconsin. Additionally, we developed a plant-based index of biological integrity from the same set of wetlands, and proposed a subjective macroinvertebrate habitat index. The two macroinvertebrate indices are derived from composited kick-net sweep samples collected from the wetland perimeter. The first index, termed the Wisconsin Wetland Macroinvertebrate Biotic Index (WWMBI), is a multimetric index based on the individual abundance of 15 selected macroinvertebrate fauna. The second macroinvertebrate index, the 100-Count Macroinvertebrate Biotic Index (100-Count MBI), is a multimetric index based on relative percentages of nine selected macroinvertebrate fauna in a random sub-sample of 100 organisms. The later index is intended to serve as a practical, rapid, field, bioassessment tool. The plant index is a multimetric index based on relative importance values (percent cover and frequency of occurrence data) of nine selected plant taxonomic groups (mixed taxonomic resolution) found to occur in 18 quadrats distributed along three transects in each wetland. The subjective macroinvertebrate habitat index is derived from six environmental attributes and was intended to serve as a predictor of biotic index scores as influenced by the combination of naturally occurring factors that are known or suspected to influence macroinvertebrate populations and human disturbance factors (which are difficult to predict).*

*The three multimetric biotic indices exhibited considerable promise in differentiating among wetland types, wetland history, disturbance condition, and water duration. Basic differences in macroinvertebrate and plant community structure that exist among wetland types (e.g., wooded kettles versus open prairie type wetlands) confounded our ability to measure the performance of the indices. However, the response of index scores and biological integrity ratings based on these scores tend to correspond to wetland history and disturbance classifications and to our preconceived hypotheses as to how human disturbance should affect macroinvertebrate and plant communities. The subjective macroinvertebrate habitat index did not perform as well as expected, suggesting that further modifications to the index are necessary.*

*The three indices demonstrated excellent potential for use in evaluating wetland restoration efforts in Wisconsin depression type wetlands. Biological purpose of wetland may be defined in terms of the macroinvertebrate and plant communities that they support, both in terms of dominant forms and rare taxa. Data collected in this study support that further hierarchical classification of depression wetlands is required to reduce inter-wetland variability and maximize understanding of wetland biological function and response to human disturbance.*

# INTRODUCTION

In the past decade, a considerable effort has been made at local, state, and federal levels to protect, restore, and create wetland habitat. This effort is based on the knowledge that wetlands perform a myriad of functions to mankind; functions which include physical, chemical, and biological components. The relative significance of these functions differs according to the spatial position of individual wetlands in the landscape or the temporal stage of the wetland with respect to dynamic climatic cycles. The biological function of wetlands, while obvious in general context to most biologists or ecologists, is often questioned by developers, engineers, and the public. This is especially true when those wetlands are small, transient or temporary, or hinder man's attempts to farm, build roads or other structures, or where the wetlands simply appear to serve as breeding areas for hoards of nuisance mosquitoes. The biological function of wetlands is addressed in Wisconsin's Wetland Water Quality Standards (Wisconsin Administrative Code NR 103.03(1)(e-f)), which provides for the protection of habitat for aquatic organisms and resident and transient wildlife, and in NR 103.03(2)(e), which protects the hydrological conditions necessary to support these biota. Due to the complexity of hydrological and ecological conditions associated with the many different types of wetlands found in Wisconsin, the code was established with simple narrative water quality criteria or conditions rather than specific numerical criteria. The intent of this study is to provide a tool, in this case a new biological index and classification system, that may be used to quantify, characterize, rank, and define biological function and ecosystem integrity of wetlands.

*NOTE: This study was directed at small, palustrine or depression basin wetlands only!*

The objective of this study is to develop a hierarchical approach to classifying and ranking wetlands as to biological purpose, condition, and relative rarity using an index based on a combination of macroinvertebrate (primarily aquatic insects) and plant communities. Biological purpose is defined in terms of function. What biota does the wetland support, and what purpose does that biota serve in the food-web (e.g., aquatic insects serve as food resources for waterfowl production) or in modifying environmental impacts or conditions (e.g., nutrient uptake and water quality preservation by aquatic macrophytes). Emphasis in the current investigation was given to depression wetlands that hold standing water for periods of less than one year and that are isolated from other waterbodies (i.e., palustrine). Temporary basins, by virtue of the lack of resident vertebrate predators (primarily fish), often harbor a unique and often diverse assemblage of organisms (Williams 1987). These wetlands serve as amphibian breeding sites and provide important food resources for migrating waterfowl, and consequently are of critical importance to waterfowl (Trochell and Bernthall 1998). Biological condition was evaluated in terms of relative community comparisons with reference wetlands (a subset of least-impacted or undisturbed wetlands sampled in this study). The response of various community attributes (e.g., taxa richness, relative abundance, indicator organisms) to disturbance was examined as a means to select metrics for incorporation into the multimetric index. Index scores were used to rate the biological integrity of each wetland. Biological rarity of wetlands was measured both in terms of their ability to support individual species deemed to be rare (i.e., regionally scarce) and community attributes, including complexity and biodiversity.

We also documented the range in "natural variation" of aquatic organisms/communities in different classes of Wisconsin wetlands (as described in Wisconsin Administrative Code NR 103.03(1)) and describe the hydrological conditions necessary to support biological characteristics naturally present in Wisconsin wetlands (per NR 103.03(2)(e)). Through the careful selection and sampling of a set of least-disturbed reference wetlands representing the three major ecoregions of Wisconsin (Omernik 1987), we developed and tested the application of a multimetric index of ecosystem integrity that incorporates aspects of community stability and resiliency to disturbance.

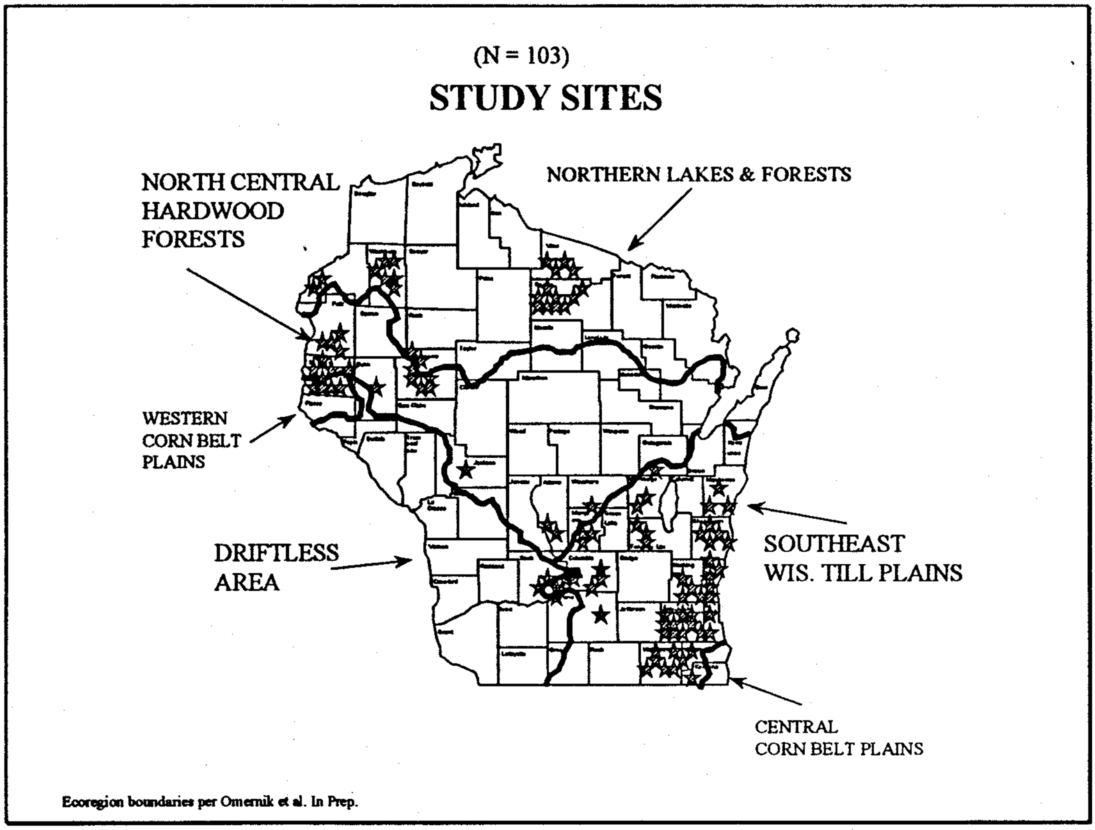
Due to the lag time necessary to process the large number of laboratory macroinvertebrate samples collected, this study spanned a two-year period. Field work was conducted in the spring and summer of 1998, laboratory work was completed by the following spring, and metric development and testing was performed in the summer of 1999. This report constitutes the completion of the project, but further testing and application of the indices will be required prior to adoption as a useful tool in the management of Wisconsin wetlands.

# METHODS

## STUDY SITE SELECTION:

Wetlands were selected for the study with assistance from many individuals throughout the state. In the fall of 1997, we completed a mailing to over 50 wetland resource managers and scientists asking for their input and names and locations of candidate wetlands that would meet the following criteria. Sample sites were to be temporary wetlands situated in land-locked basins (i.e., palustrine or depressional). This restriction was intended to minimize biological cross-contamination effects from permanent water-bodies containing fish. We asked for wetlands representing different sizes, shapes, water duration, hydrologic class, dominant plant communities, and origin as distributed among the three major ecoregions of Wisconsin. Because the presence of aquatic macroinvertebrates was essential to our work, we needed wetlands that contained standing water through early May. We desired 25 least-disturbed wetlands in each of the three ecoregions (75 total) to serve as reference wetlands for index development. The other 25 wetlands (distributed across the state) were to represent various forms and levels of disturbance (or restoration and histories). Additionally, we wanted wetlands with either public access or cooperative landowners.

This recruitment effort was successful in that it generated a long list of candidate wetlands, but it also presented a difficult task to sift out wetlands that did not meet our criteria. As will be apparent when examining the wetlands that were actually sampled, not all criteria or hopes were met and large gaps in distribution were evident. Numerous phone calls were made to gather additional background information on various wetlands and narrow the list to the desired 100. In the end, we selected 103 wetlands (one wetland consisted of a double basin) to represent the three major ecoregions of Wisconsin (Figure 1). Characterization of the wetlands as to their origin, disturbance history, and water duration class will be presented in the findings section.



## FIELD SAMPLING:

### General:

We visited each wetland during early spring (mid-April to early-May) and mid-summer (July) of 1998. The purpose of the early spring sampling period was primarily to collect aquatic macroinvertebrates and water chemistry samples prior to pond desiccation and to minimize effects on community composition related to immigration (colonization) or emigration. The summer sampling period was intended to coincide with maximum emergent vegetation production, thereby enhancing the plant identification process. During each visit, we took 35mm photos and completed a field observation form (see Appendix A), which included riparian land-use data, water color and clarity observations, maximum water depth, a sketch or map of the wetland and its surrounding environment, and weather related data. We carefully recorded the location of each wetland on the hand drawn map by describing distances or relative positions to surrounding roads or other natural landmarks. We recorded the Township, Range, Section, and quarter-quarter locations on the field sheet and used GPS units to obtain decimal degree locations for the majority of the wetlands in the study. Eight wetlands were sampled at approximately one-month intervals in order to examine temporal changes in macroinvertebrate metrics (Table 1 below).

Table 1. Wetlands sampled periodically through dry season.

| Wetland | Months sampled | | | | | |
|---|---|---|---|---|---|---|
| | April | May | June | July | Aug | Sep |
| French Creek NE | X | X | X | X | dry | dry |
| French Creek West | X | X | X | X | dry | dry |
| Hwy Z kettle | X | - | X | X | dry | dry |
| John Muir | X | X | X | - | - | - |
| Old World Wisconsin | X | X | X | X | - | X |
| Patrick West | X | X | X | X | X | X |
| Scuppernong Prairie | X | X | X | X | - | X |
| W P & L South | X | X | X | X | X | X |

### Macroinvertebrate sampling:

Types of Macroinvertebrate samples:
- Concentrated D-frame Net Sweeps
- "Bagged" D-frame Net Sweeps
- Screened Samples

Two sets of macroinvertebrate samples were collected at each wetland during mid to late April 1998. The first set of samples consisted of a field-concentrated composite of three standardized D-frame net sweeps (each sweep 2 m in length) collected from representative shoreline substrates in water less than 60 cm deep. The position within each wetland from which the samples were collected generally represented a trisection of the wetland perimeter, but in some larger wetlands the samples were simply collected from three widely separated locations along one shoreline. The mesh size of the D-frame net was 800 by 900 microns (a smaller mesh size would clog and make sampling in many muck and peat bottomed wetlands impossible). The three net sweeps were composited in a bucket equipped with a #30 mesh (600 micron) screen, and the sample was then concentrated in the field by hand-washing or rinsing substrates. Large particles, consisting of plant stems, woody debris, leaves, etc., were shaken vigorously to remove attached or entangled macroinvertebrates, examined, and then discarded. The remaining litter and macroinvertebrates were transferred to one-liter containers and preserved with 95% ethanol.

A second set of three composited net sweeps, collected from three undisturbed stations spaced intermediately between the previous trisected stations, was transferred intact (i.e., without concentrating) to one or more 1-gallon plastic bags (lock-type) and preserved in 95% ethanol. This second set of macroinvertebrates was originally intended to serve as comparative data to determine what organisms, if any, we were losing using the net-concentration method. However, two conditions prevented us from completing this objective. First, laboratory processing of the smaller, net-concentrated samples took much longer than expected. The "bag" samples took even longer to process and ultimately required sub-sampling to make the process even remotely feasible. The increased variability introduced by sub-sampling, together with generally poorer sample preservation, discouraged us from processing these bulky, messy samples. However, the bag samples did serve as a backup for lost net-concentrated samples (none lost) or where the number of organisms in the net-concentrated sample was less than the desired minimum of 100 organisms. Both sets of samples were labeled appropriately in the field and transported to the laboratory for processing.

On an additional subset of 17 wetlands, we employed a third method of collecting macroinvertebrates modeled after the Minnesota wetland biotic index studies (Helgen and Gernes 1996). The contents of two net sweep grabs were placed on a coarse screen (0.5 by 1.25 inch grid) above a rectangular plastic container and allowed to sit for approximately five minutes. We turned the substrate material over once during the process. Macroinvertebrates and detritus that fell or crawled through the mesh into the container below were transferred to one-liter containers and preserved with 95% ethanol.

<u>Plant Surveys</u>:

Plant Samples:
- Subjective Cover Estimates
- Transect surveys
- Voucher specimens

We conducted plant surveys during July 1998 using a combination of techniques. The first method was a simple, subjective estimate of cover. We drew a sketch of the wetland plant cover and estimated percent cover of total emergent, submergent, and floating-leafed vegetation, and open water for each wetland. Within each plant cover type, we recorded the plant taxa (family or genus level) present and their relative abundance on a scale of 1=present and rare, 2=occasional, 3=common, 4=abundant, 5=co-dominant, and 6=clearly dominant.

The second method of assessment was more objective. We conducted quantitative surveys of percent cover and frequency of occurrence within six equidistantly spaced 20 by 50 cm rectangular quadrats (Daubenmire 1959) positioned along each of three transects that trisected the basin (total of 18 quadrats per wetland). Sampling proceeded from the 60 cm depth contour to shore. In those wetlands which had dried up or had very little standing water in July, each transect extended from the center of the basin to a point where the shoreline had been during April. Specimens not identified in the field were collected and pressed for voucher preparation. Voucher specimens are divided between the WDNR research collection and the UW-Madison herbarium (deposited with T. Cochrane).

Riparian Data:

Riparian cover type surrounding the perimeter of each wetland was classed as one of the following classes: woods, shrubs, grass, wetland, urban, and agriculture. The percent cover of each riparian type within the 100-foot buffer zone was estimated visually (generally to the nearest 5%). Agriculture included land currently in agricultural production (grain and row crops) and pastured land. Urban included commercial, industrial, and residential areas including lawns. Grassland included prairie restorations and former agricultural lands reverting to natural cover. In addition to shoreline land cover, we estimated shade canopy (trees) cover above each wetland at noon.

Physical, chemical, and miscellaneous biological data:

We collected water samples for pH, alkalinity, and conductivity measurements at each wetland during the April 1998 macroinvertebrate surveys. Water samples were collected from a central basin location (undisturbed) within each wetland with a 250 ml plastic jar provided by the Wisconsin State Laboratory of Hygiene (SLH) and placed in an iced cooler until delivered to the SLH in Madison within one week of collection. Other field measurements taken included air and water temperature (standard pocket thermometer), apparent water color (subjective), water clarity (categorical), and field conductivity (meter) on a subset of wetlands. We also collected zooplankton samples (grab bucket filtered through 60 micron mesh), and we recorded the presence of algae blooms or excessive amounts of duckweed. We completed a field observation form (Appendix A) for each wetland, which included a rough sketch or map of the wetland and its surrounding environment, and completed a standardized WDNR Rapid Assessment Form with Amphibian supplement. We also categorized the dominant type(s) of bottom substrate material within the 0-60 cm water depth zone.

## LABORATORY METHODS:

Macroinvertebrates:

| TWO-TIERED PROCESS |
|---|
| • Random 100+ Count<br>• Total Count |

We used a two-stage procedure to process all macroinvertebrate samples. Each sample was rinsed with tap water on a #30 mesh sieve to remove the ethanol and then placed in a grid-marked tray with 24 cells. Samples were picked under lighted 3X magnification. Cells were randomly selected and macroinvertebrates were removed from the detritus and identified to order or family level, as appropriate, until a minimum of 100 organisms were picked.. In general, aquatic insects were identified to family; all other taxonomic groups were identified to order or class (please see Table 5 for a complete taxonomic listing). This method essentially follows the procedure established by Hilsenhoff (1982) in the stream arthropod biotic index. Once a cell was started, all organisms in that cell were picked and included in the 100-count. All data and specimens representing the 100-count subsample were kept separate from the remainder of the sample. Following completion of the 100-count pick, all organisms in the balance of the sample were picked, identified, and placed in vials for further examination and/or voucher preparation in 70% ethanol. We processed the entire contents of the 17 "screened" samples although most were relatively clean and contained less than 100 total organisms. As mentioned earlier a few of the backup "bag" samples were processed to supplement a few net-concentrated samples that did not contain a minimum of 100 organisms.

Training of staff and Quality Assurance/Quality Control issues: staff received orientation and training in processing macroinvertebrates prior to working independently on wetland samples. The senior biologist verified all identifications for the first 10 samples and randomly checked 10% of all samples thereafter. Taxa identification was deemed adequate when the number of incorrectly identified taxa was less than 10% of the total taxa and the number of total specimens incorrectly identified was less than 10% of the total number of organisms in the entire sample. The senior biologist also repicked the first 10 sample remnants (detritus remaining after the staff completed processing a sample) as an additional quality control check. Sample processing was judged adequate if the number of new taxa recovered was less than 10% of the original number of taxa and the number of additional organisms recovered was less than 10% of the original total. After the initial 10 samples were processed and checked as outlined above, we randomly chose one sample for QAQC checks from each batch of 10 samples processed thereafter. If that sample failed to meet QAQC criteria, the remaining nine samples in that batch were assumed to also fail, and each sample was checked for Ids and sample remnants were searched. If the random sample passed the QAQC check, all samples in that batch were assumed to meet the QAQC criteria.

We processed a total of 165 macroinvertebrate samples. Of this total, 148 were net concentrated samples and 17 were "screened" samples. Of the 148 net samples, 16 were duplicate or triplicate field samples, and of the remaining 132 samples, 104 were April samples and 28 were monthly samples from the eight basins so sampled (see Table 1).

Plants:

Field plant specimens were identified to family, genus, or species as far as possible. At least one taxonomic voucher was prepared for each taxon encountered. Vouchers of unknowns and specimens of uncertain determinations were delivered to the University of Wisconsin – Madison herbarium for identification and/or verification. All other voucher specimens were retained for the WDNR research herbarium collection. Duplicate or replicated specimens were not retained and were discarded. A record of laboratory identifications for each wetland was compiled and matched with field notes concerning unknowns or questionable field identifications, and the necessary corrections to field identifications were made accordingly.

Water Chemistry:

Samples for water chemistry analysis were completed under contract with the Wisconsin State Laboratory of Hygiene. The measurements conducted in this study are described in the SLH's 'Manual of Standard Operating Procedures Inorganic Chemistry Unit', Method 115.1, automated alkalinity, pH, and conductivity. Duplicative QC limits for alkalinity are 1.08% < 250 and 0.95% >250, and for conductivity 1.69% <100 and 1.24% > 100.

## DATA ANALYSIS:

Field sheets and field notes were reviewed each week by the senior staff member for completeness and consistency. Backup copies of data were made for security purposes. Data were transcribed into a computerized data base, and data transcription errors were detected by comparing computer calculated sums of macroinvertebrates with manual sums as recorded on laboratory sheets. Discrepancies triggered recalculations of laboratory sums and examination of individual taxon data for errors. Errors were corrected and sums were recalculated to confirm that the computer data matched lab totals. We used SYSTAT (SPSS 1997) to perform all statistical and graphical analyses. Percentage data were transformed using the arc-sine square-root transformation, and abundance data were either log-transformed (X + 1) or power-transformed as applicable. Metric development was based on a series of visual comparisons of community attribute responses to suspected measures of disturbance using box plots and jittered dot density plots (SYSTAT). Those attributes that exhibited evidence of separation between reference wetland conditions and wetlands known or wetlands suspected to be impacted by human disturbance were selected as potential metrics. Attributes that exhibited inconsistent or overlapping responses between impacted and reference systems were eliminated from further examination.

# FINDINGS

## GENERAL CHARACTERIZATION OF WISCONSIN DEPRESSIONAL WETLANDS:

Locations: 103 wetlands (104 basins) were sampled during this study (Figure 1 and Appendix B). The majority of wetlands were located in the Southeast Wisconsin Till Plains (N=58). Twenty-six wetlands were in the Northern Lakes & Forests, and 18 wetlands were from the North Central Hardwood Forests. Two wetlands were just across the border in the adjacent Western Corn Belt Plains.

Wetland Typology: Definitions of wetlands and types of disturbances are provided in Tables 2 and 3, respectively. Of the 104 wetland basins included in this survey, the majority represent undisturbed or only moderately disturbed "natural" wetland ecosystems (67), while the balance represent either restored (24), created (5), or heavily disturbed wetlands (8). Most are either "kettle" (46) or "prairie" (32) depressions. Eight wetlands are diked wetlands. The rest include a few representative bog (7), fen (2), interdunal (3), seep (2), lacustrine (2), and riverine (3) wetlands. In terms of disturbance, 47 wetlands represent least-disturbed or undisturbed systems that have no visible signs of impact. The balance are disturbed (or suspected to be disturbed) to varying degrees by one or a combination of the following: roads (18), substrate excavation (9), agriculture-current (8), agriculture-past (8), houses or buildings (2), railroad (1), pasture (1), beaver dam (1), lake influence (2), and managed watersheds (11). The later group of wetlands is managed as grassland for waterfowl production by periodic burning.

Table 2. Definitions of wetland classes used in this study.

| Type: | Definition: |
|---|---|
| Kettle | Primarily steep-sided depressions in woodland settings (but land may be managed otherwise). See also Bog. Includes some wooded, vernal pools. |
| Prairie wetland | Depressions in grassland settings; generally flatter, gentle slopes; includes some vernal depressions. |
| Fen | Groundwater-fed; chemistry generally alkaline; may be springs with surface flow. |
| Seep | Groundwater-fed wetland representing springs, snow-melt, or some combination of surface runoff (normally non-alkaline); may be flow-through or discharge area; possibly intermittent. |
| Bog | Deeper, more permanent kettle hole with floating mat either around perimeter or in central basin with open margins. Mat may contain sedges, spahgnum, leatherleaf, or tamaracks. |
| Interdunal | Wetland formed between dune swales adjacent to Great Lakes. |
| Diked wetland | Wetland created or restored by artificial damming of drainage-way. |
| Riverine | Wetland system interconnected during spring snowmelt or high water to adjacent river or stream. |
| Lacustrine | Wetlands connected to a lake (not boggy in nature). |

Table 3. Types of disturbances influencing wetlands in this study.

| Class: | Description: |
|---|---|
| None | Basically undisturbed, natural, unaltered. |
| Road | A nearby road may contribute road salt runoff or interfere with hydrology. |
| House | A nearby building may or may not impact wetland. |
| Railroad | Railroad tracks may impact wetland hydrology or chemistry. |
| Agriculture | Current agricultural practices probably contribute to some degree of impact on wetland; nutrient, sediment, or herbicide/pesticide runoff likely. |
| Old Agriculture | Land surrounding wetland was formerly in agricultural production; land now reverting to grass or woods, not managed (see below). |
| Excavation | Scrapes or dug-outs; sediment disturbed during construction or restoration. |
| Managed | Former agricultural lands (Old Ag.) that are currently managed by state or federal agencies as grassland to enhance waterfowl production. |
| Pasture | Extensive portion of wetland riparian area or watershed managed as pastureland, with extensive grazing by cattle, sheep, etc. |
| Lake | Biota probably influenced by interconnections to permanent water-body containing fish. |

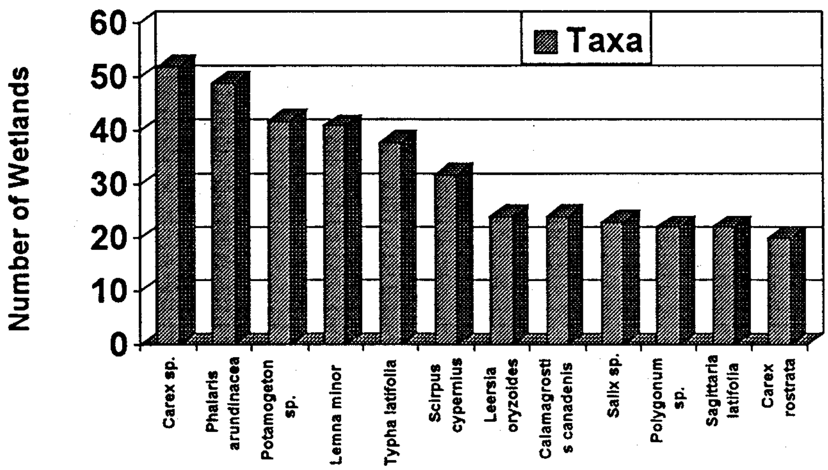
In an earlier study of 54 Wisconsin wetlands, Lillie & Garrison (In Prep.) found that water duration was the single most significant of 12 measured environmental factors associated with the structure of macroinvertebrate communities (based on canonical correspondence analysis). Consequently, the distribution of wetlands by water duration (defined as the number of months of standing water during the non-ice period) is an important consideration in interpreting the macroinvertebrate data in this study. Sixteen wetlands had relatively short water duration as they were dry during the July plant surveys. Another 45 wetlands had maximum surface water depths in July less than 40 cm and, with the exception of a few of the bog-type wetlands, probably dried up before the end of the summer. Two of these basins were episodic, rain-filled depressions and, as will be discussed later, should not have been classed as wetlands. The remaining 43 wetlands had deeper standing water in July and, consequently, are believed to hold water throughout the year (in a typical year). A very severe drought, which occurred during 1998 in the northern half of the state, caused several wetlands known to have typically longer water duration to dry out prior to the July plant surveys. However, because the drought occurred after the April sampling period, the macroinvertebrate communities present in the April samples should reflect the previous year's conditions.

Riparian Zone characteristics: The predominant riparian cover type within the 100-foot buffer zone around each wetland was woodland (> 50% cover in 59% of all wetlands). Grassland was the next most important cover type (36%). Although only two wetlands had more than 50% urban or agricultural land cover, 21% of the wetlands had either some urban or agriculture influence in the 100-foot buffer zone. Shade cover (tree cover above wetland) was less than 5% in about half of the wetlands surveyed; only about 17% of the wetlands had a canopy cover greater than 50% cover.

Basin characteristics - physical, chemical, and biological attributes: Wetlands ranged in size from small woodland pools less than 0.1 acre to a large 27 acre wetland restoration site. The majority of wetlands (59%) were less than 1 acre; 31% were between 1-5 acres; and the balance (10%) were greater than 5 acres. Most wetland waters were lightly-stained (70%) or darkly-stained (14%), with pH units in the 6.0-7.0 (52%) or 7.0-8.0 (43%) range. Algae blooms were observed in only 5% of the wetlands during the April sampling period, while 15% of the wetlands had dense duckweed cover. The occurrence of algae blooms and dense duckweed increased to 23% and 33%, respectively, by the July sampling period. The predominant bottom substrate was mud (45%), combinations of mud with other substrates (26%), purely organic (12%), leaf (10%), peat (4%), roots (11%), or inorganic (5%). About 10% of the wetlands had some form of floating mat either around some portion of the wetland perimeter or throughout the entire basin.

Plant Community characteristics: Wetland plant cover, as estimated visually according to major plant community type, was dominated by emergents (68%). Thirteen percent of sampled wetlands had a significant submergent plant cover, and another 13% were dominated by the floating-leafed community. Twenty-six percent of the wetlands had areas of 50% or more open water (note: totals exceed 100% due to co-dominance among some plant types). The dozen most commonly occurring plant taxa are shown in Figure 2 below. The leading dominant plant taxa as determined by the frequency of occurrence of the top 3 relative importance values (based on the average of percent cover and frequency of occurrence) per wetland (see Figures 3-5; provided at end of text) were Carex spp. (17%), duckweeds (14%), reed canary grass (7%), other grasses ( 6%), pondweeds (6%), cat-tails (4%), and various bulrushes (4%). Lemna minor was the most commonly occurring dominant species and Carex was the dominant genus (Figure 3).

Figure 2. Most Common Plant Taxa



Macroinvertebrate characteristics: A total of 94,793 macroinvertebrates were collected from the 104 wetland basins during the course of the study (partial list in Appendix E). In order of ranked abundance, midges (24,928), mosquitoes (10,555), clams (13,126), and snails (8,883) comprised 60% of all organisms captured. Mollusks (snails and clams) were the dominant macroinvertebrate in reference prairie wetlands (Figure 6), and shared dominance with mosquitoes in reference kettles (Figure 7). Midges dominated impacted prairie wetlands and impacted kettles. Scuds (primarily *Hyallela azteca*) frequently dominated managed prairie wetlands. Midges also were frequently dominant in bogs and other wetland types (Figure 8). Proportionately, midges were more frequently dominant in disturbed and restored wetlands than in natural wetlands (Figure 9). Clams and mosquitoes were the second and third most frequently dominant organism in natural wetlands. Characterization of macroinvertebrate communities in wetlands must recognize differences attributable to water duration and location. Mosquitoes were more frequently dominant in short duration wetlands, while midges and mollusks almost exclusively dominated long duration wetlands (Figure 10). Wetlands with medium water duration (i.e., temporary) were occasionally dominated by caddisflies, phantom midges, and a mixture of taxonomic groups, but midges, clams, and mosquitoes continued to predominate. Differences in macroinvertebrate dominance among ecoregions were evident (Figure 11). Wetlands located in the Southeast Wisconsin Tills Plain and North Central Hardwoods Forest ecoregions were mostly dominated by midges, while mosquitoes dominated Northern Lakes and Forests ecoregion. Many of the ecoregional differences reflect differences in the types of wetlands sampled in each region (e.g., mostly wooded kettles in the north and open prairie wetlands elsewhere) rather than due to basic compositional differences among macroinvertebrate communities. Ideally, such comparisons should be made between wetlands of the same physical, hydrogeomorphical, and land cover characteristics.

# METRIC DEVELOPMENT

## Wetland Habitat Index:

The WETLAND HABITAT INDEX is derived from six attributes and is used to predict general habitat conditions for macroinvertebrate production in depression basin wetlands.

A metric has been defined as "...a measurable component of a biological system with an empirical change in value along a gradient of human disturbance" (Danielson 1998). The response of an attribute (i.e., potential metric) is usually plotted as the dependent variable along the y-axis against some measure of human disturbance (independent variable) along the X-axis as a means to identify candidate metrics. This procedure may appear quite straight-forward in theory, but some complications arise due to difficulties in measuring "human disturbance". While it was relatively easy to measure various macroinvertebrate and plant community attributes in this study, we did not have a clear picture of what represented the human disturbance gradient. Initial efforts at identifying potential metrics were confounded by the natural variability existing in the data base. Natural differences in macroinvertebrate community structure masked or otherwise counteracted impacts produced by human disturbance. It was not a surprise to discover that bogs, fens, and seeps have relatively distinct communities from other depression type wetlands. However, we also discovered that kettle wetlands differed structurally from prairie wetlands. These differences together with our inability to identify the "best' human disturbance gradient forced us to take a somewhat different approach at selecting metrics. First we developed a wetland habitat rating for macroinvertebrates based on a combination of six subjective and objective criteria (Table 4, below). This rating index represents a prediction of wetland habitat condition for macroinvertebrate production. The suspected influence of human disturbance is designed into the index as follows. Within the land use attribute, urban and agricultural impacts are assumed to have detrimental impacts on macroinvertebrate production. This is not necessarily true because small amounts of nutrient inputs into an otherwise oligotrophic wetland might prove "beneficial" depending upon which macroinvertebrate community attributes are identified as metrics. Likewise, the algae & duckweed scores reflect supposed nutrient gradients, and the assumption is made that more nutrients lead to increased frequency of blooms and that blooms are detrimental to macroinvertebrate production. This also can be debated. The water color-turbidity gradient also assumes an association exists between water quality and conditions favorable for macroinvertebrate production, which may or may not exist. Highly stained waters resulting from accumulations of tannic or organic acids occur naturally, and high color is generally associated with low pH conditions that are unfavorable for macroinvertebrate production. The colonization potential scores are based on the consensus gained from numerous wetland studies of macroinvertebrate communities, which generally illustrate that a diverse mixed plant community is more beneficial to macroinvertebrates than dense or very sparse communities. The watershed disturbance history attribute makes a number of assumptions that are acknowledged to be inaccurate in many circumstances. The scores given to restored and created wetlands represent expected averages; some restored or created wetlands actually may provide more suitable habitat for macroinvertebrates than unimpacted wetlands. Lacking additional data on the history of an individual wetland, an intermediate score (3-6) is assigned. The natural disturbance attribute is based primarily on the assumption that an intermediate period of standing water is desirable from the viewpoint of macroinvertebrate communities. As water duration increases from very short to intermediate, taxa richness increases (primarily due to increase in predators) and food web chains lengthen (Schneider and Frost 1996). A shift in water duration from intermediate to permanent may be accompanied by a decrease in taxa richness as the system becomes more stable (hydrologically) and food web linkages become more simplified. In previous investigations of Wisconsin wetlands, we found taxa richness and abundance to be highest in basins of intermediate water duration. Longer duration wetlands contained numerous predators that reduced total macroinvertebrate populations, and very short duration wetlands generally contained fewer taxa and lower abundance.

## Table 4. WETLAND HABITAT RATING FOR MACROINVERTEBRATES

| Attribute/scale | 10-9 | 8-7 | 6-5 | 4-3 | 2-1 | 0 |
|---|---|---|---|---|---|---|
| LAND USE: Dominant Riparian Land-use in 100 ft buffer | Wetland | Wooded | Shrub | Grassland | Agriculture > 10% (over-rides natural covers) | Urban > 10% (over-rides natural covers) |
| TROPHIC: Duckweed – Algae status (surrogate for nutrients) | Both absent, both seasons | One present, one season only | Either one, both seasons | One both seasons, one only one season | Both present both seasons | Heavy blooms, both seasons |
| CLARITY: Water clarity, turbidity, color | Clear, both seasons | Only one condition present in one season | Either dirty or dark-stained in both seasons, but not both together | Both dirty or stained in one season, and one condition in other season | Both dirty and stained in both seasons | Both very turbid and dark-stained or colored in both seasons |
| Colonization potential: Habitat substrate conditions * A. vegetation B. sediments | Hemi-marsh conditions with good mixture of plants and sediment types | Some evidence of vegetative dominance with mixture of bottom substrates | Generally one or two plants dominant, with mud, roots, or leaf-pack bottoms | Fewer plants, mud or leaf-pack, or occasionally peat bottom | Very few plants, generally leaf-pack or mud bottom | No plants, mud, silt bottom with sedimentation or other signs of disturbance |
| WATERSHED: Watershed – disturbance history | None – pristine conditions | Minimal impacts possible; managed WPAs, etc.. | Restored wetlands | Created wetlands | Moderately-impacted, hydrology altered | Severely-impacted, sedimentation, hydrology, or toxics |
| NATURAL: Natural Disturbance – Hydroperiod | Temporary to semi-permanent (5-7 months), not permanent | Other types, including moderately-short duration (3-4 months) and long-duration or permanent wetlands (8-9 months): assign a score of "3-8", giving some consideration to abnormal precipitation or drought conditions. | | | Very short water duration, less than 2 months | Fish present – or known inter-connection with permanent waterbody |

* floating bog mats without standing water rated separately.

The sum of the individual habitat rating scores for each wetland represents the subjective Wetland Habitat Index. The index score represents the general, overall, environmental conditions for macroinvertebrate production, taking into account both human disturbance and natural disturbance. Consequently it should reflect potential macroinvertebrate biotic integrity potential. Large disparities between predicted biotic integrity and measured biotic integrity should be informational in detecting causes of impacts (see last section). Habitat index scores of wetlands in this study ranged from 24 to 54 (mean 41.6, mode 47).

The Wetland Habitat Index represents the average of six factors that could influence macroinvertebrate community structure. However, macroinvertebrate production does not necessarily relate to average conditions, but rather may respond to a single limiting resource condition. Consequently, we also computed a **limiting resource index**, which was the lowest score (0-9) recorded for each wetland among the six attributes used in the Wetland Habitat Index. This limiting resource index score may be a more accurate predictor of macroinvertebrate production than the averaged Wetland Habitat Index rating. We address this issue in another section. Limiting Index scores ranged from 0 to 8 (mean 3.4, mode 4).

## WISCONSIN WETLAND MACROINVERTEBRATE INDEX

> The WWMI is a multimetric index based on 15 macroinvertebrate metrics derived from a total count of organisms in three composited net sweeps. The WWMI is used to rate, rank, or compare wetland biological condition.

Development of the Wisconsin Wetland Macroinvertebrate Index (WWMI) was a three-stage process. We did not apply the standard procedure of plotting community attributes (dependent response variables) versus some selected measure of human disturbance (independent variable) for several reasons. First, we believed that none of our measured environmental variables accurately portrayed human disturbance. Percent agriculture and percent urban area in the riparian zone may have been adequate had we included a larger number of wetlands with degraded riparian zones in the study. Only a small fraction of the wetlands included in the study were heavily impacted, and the majority of wetlands that were classified as "impacted" were only minimally impacted. To further complicate matters, we had an imbalance in wetlands types represented in the study. Among the kettle wetlands, we had 33 reference or least-disturbed representatives and only 17 "impacted" kettles. Among the prairie wetlands, we only had five reference basins; the remainder of the prairie wetlands consisted of 19 "impacted" and 11 managed wetlands. Upon examination of the data, we now suspect that many of the wetlands categorized as "impacted" were not seriously degraded, and some "impacted" wetlands may have been in better condition (from the point of view of providing macroinvertebrate habitat) than some so-called reference sites. An additional complication, anticipated by our earlier research (Lillie & Garrison, In Prep.), was the masking influence of water duration on the macroinvertebrate communities. Because macroinvertebrate community composition differs dramatically across the water duration continuum (function of life cycle dependencies related to drought and predation), water duration or some surrogate thereof should be accounted for in searching for metrics and assigning index scores.

The above factors forced us to use an alternative approach in choosing metrics and in developing an index scoring system. We began by examining the frequency of occurrence of the various taxonomic groups and eliminated those taxa that occurred too infrequently to be of any practical value in metric development. As a precaution against tossing out infrequent but potential "indicator" taxa, we plotted abundance (or taxonomic richness) of each group against the water duration gradient to identify those taxa that were water duration dependent (exhibited either a trend or were restricted to long or short duration wetlands). Those taxa that were infrequent but restricted to long or short duration wetlands were reconsidered for inclusion as a potential metric. Data transformations were necessary to convert taxa abundances to a useful scale for assigning scores, including various power and logarithmic transformations (see Table 5). Of the original 69 community attributes tested, 40 were dropped from further consideration. We then examined labeled bi-plots (labeled as to reference – impact class) that compared the response of community attributes versus water duration (summary in Table 5) as a means to select metrics that appeared to be influenced by human disturbance. Those community attributes that exhibited some degree of separation between impact and reference wetlands were selected as metrics for inclusion in the multimetric index. In all cases but one (water boatmen) the direction of the response exhibited by the selected metric corresponded with the expected response to disturbance.

Fifteen metrics were selected for inclusion in the WWMI (Table 6). As noted, several metrics have restricted uses. Some perform better in prairies than kettles; some are only useful in long or short duration wetlands. Some adjustments in expectations seem appropriate when assigning scores among the various wetland types. However, due to the limited number of basins falling into each of the various wetland classes (e.g., short duration, reference kettles; long duration reference prairie wetlands, etc.) we have developed a single scoring scheme for all wetland classes. These scores may need to be modified or adjusted as more data become available. To assign scores to the 15 metrics we examined two sets of bi-plots between community attributes and water duration. The first set of observations was restricted to only reference kettles (there were too few reference prairies to use same method for prairies), which allowed us to view the distribution of data in what was believed to represent least-disturbed wetlands. For those metrics that declined with human disturbance, we used the reference wetland plots to establish index scores. For those metrics (e.g., pigmy backswimmers and soldier flies) that increased with human disturbance, we compared the reference plots with plots using all data points including disturbed wetlands. In the latter case the total absence or an excessive abundance of the taxon may represent poor or inadequate conditions while intermediate abundance may represent optimal or natural conditions. Consequently, we used the combination of plots to assign scores to several metrics. For those metrics that exhibited a response to water duration, we adjusted scores as noted in Table 6. This adjustment may not be appropriate however, in that it boosts index scores for either short or long duration wetlands where such an adjustment may not be warranted. An alternative might be to use a different set of metrics for short, medium, and long duration wetlands.

The Wisconsin Wetland Macroinvertebrate Index (WWMBI) is composed of 12 abundance metrics, two richness metrics, and one percentage metric (Table 6). Mollusks (clams and snails), annelids (worms and oligochaetes), and Anostraca (fairy shrimp) are non-insect groups that generally are more abundant in natural, undisturbed wetlands. The number of total non-insect taxa groups is greater in reference wetlands. Among the insect taxa, damselflies, water boatmen, limnephelid caddisflies, total caddisflies, phantom midges, and mosquitoes decline with disturbance, while pigmy backswimmers and soldier flies increase with degree of disturbance. Total invertebrate abundance and taxa richness both decline with disturbance. Metric scores were assigned as follows. For those attributes that declined as disturbance increased, we drew a line through the distribution of data points (reference kettles only) that represented the approximate 95th percentile. The remaining data points were trisected and scores assigned as follows: the upper third = 5, middle third = 3, and the lower third = 1. In most cases, the complete absence of a particular taxon in a wetland was scored as a zero. Because fairy shrimp are generally naturally absent from long duration waterbodies (8-9 months), we assigned a score of 5 to all long duration wetlands, irrespective of the abundance of fairy shrimp present. Likewise phantom midges (Chaoboridae) are generally restricted to medium and long duration wetlands, so wetlands less than four months in duration were scored as a 5. Mosquitoes also may deserve an adjustment, particularly in longer duration wetlands where their presence may suggest an abnormal condition indicating the lack of predators, which should be controlling their numbers. We did not make this adjustment because there were too few data points in the range to reach a decision. For soldier flies (Strayiomyidae), we drew a line through the distribution of all data points (all wetlands except "others") that represented the 95$^{th}$ percentile. The remaining data were trisected and scores were assigned as follows: the upper third = 1, the middle third = 3, and the lower third =5. Because soldier flies were generally found only in wetlands with water durations greater than four months, we assigned a score of 5 to all short duration wetlands. Pigmy backswimmers (Pleidae) were assigned scores adjusted as follows. First because pigmy backswimmers were restricted exclusively to long duration wetlands, all wetlands with water durations less than seven months were assigned a score of 5. In longer duration wetlands a small number of backswimmers per sample appeared common in natural systems. Hence, scores were assigned at an increasing level from 0 to 10 (see Table 6). However, greater abundances of backswimmers in a wetland were believed to be associated with increasing levels of algae and duckweed and consequently were assigned lower scores.

As noted in Table 6, some metrics are more applicable to kettle wetlands while others appear to be more appropriate in prairie wetlands. Similarly, some further adjustments may be required for water duration and perhaps for other environmental variables as well (e.g., pH, alkalinity, vegetation dominant). This particular set of metrics is designed for use in prairie and kettle wetlands; a different set of metrics may be required for bogs, fens, lacustrine, and riverine wetlands. Some metrics may be redundant (e.g., 3 caddisfly metrics), and some weighting of metrics may be desirable to refine the system. The absence of particular taxonomic groups from a wetland may not be associated simply with human disturbance and consequently may not deserve a score of zero. Many natural causes may explain such absences, but until such time as we can model macroinvertebrate community composition in wetlands we should maintain the current practice. It may be advisable to drop the Limnephelid metric as it is the only taxa group that is relatively difficult to identify (young larvae) and it is somewhat redundant with total caddisflies. We have retained it for the current presentation because it may signify the presence and availability of sufficient supplies of coarse particulate matter in "natural" wetlands.

Table 5. Metric evaluation based on total macroinvertebrate sample data – April 1998 data only (N=113).

| Taxonomic Group | Community Attribute | Metric Potential & Response |
|---|---|---|
| | | |
| Gastropods (=Snails) | Abundance | Weak |
| Pelecypods (=Clams) | Abundance | Weak |
| Mollusks (Snails & Clams) | Abundance | Log 10 positive, good potential |
| Amphipods (=Scuds) | Abundance | Infrequent; generally restricted to wetlands with wooded shorelines or longer duration wetlands. |
| Isopods (=sowbugs) | Abundance | Too infrequent |
| Collembola (=springtails) | Abundance | Too infrequent |
| Arachnidae (=spiders) | Abundance | Too infrequent |
| Hydracarina (=mites) | Abundance | Too infrequent |
| Annelids (=worms) | Abundance | Possible with 0.3 power transformation; marginal? |
| Hirudinea (=leeches) | Abundance | Log 10, adjusted for negative slope with duration; poor, inconsistent. |
| Anostraca (=fairy shrimp) | Abundance | Log 10 positive; not useful in long duration wetlands. |
| Conchostraca (=clam shrimp) | Abundance | Too infrequent |
| Ostracoda (=seed shrimp) | Abundance | Too infrequent |
| Other non-insect invertebrates | Abundance | Too infrequent |
| "Nonbugs" (sum non-insects exclusive of mollusks) | Abundance | Log 10 positive with negatively sloped scale for long duration; marginal, inconsistent. |
| "NonTaxa" (all non-insects) | Taxa richness | Untransformed positive; good, best for prairies. |
| Unidentified mayflies (immature) | Abundance | Too infrequent |
| Caenidae (mayfly family) | Abundance | Too infrequent |
| Baetidae (mayfly family) | Abundance | Too infrequent |
| Other mayflies (misc. families) | Abundance | Too infrequent |
| Sum all mayflies | Abundance & richness | Too infrequent |
| Zygoptera (sum of damselflies) | Abundance | Log 10 positive; fair for kettles; not effective for prairies? |
| Coenagrionidae (family) | Abundance | Too infrequent |
| Lestidae (family) | Abundance | Too infrequent |
| Other damselflies | Abundance | Too infrequent |
| Unidentified Anisoptera (=dragonflies) – (immature) | Abundance | Too infrequent |
| Aeshnidae (family) | Abundance | Too infrequent |
| Libellulidae (family) | Abundance | Too infrequent |
| Corduliidae (family) | Abundance | Too infrequent |
| Odonate Taxa | Richness | Positive; uncertain use for short duration wetlands; poor, not effective. |
| Sum Odonates (all taxa) | Abundance | Log 10 positive, no separation. |
| Unidentified bugs | Abundance | Too infrequent |

| | | |
|---|---|---|
| Pleidae (=Pigmy backswimmers) | Abundance | Present in long duration wetlands; optimum scores for intermediate abundance; negative indicator? |
| Corixidae (waterboatmen) | Abundance | Weak; Power 0.3 transformed positive; some inconsistencies. |
| Other Hemiptera (=bugs) | Abundance | Too infrequent |
| Sum Hemiptera (all taxa) | Abundance | Same as for Corixidae |
| "Bugtaxa" | Richness | Positive (step function) adjusted for over abundance; poor, not effective. |
| Unidentified Caddisflies | Abundance | Too infrequent |
| Polycentropodidae (family) | Abundance | Too infrequent |
| Phyrganeidae (family) | Abundance | Too infrequent |
| Limnephilidae (family) | Abundance | Negative slope for wetlands with intermediate water duration only; very good, looks promising. |
| Leptoceridae (family) | Abundance | Too infrequent |
| Other trichopteran families | Abundance | Too infrequent |
| "Centcaddis" (of all inverts) | Percent | Possible: negative slope with duration, scoring in long duration wetlands uncertain; marginal in kettles; good in prairies. |
| Sum Trichoptera (all taxa) | Abundance | Possible: even or sloped scales are possible – not good for very short duration wetlands, but good for both kettles + prairies. |
| "Caddistaxa" | Richness | Too small of spread |
| Unidentified Coleoptera (=beetles) | Abundance | Too infrequent |
| Haliplidae (=Crawling water beetles) | Abundance | Too infrequent |
| Dytiscidae (=Predaceous diving beetles) | Abundance | Too infrequent |
| Hydrophilidae (=Water scavenger beetles) | Abundance | Too infrequent |
| Scirtidae (=Marsh beetles) | Abundance | Too infrequent |
| Curculionidae (=Weevils) | Abundance | Too infrequent |
| Other beetles | Abundance | Too infrequent |
| Sum Coleoptera (all taxa) | Abundance | Log 10 positive; negative slope with water duration; poor separation. |
| "Beetletaxa" | Richness | Untransformed positive; no separation. |
| Unidentified Diptera (=flies) | Abundance | Too infrequent |
| Chironomidae (=Midges) | Abundance | Log 2 with central optimum; positive slope with duration; no separation. |
| Ceratapogonidae (=Biting midges) | Abundance | Log 10 positive; no separation. |
| Chaoboridae (=Phantom midges) | Abundance | Log 10 positive; tapered for short duration wetlands; marginal, better in longer duration wetlands. |
| Tipulidae (=Crane flies) | Abundance | Too infrequent |

| | | |
|---|---|---|
| Culicids (=Mosquitoes) | Abundance | Log 10 positive; reversed scores for long duration wetlands? |
| Stratiomyidae (=Soldier flies) | Abundance | Log 10 negative; not valid for short duration wetlands; possible negative indicator? |
| Sum Flies (all taxa) | Abundance | Log 10 positive; central optimum; inconsistent. |
| Percent Flies | Percent of total | Untransformed negative; sloped for low to moderate alkalinity, straight for high alkalinity; inconsistent. |
| "Flytaxa" | Richness | Untransformed positive; step function for short duration; poor. |
| Total Invertebrates | Abundance | Log 10 positive; fair. |
| Total Taxa (all invertebrates) | Richness | Untransformed positive; marginal – may need adjustment between prairies and kettles. |
| "CentEOT" (Percent of Ephemeroptera, Odonata, and Trichoptera individuals) | Percent | Log 10 positive: negative slope with water duration; marginal, caddis alone may be fine. |
| E-O-T taxa (Ephemeroptera, Odonata, and Trichoptera taxa) | Richness | Untransformed positive; poor. |

Table 6. Assignment of scores for macroinvertebrate metrics included in the Wisconsin Wetland Macroinvertebrate Index.

| **Taxa Group** | **Attribute** | **Limitations** | **Response** | **Scores:** 0 | 1 | 3 | 5 | **Modifica tions** |
|---|---|---|---|---|---|---|---|---|
| Mollusks | Abundance | None | Decrease | 0 | 1-10 | 11-99 | >99 | - |
| Annelids | Abundance | none | Decrease | - | 0-10 | 11-25 | >25 | - |
| Fairy Shrimp | Abundance | Short-medium duration | Decrease | - | 0-8 | 9-25 | >25 | 8-9 mon. = 5 |
| Non-insects | Richness | Useful in prairies | Decrease | 0 | 1-2 | 3-5 | >5 | - |
| Damselflies | Abundance | Useful in kettles | Decrease | 0 | 1-2 | 3-15 | >15 | - |
| Pigmy backswimmers | Abundance | Longer duration only | Increase | 0 | 1-2 and >100 | 3-5 and 11-99 | 6-10 | < 7 mon. = 5 |
| Water boatmen | Abundance | none | Decrease | 0 | 1-4 | 5-10 | >10 | - |
| Limnephelids | Abundance | Best in intermediate water duration | Decrease | 0 | 1-10 | 11-50 | >50 | - |
| Caddsiflies | Percent | Redundant? | Decrease | 0 | <8% | 8-15% | >15% | > 7 mon. ? |
| Caddisflies | Abundance | May need duration adjustment | Decrease | 0 | 1-10 | 11-60 | >60 | ? < 4 mon. |
| Phantom midges | Abundance | Longer duration only | Decrease | 0 | 1-8 | 9-25 | >25 | < 4 mon. = 5 |
| Mosquitoes | Abundance | Short-medium duration? | Decrease ? | 0 | 1-10 | 11-99 | >100 | ? |
| Soldier Flies | Abundance | Long-medium duration? | Increase | - | <25 | 8-24 | <7 | < 4 mon. not used |
| Total Invertebrates | Abundance | none | Decrease | <15 0 | 150-500 | 500-1500 | >1500 | |
| Total Taxa | Richness | Base adjustment for kettles vs prairies? | Decrease | <5 | 6-11 | 12-19 | >19 | |

## THE 100-COUNT MACROINVERTEBRATE BIOTIC INDEX

> The 100-Count MBI is a multimetric index based on ten macroinvertebrate metrics derived from a random pick of 100 organisms found in three composited net sweeps. The 100-Count MBI can be applied in the field by experienced staff as a means as rapid bioassessment.

A second macroinvertebrate index was derived using the data from the 100-count data set. This index can serve either as a rapid field assessment tool or as a supplement to the laboratory derived WWMBI. Because the 100-count data are a subset of the larger total invertebrate data, abundance was artificially capped by the laboratory procedure employed. Consequently, we used the percentage of total organisms of each taxonomic group and selected richness measures in developing the 100-count Macroinvertebrate Biotic Index (100MBI). We essentially followed the same procedures described for the total count data set in selecting metrics and assigning scores with the following modifications. Percentage data were transformed using the arcsin square-root transformation prior to data analysis. Taxa that occurred infrequently or in very low percentages were eliminated as potential metrics. Next, we examined bi-plots of percentages (transformed) and water duration to identify taxa that were water duration dependent. Taxa that exhibited potential utility as a metric (i.e., plots showed some logical and consistent response) were examined more closely using a series of plots with wetland classes identified as to disturbance type. The results of these examinations are provided in Table 7.

Based on more careful examination of the data, we selected 10 metrics for inclusion in the field rapid bioassessment index – the 100-Count MBI (Table 8). Five metrics decline with disturbance and five increase with disturbance. Nine represent percentage metrics and one is a relative richness metric. Scores were assigned as described for the WWMBI. The relative percentages of mollusks, limnephilid caddisflies, total caddisflies, and EOT (sum of Ephemeroptera-Odonata-Trichoptera) taxa in a random subsample decline as disturbance increases. Pigmy backswimmers, water boatmen (and total bugs), chironomid midges, and soldier fly larvae all tend to increase with enrichment or disturbance. The number of non-insect taxa groups encountered in the 100-count random subsample also declined with an increase in disturbance. Pigmy backswimmers are generally restricted to longer duration wetlands (i.e., greater than 6 months standing water during the ice-free season). Consequently most short and medium duration wetlands will probably receive a score of 5 (due to the absence of pigmy backswimmers), whether they deserve it or not. Therefore, some consideration should be given to not using this metric in medium and short duration wetlands. There also appears to be a great deal of redundancy built into the 100-count index in that the total bug metric encompases the pigmy backswimmer and water boatmen metrics, and the total caddisfly metric probably reflects the limnephilid metric. It might be advisable to drop the limnephilid metric and rely on the total caddisfly metric alone. This would allow a simplification of the field identification procedures as all caddisflies would need only to be identified as caddisflies; family level separation would not be necessary. It would be desirable to eliminate the chironomid metric (many midge larvae are small and easily missed in field-picking), but it is not certain how this would affect the performance of other metrics because chironomids often comprise a large percentage of the total organisms in the current 100-count methodology. The fact that three metrics represent bug (=Hemiptera) taxa essentially gives more weight to the Hemiptera than may be warranted. It might be desirable to use total bugs and drop pigmy backswimmers and water boatmen. From a field perspective this would not save much time (it is easy to identify pigmy backswimmers and water boatmen), and the impact of this action on the performance of the "new" index would have to be evaluated.

Table 7. Summary of metric evaluation of 100-count macroinvertebrate sample data – April 1998 (N=113).

| Taxonomic Group | Community Attribute | Metric Potential & Response |
|---|---|---|
| Gastropods (=Snails) | Percentage | Higher in prairies than kettles; possible. |
| Pelecypods (=Clams) | Percentage | Good potential; better separation in prairies. |
| Mollusks (Snails + Clams) | Percentage | Has potential in prairies; but too much overlap in kettles. |
| Amphipods (=Scuds) | Percentage | Too infrequent |
| Isopods (=Sowbugs) | Percentage | Too infrequent |
| Collembola (=Springtails) | Percentage | Too infrequent |
| Arachnidae (=Spiders) | Percentage | Too infrequent |
| Hydracarina (=Mites) | Percentage | Too infrequent |
| Annelids (=Worms) | Percentage | Too infrequent, very slight potential. |
| Hirudinea (=Leeches) | Percentage | Too infrequent |
| Anostraca (=Fairy Shrimp) | Percentage | Too infrequent |
| Conchostraca (=Clam Shrimp) | Percentage | Too infrequent |
| Ostracoda (=Seed Shrimp) | Percentage | Too infrequent |
| Other Non-insect invertebrates | Percentage | Too infrequent |
| "Nonbugs" (sum non-insects exclusive of mollusks) | Percentage | Inconsistent, no pattern. |
| "NonTaxa" (all non-insects) | Percentage | Unclear, somewhat higher in prairies, but inconsistent in general |
| "NonTaxa" (all non-insects) | Richness | Positive response in reference wetlands; good potential indicator. |
| Aquatic Insects (all) | Percentage | Generally higher in kettles, but not useful |
| Unidentified mayflies | Percentage | Too infrequent |
| Caenidae (mayfly family) | Percentage | Too infrequent |
| Baetidae (mayfly family) | Percentage | Too infrequent |
| Other mayflies (misc. families) | Percentage | Too infrequent |
| Sum of all mayflies | Percentage | Too infrequent |
| Zygoptera (all damselflies) | Percentage | Too much overlap, not useful |
| Coenagrionidae (family) | Percentage | Too infrequent |
| Lestidae (family) | Percentage | Too infrequent |
| Other damselflies | Percentage | Too infrequent |
| Unidentified Anisoptera (=Dragonflies) | Percentage | Too infrequent |
| Aeshnidae (family) | Percentage | Too infrequent |
| Libellulidae (family) | Percentage | Too infrequent |
| Corduliidae (family) | Percentage | Too infrequent |
| Anisoptera (all dragonflies) | Percentage | Marginal, mostly in medium-long duration wetlands; higher in kettles, but not useful |
| Odonates (combined) | Percentage | Marginal, possibly useful in kettles only? |
| Unidentified Bugs | Percentage | Too infrequent |
| Pleidae (=Pigmy backswimmers) | Percentage | May be fair negative indicator in longer duration wetlands? |
| Corixidae (=Water boatmen) | Percentage | Potential negative indicator: better in prairies? |
| Other Hemiptera (=Bugs) | Percentage | Too infrequent |

| | | |
|---|---|---|
| Sum Hemiptera (all bug taxa) | Percentage | Some utility as negative indicator, follows pattern of Pleidae + Corixidae) – redundant? |
| Unidentified Trichoptera (=caddisflies) | Percentage | Too infrequent |
| Polycentropodidae (family) | Percentage | Too infrequent |
| Phyrganeidae (family) | Percentage | Too infrequent |
| Limnephilidae (family) | Percentage | Potentil positive indicator in short-medium water duration wetlands |
| Leptoceridae (family) | Percentage | Too infrequent |
| Other caddisflies | Percentage | Too infrequent |
| Sum Trichoptera (combined) | Percentage | May work well for short-medium water duration wetlands; positive indicator |
| Unidentified Coleoptera (=Beetles) – mostly larvae | Percentage | No pattern, great amount of overlap |
| Haliplidae (=Crawling Water Beetles) | Percentage | Inconsistent pattern; may be higher in impacted systems? |
| Dytiscidae (Predaceous Diving Beetles) | Percentage | Water duration important, patterns inconsistent, not useful (at this coarse taxonomic level) |
| Hydrophilidae (Water Scavenger Beetles) | Percentage | Not useful, inconsistent |
| Scirtidae (=Marsh Beetles) | Percentage | Too infrequent |
| Curculionidae (=Weevils) | Percentage | Too infrequent |
| Other Coleoptera | Percentage | Too infrequent |
| Sum Coleoptera (combined) | Percentage | Weak, inconsistent. |
| Unidentified Diptera (=Flies) | Percentage | Too infrequent |
| Chironomidae (=Midges) | Percentage | Slight potential as negative indicator; much overlap, better for prairies? |
| Ceratapogonidae (=Biting Midges) | Percentage | Slightly higher in disturbed systems, but too much overlap to be useful |
| Chaoboridae (=Phantom Midges) | Percentage | Higest in reference kettles; long-medium water duration only, weak, possible |
| Tipulidae (=Crane Flies) | Percentage | Too infrequent |
| Culicidae (=Mosquitoes) | Percentage | Possible; best for medium-short duration? Possible negative indicator in long duration?; many problems |
| Stratiomyiidae (Soldier Flies) | Percentage | In medium-long water duration only; marginal, higher in impacted or enriched systems = negative indicator potential |
| Other Diptera | Percentage | Too infrequent |
| Sum Diptera (combined) | Percentage | K > P; lack of separation |
| Total Taxa | Richness | Weak, too much overlap |
| EOT (sum of Ephemeroptera, Odonata, + Trichoptera) | Percetnage | Slight separation; moderate potential; highest in reference systems |

Table 8. Assignment of scores to macroinvertebrates used in the 100-Count MBI rapid assessment Index (scores based on arcsin square-root transformed numbers).

| Taxa Group | Attribute | Limitations | Response | Scores 0 | 1 | 3 | 5 | Modifications |
|---|---|---|---|---|---|---|---|---|
| Mollusks* | Percent | None | Decrease | 0 | 0-0.3 | 0.3-.7 | >0.7 | None |
| Non-Insects | Richness | None | Decrease | 0 | 1-2 | 3-4 | > 4 | None |
| Pigmy Backswimmers | Percent | Long duration only | Increase | - | > 0.3 | > 0 & < 0.3 | 0 | < 7 mon. = 5? |
| Water Boatmen | Percent | None | Increase | - | > 0.2 | 0.1-0.2 | < 0.1 | None |
| Total Bugs | Percent | None | Increase | - | > 0.5 | 0.3 - 0.5 | < 0.3 | None |
| Limnephildae | Percent | None | Decrease | 0 | > 0 & < 0.2 | 0.2 - 0.35 | > 0.35 | None |
| Total Caddisflies | Percent | None | Decrease | 0 | > 0 & < 0.2 | 0.2 - 0.4 | > 0.4 | None |
| Chironomidae | Percent | None | Increase | 0(?) | > 0.6 | 0.3 - 0.6 | > 0 - < 0.3 | None |
| Soldier Flies | Percent | None | Increase | - | > 0.25 | 0.05 - 0.25 | < 0.05 | None |
| EOT taxa | Percent | None | Decrease | 0 | > 0 & <0.2 | 0.2 – 0.4 | > 0.4 | None |

* dropped after further consideration; may be applied to prairies only?

# WISCONSIN WETLAND PLANT BIOTIC INDEX

> The WWPBI is a multimetric index based on nine plant metrics derived from transect data (18 quadrats) and is intended as a supplementary index to the WWMBI to rate, rank, and compare wetland biological condition.

We developed the Wisconsin Wetland Plant Biotic Index (WWPBI) applying the same procedures used in formulating the two macroinvertebrate-based indices. The WWPBI index is designed to serve as a tool for evaluating the biotic integrity of depression wetlands in Wisconsin. Although we identified many plants to the species level for research purposes, we believe that a practical tool for managers with limited botanical training should be based on easily identifiable taxa. Consequently, for the most part, we lumped taxa at various taxonomic levels (e.g., family, genus) or structural groups (e.g., grass-like, emergents) for analysis. We did include those species that were of common occurrence and were fairly easy to identify in the field (e.g., reed canary grass, rice cut-grass, woolgrass, and lesser duckweed). We used importance values (average of percent cover and frequency of occurrence) as the attribute of concern at the family-genus-species levels and percent cover for emergent, submergent, floating-leafed, and open water attributes. The number of total plant taxa per wetland was the only richness measure included. Most managers will be able to identify or otherwise separate the plant taxa in the field with a minimal of background training.

We evaluated 24 plant community attributes representing the major taxa groups found in 104 wetland basins (Table 9) as candidate metrics. The plant taxa attribute was the only richness measure examined, and included both identified and unidentified specimens found in the combined quadrat and general basin surveys. The majority of attributes tested were sums of importance values for all species in each taxa group (e.g., Carex IV = sum of individual Carex species). The importance value represented the average of the percent cover and frequency of occurrence of each taxon in the 18 quadrats surveyed within the 0-60 cm zone (please refer to methods section for more details). We only considered those taxa groups that commonly occurred and were easily identifiable. While some rare taxa undoubtedly would serve as good indicators, incorrect identifications could lead to many difficulties in developing a simple, field-employed, plant index. Consequently, we tried to keep the index simple and easy to apply. Admittedly, some taxa (e.g., Calamagrotis canadensis = Bluejoint grass) may at first be difficult to identify in the field, but with experience the investigator should be able to easily separate between look-alike species. The MaxIV attribute represents a dominance measure and is simply the highest IV of any taxon in an individual wetland. The "GoodIV" attribute is the sum of IVs of Carex spp., bladderworts, pondweeds, Leersia spp., Calamogrostis canadensis, arrowheads, spikerushes, smartweeds, and horsetails. The "GoodIV" was intended to serve as a measure of the more desirable plants in the wetland community. We also examined the four percentage attributes based on major plant community types (e.g., emergents, submergents).

We selected nine plant community attributes as metrics (two metrics were combined as an adjustment for longer duration wetlands) for development of the Wisconsin Wetland Plant Biotic Index (WWPBI) based on their response to potential sources of human impact (Table 10). The index is formed using one richness metric, seven importance value metrics, and one percentage metric. Four metrics, including the number of plant taxa, Carex Ivs, CalamIV (Bluejoint grass), and GoodIV (see list in table), demonstrate good positive associations with "natural" or relatively least-disturbed situations. Consequently these metrics are expected to decline with increased levels of human disturbance. RCGIV (reed canary grass), TYPHAIV (cat-tails), and DUCKWIV (Lemnaceae) are good negative indicators and respond to human disturbance by increasing in impacted wetlands. Because the original seven selected metrics were mostly members of the emergent plant community, the index held a bias in favor of less permanent wetlands. To counteract this bias, we adjusted the WWPBI by including a metric that would account for the submergent and floating leafed plant communities, which become increasingly important in longer duration wetlands. After screening all candidate attributes we choose a composite metric consisting of the average of the PONDIV (pondweed) and % floating-leafed attributes. Pondweeds (several species) commonly occur in undisturbed wetlands, and the coverage of floating-leafed plants generally was higher on reference wetlands than disturbed wetlands. The addition of this combination metric allows an "upward" adjustment ranging from 1 to 5 in the standard cumulative score. A number of other attributes showed some promise as metrics but were dropped for various reasons (Table 9). Of particular significance was the maximum importance value, which functioned as a pseudo dominance indicator. We hypothesized that high maximum dominance values would be associated with disturbed wetlands, but the data did not support this assumption. Several taxa, including sphagnum, spikerushes, and percent submergent, showed some promise but either were inconsistent or occurred too infrequently to permit their incorporation into the final index (Table 9).

The Wisconsin Wetland Plant Biological Integrity Index consists of eight component metrics (Table 10). We assigned scores to each of the selected metrics using the trisection technique as described for the macroinvertebrate metrics. The plant taxa metric may require some modification and further evaluation. We did not apply a consistent level of taxonomic resolution across the 103 wetlands. In some wetlands, sedges were collected and returned to the laboratory for species level identification, while in other wetlands, we combined all Carexes, for example, simply as Carex spp. Some grasses and other taxa were labeled as unknowns, and despite the collection of voucher specimens in some cases, we were unable to carry the identification beyond the family or genus level. Consequently, the inclusion of the plant richness metric is somewhat questionable. Ideally, the metric should be tested at a selected taxonomic resolution that is applied across all wetlands in the study. Some form of future adjustment to index scores may be required to compensate for natural differences in plant community composition within particular wetland types (e.g., sedge meadows) that occur among ecoregions or across the tension zone (Curtis 1959). The deep water community adjustment as proposed boosts index scores by 1 to 5 depending upon the pondweeds and other floating-leafed plants present. In retrospect, the assignment of 1 point for the absence of either two attributes seems unnecessary. Rather, a standard trisection of the two attributes, with no points assigned to absences, may have been more appropriate.

Table 9. Plant community attributes evaluated as potential metrics for the Wisconsin Wetland Plant Biotic Index.

| **Taxonomic Group** | **Community Attribute** | **Metric Potential & Response** |
|---|---|---|
| Total Plant Taxa | Richness | Marginal, slightly higher in natural systems but much overlap; better in prairies? |
| Dominance (Ivs) | Highest IV among all taxa | Lower in kettles; but pattern in prairies contrary to hypothesis; no use in kettles |
| Grasses (combined) | IV | Potentially useful, but inconsistent?; problem with zeroes |
| *Carex* spp. (all) | IV | Bogs in separate class; higher in reference kettles; lower in impacted kettles and prairies; good potential metric |
| Reed Canary Grass | IV | Good negative indicator! |
| *Sphagnum* moss | IV | Too infrequent; of potential use |
| Cat-tails (=*Typha*) | IV | Good negative indicator |
| Bladderwort (=Utricularia) | IV | Too infrequent; long duration only |
| Duckweeds (*Lemna, Wolffia, + Spirodela)* | IV | More commonly in M + Qs; L > K when adjusted for sample sizes; Good negative indicator in water duration > 6 months |
| *Lemna* minor | IV | Subset of above; more overlap; somewhat redundant |
| Pondweeds (=*Potamogeton*) | IV | Useful in long duration wetlands only; may not work in prairies |
| Woolgrass (=*Scirpus cyperinus* | IV | Generally restricted to kettles; not useful in prairies? |
| Rice Cut-grass (=*Leersia oryzoides*) | IV | Highest in impacted kettles and managed prairies; may be weak negative indicator? |
| Bluejoint Grass (=*Calamagrostis canadensis*) | IV | When found, generally in reference kettles; almost always restricted to wetlands of natural condition; good positive indicator |
| Willows (=*Salix*) | IV | Generally uncommon; too infrequent |
| Arrowheads (=*Sagittaria* spp.) | IV | Too uncommon |
| Spikerushes (=*Eleocharis* spp.) | IV | Mostly in impacted prairie wetlands, but also in long duration reference kettles; inconclusive, contradictory? |
| Smartweeds (=*Polygonum* spp.) | IV | Inconclusive; may be a negative indicator in prairies? |
| Horsetials (=*Equisetum* spp.) | IV | Too infrequent |
| Others (sum of those not listed above) | IV | Potential, but inconsistent among water duration classes |
| "Good" taxa sum* | IV | Good performance in kettles; not as well in prairies; good potential |
| Emergents | Percent Cover | Potential, but too much overlap |

| | | |
|---|---|---|
| Submergents | Percent Cover | Potential, but too infrequent in this data set; too much overlap |
| Floating-leafed | Percent Cover | Higher in natural systems; good potential – useful in longer duration only?; better in kettles |
| Open water | Percent Cover | Too much overlap |

* includes all *Carex, Utricularia, Potamogeton, Leersia, Calamogrostis, Sagittaria, Polygonum*, and *Equisetum* species.

Table 10. Assignment of scores for the Wisconsin Wetland Plant Biotic Index.

| **TAXA** | **Attribute** | **Limitations** | **Response** | **Scores** 0 | 1 | 3 | 5 | **Modifications** |
|---|---|---|---|---|---|---|---|---|
| Total Taxa | Count | Taxonomic resolution | Decrease | 0-1 | 2-8 | 9-16 | > 16 | Future (?) |
| Carex | IV | None | Decrease | 0 | < 0.1 | 0.1-0.36 | >0.36 | None |
| Reed Canary Grass | IV | None | Increase | >0.5 | 0.05-0.5 | >0 - 0.05 | 0 | None |
| Cat-tail | IV | None | Increase | >0.25 | 0.03 - 0.25 | > 0 - 0.03 | 0 | None |
| Duck-weed | IV | None | Increase | > 0.6 | 0.2 – 0.6 | > 0 – 0.2 | 0 | None |
| Bluejoint grass | IV | None | Decrease | - | 0 | > 0 – 0.05 | > 0.05 | None |
| Good* | IV | None | Decrease | 0 | > 0 – 0.3 | 0.3 – 0.6 | > 0.6 | None |
| **Deep water Community Adjustment (+ 1 to 5 maximum)** | | | | | | | | |
| **(PondIV + %Floating-leafed)/ 2** | | | | | | | | |
| Pond-weed | IV | > 7 months duration | Optimum | - | 0 | > 0 – 0.12 & > 0.4 | 0.12 – 0.4 | None |
| Floating-leafed | Percent | > 7 months duration | Decrease | - | 0 | > 0 – 0.3 | > 0.3 | None |

# METRIC EVALUATION AND APPLICATION

The interpretation of metric performance is influenced by the composition of wetlands in the data set and their distribution across the landscape. Wetlands were not randomly selected nor were they evenly distributed among classes. Wetlands within a particular "reference" type differed according to history, disturbance factor, water chemistry, or ecoregion. Bogs and "other" wetlands represent totally different classes of wetlands for which a separate scoring system should be developed. Although we present data for these types of wetlands in the report, the indices most likely fail to accurately portray their condition.

We measured the performance of the three wetland indices (WWMBI, 100-CountMBI, and WWPBI) in terms of the response of each composite index and its ability to separate least-disturbed reference wetlands from wetlands suspected to have been impacted. We use the term "suspected" to emphasize the fact that we have no single, pre-determined measure of "human disturbance" which can be used as an objective scale. Watershed condition, historical disturbances, atmospheric inputs, transient disturbances, groundwater contamination, and other such factors individually or in combination influence the biological integrity of an individual wetland. Wetlands categorized as impacted (either kettle or prairie type) may not be adversely impacted by roads, homes, or agricultural land uses occurring in the watershed. Conversely, some of the "reference" wetlands (both kettles and prairies) may have experienced past impacts that were not visible based on our subjective view of the wetland or review of its watershed history. Consequently, in assessing the performance of an index, we did not expect complete separation between reference and impacted wetlands. Some "impacted" wetlands will score better than some reference wetlands, but most reference wetlands should score better than most impacted wetlands if the index is to be of any potential value.
We examined index performance on the basis of response by (1) wetland history, (2) disturbance type, (3) water duration, and (4) prairie versus kettle. We also compared how the three indices related to the subjective habitat index and other physical and chemical parameters.

## PERFORMANCE OF THE WISCONSIN WETLAND MACROINVERTEBRATE BIOTIC INDEX:

The WWMBI performed well in separating impacted prairie wetlands from reference wetlands (Figure 12). The majority of impacted kettles also scored less than the median reference kettle. The performance of the WWMBI did not decline substantially when restricted to short and medium duration wetlands only (Figure 13). The impact of water duration on the index does appear of some consequence, however. Within wetland types, the WWMBI increases with increasing water duration in reference kettles and decreases in reference prairies (Figure 14). The reasons for this are not clear, but simply may reflect the inclusion of a few slightly impacted wetlands at either end of the spectrum in the study group.

The distribution of WWMBI scores by history classification reveals a great amount of spread within natural wetlands (Figure 15). The natural wetlands include all reference wetlands (both kettle and prairie), bogs, a few managed prairies, and several moderately-impacted wetlands (but excluding restored wetlands). The wetlands categorized as "disturbed" (N=8) represent those wetlands known (or highly suspected prior to this analysis) to be impacted by agricultural or other impacts. Using the median WWMBI score for reference kettles as a goal, the best wetlands within each of the impacted wetland classes are generally those wetlands of natural origin (Figure16). Restored wetlands score less favorably than do created (albeit few in number) or disturbed wetlands. Differences in water duration may force modification of scores as demonstrated by plots shown in Figure 17. While the pattern in WWMBI scores between reference and impacted wetlands is fairly similar among the three water duration classes, the scores for reference kettles in longer duration wetlands are generally lower than in short or medium duration wetlands. A plot of just the reference kettle data illustrates the decline in WWMBI scores within long duration wetlands (Figure18). A larger data base will be essential before refining the system to account for these trends.

A comparison of WWMBI scores within each wetland class by type of disturbance suggests that pasture (N=1), roads, agriculture (current), and excavation have the greatest impact on macroinvertebrate communities (Figure 19). Old agriculture and houses have less impact, and in one of the two watersheds dominated by urban land use, this actually appeared to have a beneficial impact. The latter conclusion may be an artifact caused in part by the fact that both urban wetlands were short duration and many short duration wetlands had WWMBI scores above the median score of 43 (see Figure 20). This again stresses the importance of making valid comparisons using the appropriate matched set of wetlands. Thus we compared the urban wetlands with the short duration reference kettles. The result was inconclusive as four or five of the reference kettles scored higher than the two impacted kettles with urban influenced watersheds, and two or three reference kettles scored lower than the impacted kettles. The data are just too few to draw any valid conclusions.

We plotted the response of the WWMBI to selected environmental attributes to examine the influence of those attributes on the index (Figure 21). Size (surface acreage) had little influence on index scores. Index scores tended to decline slightly in wetlands experiencing high pH and/or high alkalinity. The decline is logical in that high pH and alkalinity generally accompany increased primary productivity associated with algae blooms and/or macrophyte production. No clear response was noted with conductivity, although one might expect a similar response to pH and alkalinity. Shade canopy cover had little influence on scores despite the probable influence on temperature regimes and selectivity among some taxa for oviposition sites or breeding sites based on shade cover (references). The percent of riparian zone composed of woodland exhibited a slight positive impact on WWMBI scores. This may be result from the increased allocthonous inputs of leaves contributing to an increased population of shedders, including Limnephilid caddisflies.

We proposed the subjective Macroinvertebrate Habitat Index (MHI) not as alternative measure of human disturbance but rather as a predictive measure that was intended to account for both human and natural factors that influenced macroinvertebrate community condition. As expected, MHI scores corresponded quite well with expected conditions among prairie (best discrimination) and kettle wetlands (Figure 22). Scores among the limiting habitat index exhibited too much overlap among wetland types to be of any practical value. To determine how each component of the habitat index influenced the macroinvertebrate community, we plotted WWMBI scores with individual component scores of the subjective MHI (Figure 23). In theory, the WWMBI was expected to respond positively to all habitat metrics. In reality, they responded poorly. The land use metric and watershed metric appeared to respond as expected, but both displayed more variability than anticipated. WWMBI scores were only weakly correlated with the trophic, colonization, and water clarity metrics. The natural metric appeared to show a slight negative relationship with WWMBI scores (Figures 23 & 24). The combined metrics form the MHI, which was a poor predictor of WWMBI scores (Figure 25). The center of the three parallel lines in the figure represents the 1:1 line and the two outside lines represent a reasonable error of ± 10. Many wetland scores were lower than predicted, while a few had higher scores than expected. Wetland history did not explain the lack of correspondence between the two indices (Figure 26); natural wetlands exhibited the same amount of variability as did created, disturbed, and restored wetlands. The poor correspondence between the two indices may result from the incorporation of the wrong habitat metrics, inaccurate scoring of individual habitat metrics, not including other important habitat factors (e.g., herbicide – pesticide impacts), or simply because the WWMBI does not relate well to average habitat conditions.

The WWMBI also did not relate well to limiting resource conditions as measured by the lowest individual habitat metric scores in each wetland (Figure 27). Some of the highest WWMBI scores corresponded to the lowest habitat scores, which suggests that some habitat metrics may not be influencing the macroinvertebrate community in the fashion we assume to be true. A cluster of high WWMBI scores (> 50, upper left of Figure 28) represented natural wetlands which scored low on the "natural" habitat metric. This, together with the fact that this metric is negatively associated with WWMBI scores, suggests that our assumptions regarding the negative impact of short water duration and predators in wetlands may have been wrong. Adjustments to the MHI are needed, and more extensive examination of the data may provide insight as to how to make those adjustments.

The WWMBI is a multimetric index and its performance is dependent upon the individual metrics upon which it is based. Individual metrics may respond differently depending upon the type and degree of disturbance or impact. Non-insect metrics, specifically those representing mollusks, worms, and fairy shrimp, responded differently across the various wetland classes (Figure 29). Mollusks scored highest in reference prairies and kettles. Worms were only slightly higher in reference kettles than in impacted kettles, but were clearly higher in reference prairies than counterpart prairie types. Fairy shrimp scores as shown in the figure are deceiving due to the fact that long duration wetlands (which normally do not contain fairy shrimp and consequently receive an automatic score of 5) are included. Some consideration should be given to excluding (or including) metrics as necessary to adjust for differences in water duration, thus accounting for the natural differences that exist in macroinvertebrate communities among the different classes. Average scores of the individual metrics provide some indication as to how well (or poorly) the various macroinvertebrate community attributes respond to various disturbances or wetland history (Figure 30). Mollusks appear to do quite well in created wetlands, but lag behind reference wetlands in restored and disturbed wetlands. Worms are impacted adversely by all forms of human interactions. Again, the adjustment of fairy shrimp scores for long duration wetlands appears to be a mistake in that it raises the average scores for long duration impacted wetlands. The non-insect taxa metric appears to be useful only among prairie wetlands; no separation in average scores among kettle wetlands was observed (Figure 31). Damselfly, pigmy backswimmer, and water boatmen metrics function differently among kettle and prairie wetlands. All three metrics appear to discriminate between reference and impacted condition in kettle type wetlands (Figure 32). Pigmy backswimmer scores are highest in reference prairie wetlands, but the other two metrics do not appear to work well in prairie wetlands. Water boatmen scores were lowest in reference prairies, and damselflies were highest in impacted prairies. The caddisfly metrics performed best among individual metrics. Caddisfly abundance, limnephilid abundance, and percent total caddisflies clearly and consistently separated reference wetlands from impacted wetlands (Figure 33). The Diptera metrics also appeared to function consistently (Figure 34). Phantom midge scores were highest in reference wetlands, with particularly good separation among prairie wetland types. The mosquito metric differed between kettles and prairies, but still discriminated between unimpacted and impacted systems within respective types. The mosquito metric may require some adjustment of scores for longer duration wetlands where large numbers of mosquitoes may indicate unfavorable conditions for other taxa. The soldier fly metric (a negative metric) appears of marginal use and may require some form of adjustment. The total abundance metric performed better than the total taxa metric in separating impacted and reference wetlands (Figure 35). The total taxa metric appears to work better among prairies than between kettle types.

The multimetric WWMBI scores show a clear separation among wetland types (Figure 36). Reference kettles and reference prairies had substantially higher average scores than all other types. The median score for reference kettles (43) represents a goal or standard for comparison with other wetlands. We set threshold lines for rating biotic integrity of wetlands based on the $10^{th}$, $25^{th}$, $50^{th}$, $75^{th}$, and $90^{th}$ percentiles among reference kettles (Figure 37). The classification of all kettle and prairie wetlands using this rating system demonstrates that most restored, created, and disturbed wetlands do not compare with natural least-impacted systems (Figure 38). All "excellent" and "very good" wetlands were classed as natural; 82% of the wetlands rated as "poor" or "very poor" were impacted or otherwise managed by man. Only 6% of the impacted prairie wetlands received a "good" rating, while 39% of the impacted kettles were in the "good" range. Two-thirds of the restored wetlands received a rating of "poor" or "very poor". Conversely, 4 of the 5 created wetlands rated as "fair". Consequently, the WWMBI appears to fairly accurately discriminate between wetlands seriously disturbed by human activities and those only minimally disturbed. Additional testing and evaluation against an independent data set will be required to more fully assess the utility of the WWMBI.

The WWMBI consists of the sum of 15 individual metric scores (Figure 39). Different types of wetlands score differently according to what habitat or environmental conditions are present and what macroinvertebrate communities develop in each. While the WWMBI can be calculated for other wetland types (e.g., bogs, fens, etc.), we do not recommend such an application because other wetland types have distinctly different macroinvertebrate communities. They naturally may score either higher or lower than depression wetlands, but the difference in scores may be meaningless. Further work will be required to develop independent metrics and indices for other wetland types. Comparisons of individual metric scores within a particular group of wetlands may provide insight into what factors are responsible for the differences in total WWMBI scores (Figure 40). In the case of impacted kettles, it appears that the major difference with least-disturbed kettles is in lower caddisfly (all three) and damselfly metrics. This may signify a decrease in allochthonous inputs of coarse particulate matter (i.e., leaves) upon which the caddisflies depend. The slight decrease in damselflies may be insignificant. The relative breakdown of metric scores in bogs is shown to illustrate the previous point that the communities differ considerably from that in kettles despite their similar setting.

---

**<u>PERFORMANCE OF THE WISCONSIN WETLAND 100-COUNT MACROINVERTEBRATE INDEX</u> (rapid assessment)**

The 100-count Index does a fairly adequate job of separating impacted prairie wetlands from reference wetlands (Figure 41-right). It does not perform quite as well in kettles. Natural wetlands scored slightly higher than restored, disturbed, and created wetlands (Figure 41-left). Within impacted wetlands, wetlands of natural origin generally scored higher than counterpart created, disturbed, and restored wetlands (Figure 42). Agriculture, excavation, roads, and pasturing appear to have about equal degrees of impact on the 100-Count MBI within impacted prairies (Figure 43). Within impacted kettles, roads appeared to have the greatest impact on the100-Count MBI (Figure 44).

The performance characteristics of the nine individual metrics that comprise the 100-Count MBI illustrate basic differences between kettle and prairie wetlands (Figures 45 & 46). The EOT metric works quite well in prairies but not so well in kettles. Likewise the same was true for the number of non-insect taxa in the100-count MBI. Pigmy backswimmers did not exhibit much difference among prairie types, but showed some promise within kettles. Water boatmen and all bugs combined appeared to function better in prairies. Limnephilids and total caddisflies showed fairly good separation within prairies and marginal separation in kettles. Midges exhibited marginal promise as a negative indicator in both prairies and kettles. Soldier flies appeared to be of some value in prairies but not in kettles.

We developed a rating system for wetland biotic integrity based on the 33 reference kettle scores (Figure 47). Some future adjustment may be required for longer duration wetlands to compensate for the apparent decline in scores in semipermanent and permanent wetlands. The distribution of all 100-Count MBI scores by wetland type is provided in Figure 48. Reference kettle wetlands were used to develop the rating system so by definition half of the wetlands are good, very good, or excellent and the remainder are fair, poor, or very poor. All reference prairie wetlands were rated as either good or excellent (Figure 49). Only five (28%) of the impacted kettles received ratings of good or very good, and only eight (30%) of the impacted prairie wetlands achieved such status. Conversely, fifty-six percent of the impacted prairies received a rating of very poor. Twenty-seven percent of the managed prairie wetlands received ratings of good or excellent.

The 100-Count MBI ratings were influenced by wetland history (Figures 50 & 51). The majority (70%) of wetlands receiving favorable ratings of good, very good, and excellent were in natural condition. The majority of created, heavily disturbed, and restored wetlands received fair or lower ratings. Restored wetlands fared worst, with over half rated as very poor. The fact that many of the longer duration, natural wetlands were categorized as very poor suggests that the index may not perform well for long duration wetlands and that some adjustment in the index may be necessary to compensate for this apparent bias.

The 100-Count MBI exhibited approximately the same amount of lack of correspondence with the subjective habitat index scores as did the WWMBI (Figure 52). Roughly 8% and 17% of the 100-Count MBI scores were better than or less than, respectively, corresponding habitat index scores (± 10 considered reasonable). Most outliers faring better than expected were impacted kettles and prairies, while many of the wetlands that did not match expectations were bogs, or reference kettles (Figure 53). The occurrence of several reference kettles as outliers suggests that our perceptions of what constitutes ideal habitat for macroinvertebrates among longer duration kettles may be in error. Modification of the subjective habitat index appears warranted.

The 100-Count MBI ratings agree within ± 1 classification of WWMBI ratings 72% of the time (Figure 54). Ratings differ by two classes in about equal numbers (high or low), suggesting no consistent bias exists between the two indices. Currently, there is no means of determining which index more accurately measures biotic integrity. Because the two indices are based on a different set of community attributes, it may be that lack of correspondence between the two is natural and the degree to which they differ may be indicative of the type of disturbance to the wetland.

## WISCONSIN WETLAND PLANT BIOTIC INDEX PERFORMANCE

The WWPBI performed well in separating most wetland types (Figure 55). Kettles scored consistently higher than prairies suggesting perhaps that the index should not be used to make comparisons between groups or that different expectations should be established for the two groups. Within kettle wetlands, created, restored (only one), and disturbed wetlands consistently scored among the lower 25% of the natural kettles (Figure 56). The two outliers among natural kettles represent episodic, rain-filled depressions that probably should not be classed as wetlands at all. Among prairie wetlands, WWPBI scores in created wetlands and a substantial number of restored wetlands equaled or exceeded that found in the natural wetlands (Figure 57). The WWPBI did not discriminate very well among wetland history and disturbance factors (Figures 58 - 61). Surprisingly, within restored prairie wetlands, excavated wetlands consistently scored higher than wetlands impacted by agriculture or roads. Even two of the three created prairie wetlands (all excavated) were on par with best of the impacted prairie wetland group. Road-impacted kettles were among the lowest scores observed.

We established separate rating systems for kettles and prairies to account for the apparent differences in the two wetland classes. For kettles, we based the ratings on the 10th, 25th, 50th, 75th, and 90$^{th}$ percentiles of least-disturbed reference kettles (Figure 62). We excluded two outliers that represented episodic depressions containing only wood-nettle (*Laportea canadensis*). Application of the WWPBI to impacted kettles showed that 67% were rated as poor or very poor (Figure 63). Only two impacted kettles rated good or better. We used a different approach in establishing standards for prairie wetlands. Because there were so few reference prairie wetlands, we set the threshold between good and fair based on the lowest score among the reference prairies (Figure 64). Other standards were based on the 10$^{th}$, 25$^{th}$, 75$^{th}$, and 90$^{th}$ percentiles of the combined prairie data. Using this rating system, four of the five reference prairies were good; the other was excellent. Five of the eleven managed prairies rated as fair to very poor, and 48% of the impacted prairies rated lower than the lowest reference prairie wetland (Figure 64).

## <u>RELATED ISSUES</u>:
**Temporal variability, replication, and screen versus dip-net samples**

Macroinvertebrate collections for the WWMBI were conducted during the early spring sampling period, April to mid-May, in order to ensure that standing water was present to support an aquatic macroinvertebrate community. Short duration wetlands may be dry by the end of May. Likewise, sampling during early spring minimizes the impact of colonization by winged insects (i.e., immigration). Larger, more mature specimens, which have likely over-wintered as residents in the wetland have not yet emerged and many adult winged aquatic insects have not yet emigrated. Macroinvertebrate communities are expected to change both in terms of abundance and taxonomic composition as seasons change. It was not certain what impact this would have on WWMBI scores collected at times other than early spring. Consequently, we sampled eight wetlands on a monthly basis from April through early October to examine the temporal variability in index scores. These data were used to determine the feasibility of applying the index to samples collected throughout the late spring and summer.

Temporal changes in the WWMBI were substantial among wetlands (Figures 65 & 66). Scores declined dramatically in some wetlands, dropping from excellent to poor in the period of two months in French Creek – West, and remaining fairly constant in others (see Old World Wisconsin). The magnitude of temporal change appeared to be related to water duration class; the changes within smaller, shorter duration, wetlands were more sudden. These data suggest that the WWMBI is very sensitive to sample collection timing, which also means that it may be sensitive to annual climatic differences from one spring to another. More studies are required to determine the extent that this sensitivity influences the WWMBI within individual wetlands. It may be possible to establish seasonal correction factors for the WWMBI based on degree-day accumulations similar to that available for the arthropod biotic index for streams (Hilsenhoff 1988). Currently, the **WWMBI does not appear to be stable** across dates. Consequently, use of the WWMBI must be restricted to the **early spring** sampling period (which may vary depending upon locale).

We collected two field replicates from nine wetlands during the April sampling period and three replicates from four wetlands during August or October to estimate the degree of variability in WWMBI scores associated with our field collection methodology and laboratory processing. Variation in WWMBI scores among replicates collected in August and October was very low (Figures 65 & 66), with most scores falling within the same ratings classification. Variation in April WWMBI scores was high in two-thirds of the wetlands sampled (Figure 66; L-17 and Figures 67 & 68). The change in classification between the two replicates usually only covered adjacent classes, but in some cases (e.g., B10 and W07) the absolute difference in scores was large. Currently, we can not separate the cause of this variation between field collections and laboratory processing. Additional studies will be required using more field replicates and examining laboratory issues to pinpoint sources of variation in scores.

The collection of screened macroinvertebrate samples (Helgen & Gernes 1996) represents a substantial cost savings in terms of laboratory processing over the large investment required to process the typical D-frame kick-net samples. Consequently, we tested this method using samples collected from 15 wetlands during April 1998. Unfortunately, most samples contained too few specimens to compute a WWMBI score. Screen samples captured, on the average, five fewer taxa per wetland than net samples. Almost all WWMBI scores calculated using screen sample data rated as very poor (Figure 69). Because of the differences in abundance between the two sampling methodologies, the rating scores for the screen samples should be tailored to compensate for the smaller specimen numbers. We can not do this with only 15 samples. However, the similarity in patterns expressed by the relative position of screen scores and net scores in Figure 69 suggest that the screen sample method may provide satisfactory results. High screen scores are generally associated with high net scores. Consequently, we recommend continuing studies incorporating the screen sampling procedure as an alternative to dip nets.

# MANAGEMENT CONSIDERATIONS & CONCLUSIONS

The three multimetric biological indices developed from this study demonstrate good potential for application in classifying and ranking kettle and prairie depression wetlands as to their relative biological integrity when required. The indices can be applied immediately in assessing the success of wetland restoration efforts in Wisconsin and in establishing goals or standards tailored for specific types and classes of wetlands. The sample data used in computing the indices can be further utilized in defining the biological purpose of individual wetlands or how they compare in terms of biological function with wetlands of similar class and regional distribution. Rarity is an issue that can not be addressing using the limited data base available in this study; construction of a statewide data base encompassing wetlands representing all ecoregions and wetland types will be required. This study has only begun to identify the hydrological and ecological conditions necessary to support a fully-functioning, aquatic ecosystem.

Limitations: The WWMBI is designed as a laboratory tool and, as such, has limitations including relatively slow turn around time and a substantial investment of labor (4-8 hours per sample). The 100-Count MBI has yet to be applied in the field, but is intended to provide rapid assessment (< 1 hour) of wetlands. Both macroinvertebrate indices are based on the macroinvertebrate community present during the early spring sampling period (April-May), which further limits their application. The impact of introduced predators, unseen interconnections with permanent water (e.g., springs and crayfish burrows), and the inherent dynamic variation in water duration associated with temporary wetlands all function to create a considerable amount of "noise" or scatter in the macroinvertebrate data. The WWPBI is most likely less affected by these factors and thus may serve better as a long-term integrator in assessing biotic integrity. This does not negate or rule out the utility of the macroinvertebrate indices as they may respond more quickly to impacts and thus serve as better ecological indicators or sentries. Expansion of our work to include other biological components, including amphibians, zooplankton, phytoplankton (diatoms), and small mammals will enhance our ability to formulate a fully-functioning, multimetric, index of ecological integrity. Screen sampling may be an alternative to dip-netting if index scores can be tailored to adjust for differences in sample sizes and community composition.

# ACKNOWLEDGMENTS

This study was funded in part by a USEPA Wetland Grant #CD985491-01-0, administered by the USEPA Region V office, and the Wisconsin Department of Natural Resources. We especially thank Richard Bautz, John Wagner, and Bob Wernerehl, for their many efforts in conducting the field and laboratory portions of this study. We also thank C. Garra (USEPA), P. Trochlell, T. Bernthal, S. Hausmann, D. Siebert, and A. Sabor for review comments, and S. Hausmann, D. Gebken, J. Addis, K. Beetham, R. Halverson, and C. Garra for project administration and guidance. Ted Cochrane, UW-Madison Herbarium, kindly verified the identifications of several plant voucher specimens. A special thanks goes to those individuals who helped us in finding wetlands to sample or who assisted with other planning phases of this project. This includes the following (affiliations other than WDNR are given in parenthesis): D. Reed (SEWRPC), D. Read (US Army), T. Grunewald, D. Simon, D. McConnell (USFWS), T. Neigus, N. Braker (TNC), L. Johnson (NRRI, U-Minn.), T. Meyer, A. Larsen, R. Hoffman, E. Epstein, M. Martin, G. Barrett (WP&L), T. Miller, R. Hay, R. Beilfuss (ICF), V. Pappas, J. Kline, J. Jackson (DOT), R. Strand, M. Sparrow, J. Pritzel, R. Karowski, K. Leithoff (DOT), J. Ruwaldt (USFWS), C. Richardson, (Mequon), B. Weihrouch (NRCS), T. Thrall (NRCS), K. Schreiber, D. Boorse (UW-Madison), A. Wilson, P. Rossa (Madison), E. Lobner, P. Garrison, and R. Arneson.

# LITERATURE CITED

Daubenmire, R. 1959. A canopy-coverage method of vegetational analysis. Northwest Science 33(1):43-64.

Helgen, J., and M. C. Gernes. 1996. Wetlands index of biotic integrity: development of invertebrate and vegetation-based indices in degraded and reference wetlands. Abstract for USEPA sponsored conference "Wetlands: Biological Assessment Methods and Criteria Development Workshop" held September 18-20, 1996 at Boulder, Colorado.

Hilsenhoff, W. L. 1982. Using a biotic index to evaluate water quality in streams. Wisconsin Department of Natural Resources Technical Bulletin No. 132. 22 pp.

Hilsenhoff, W. L. 1988. Seasonal correction factors for the biotic index. Great Lakes Entomologist 21(1):9-13.

Lillie, R. A., and P. J. Garrison. In Prep. Using canonical correspondence analysis of macroinvertebrate and diatom community composition to differentiate between lakes and wetlands.

Omernik, J. O. 1987. Ecoregions of the conterminous United States – map supplement. Annuals of the Association of American Geographers 77(1):118-125.

Schneider, D. W., and T. M. Frost. 1996. Habitat duration and community structure in temporary ponds. J. N. Am. Benthol. Soc. 15(1):64-86.

State Laboratory of Hygiene. 199?. Manual of standard operating procedures inorganic chemistry unit. Method 115.1, automated alkalinity, pH, and conductivity.

SPSS. 1997. SYSTAT 7.0 Graphics. SPSS, Inc., Chicago, IL 60611. 450 pp.

Trochell, P., and T. Bernthal. 1998. Small wetlands and the cumulative impacts of small wetland losses: A synopsis of the literature. Wisconsin Department of Natural Resources Publ-FH226-98, P.O. Box 7921, Madison, WI 53707. 16 pp.

Williams, D. D. 1987. The ecology of temporary waters. Pub. Timber Press, Portland, OR. 205 pp.

# FIGURE CAPTIONS

**FIGURE 3. Frequency of occurrence of dominant plant cover.**

**FIGURE 4. Frequency of occurrence of taxa representing second most important plant cover.**

**FIGURE 5. Frequency of occurrence of taxa representing third most important plant cover.**

**FIGURE 6. Frequency of occurrence of dominant macroinvertebrates within three classes of prairie wetlands.**

**FIGURE 7. Frequency of occurrence of dominant macroinvertebrates in kettle wetlands.**

**FIGURE 8. Frequency of occurrence of dominant macroinvertebrates in bogs and "other" wetlands.**

**FIGURE 9. Frequency of occurrence of dominant macroinvertebrates by wetland history classification.**

**FIGURE 10. Frequency of occurrence of dominant macroinvertebrates by water duration classification.**

**FIGURE 11. Frequency of occurrence of dominant macroinvertebrates by ecoregion.**

**FIGURE 12. Distribution of Wisconsin Wetland Macroinvertebrate Biotic Index (WWMBI) scores by wetland reference classification. Box plot data represent median, first and third quantiles, and extremes.**

**FIGURE 13. As in figure 12 except restricted to medium and short duration wetlands only.**

**FIGURE 14. Distribution of WWMBI scores by wetland type and duration class.**

**FIGURE 15. Distribution of WWMBI scores by wetland history classification.**

**FIGURE 16. Distribution of WWMBI scores by wetland type and history. Horizontal line represents median for reference kettles.**

**FIGURE 17. Distribution of WWMBI scores by wetland type, history, and duration classification.**

**FIGURE 66. Temporal changes in WWMBI scores in four south-central Wisconsin wetlands. Field replicates numbered 1-3; screen samples designated as "SC". Field replicates circled.**

**FIGURE 67. Field replicate WWMBI scores for four southeastern Wisconsin wetlands. April 1988 samples.**

**FIGURE 68. Field replicate WWMBI scores for four northern Wisconsin wetlands. April 1988 samples.**

**FIGURE 69. Comparison of WWMBI scores between screen samples (=SC1) and composited kick-net samples (=1 or 2) in 15 south-central Wisconsin wetlands during April 1998.**