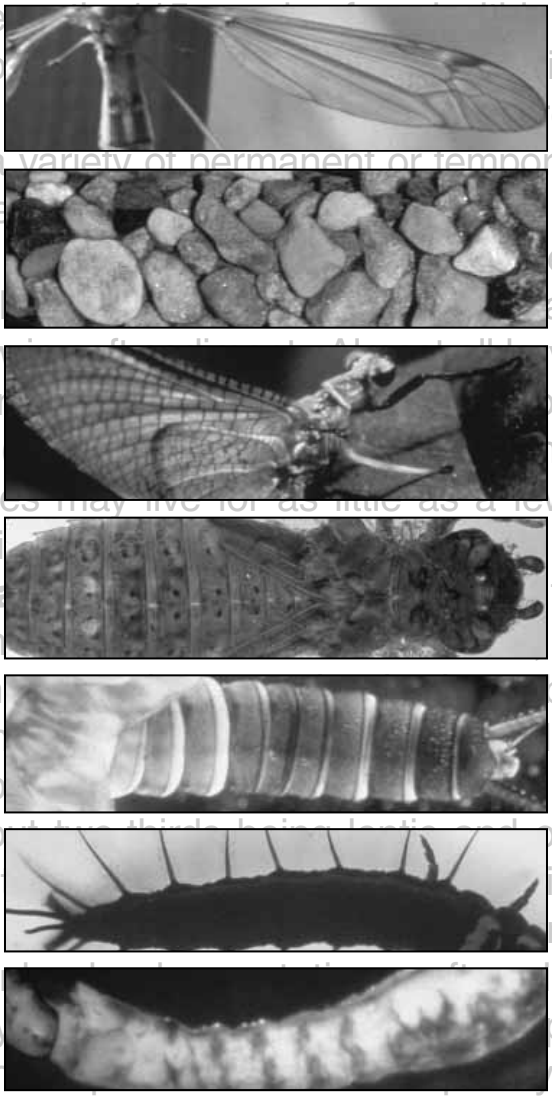

MACROINVERTEBRATE

Data Interpretation Guidance Manual

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Madison, WI
2003

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Contents

Overview 2

Introduction 3

Metrics Generated by the BUGPROGRAM 5

Interpretation of Metrics 5

Descriptive Metrics 6
- Biotic Indices 6
- Richness Measures 9
- Diversity Measures 14
- Functional Feeding Classes 16
- Ratio Measures 18
- Dominance Measures 19

Comparison Metrics 19
- Paired Comparison Metrics 20
- Ratio Metric Comparisons 20
- Presence/Absence and Percentage Comparison Metrics 20

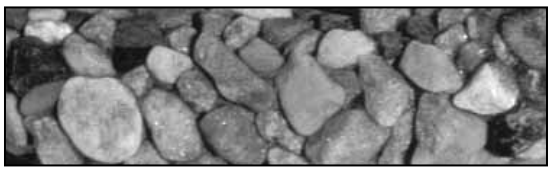
Recommendations 23
- Objectives 23
- Microhabitat Considerations 25
- Choosing the Appropriate Metrics 25

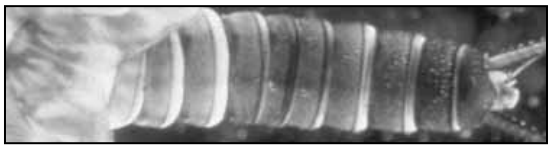
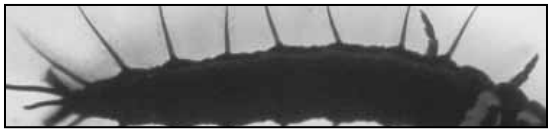
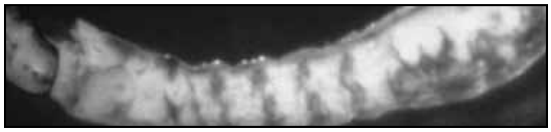
Future Directions 28

References 31

Appendix A. Selected Literature Review 47

Appendix B. A History of the Development of Macroinvertebrate Biological Indicies in Wisconsin 57



Overview

This guidance manual is designed to assist Departmental staff interpret macroinvertebrate data reported to the Department through contractual arrangements with the University of Wisconsin Stevens Point (UWSP). Data are analyzed, and the computer software program **BUGPROGRAM** [1] version 6.01 reports results.

The electronic database as of November 2003 had 8,940 individual samples (11,970 samples when including replicate samples) and includes data for all macroinvertebrate samples processed by the UWSP from 1983 to the present. Approximately 90 percent of the samples in the database have associated latitude and longitude data. The database includes macroinvertebrate samples collected by: Wisconsin Department of Natural Resources (DNR), United States Geological Survey (USGS), and United States Forest Service (USFS), University, and private research projects.

By querying the electronic database, the BUGPROGRAM produces reports consisting of a series of over 30 biological community attributes for each sample. These results include measures of richness, diversity, dominance, functional-feeding classes, and biotic indices. This guidance manual is intended to: 1) provide a detailed description of each biological attribute (commonly referred to as a metric), 2) provide a simplified explanation of the biological significance of each metric, and 3) instructions as to how and when each metric should be applied. A comprehensive list of references on biological indices is provided at the end of this document for those who wish to explore these issues in greater depth.

Please note, this document does not tell you how, when, or where to sample (please see DNR Field Procedures Manual at **http://intranet.dnr.state.wi.us/int/es/science/ls/fpm/table.htm**). It does not tell you how to conduct statistical procedures or suggest which procedure is best to use under given circumstances, nor does it specifically address other macroinvertebrate biomonitoring issues such as lake, wetland, or contaminated sediment assessments. Order descriptions of insects, presented in this document as side-bars, are summarized from Hilsenhoff (1995).

[1] The BUGPROGRAM, program manual, installation instructions, and historical databases are currently available using the following URL:**http://www.uwsp.edu/water/biomonitoring/BUGPRO.HTM**. Program structure and capabilities of the BUGPROGRAM 6.01 are described in further detail in the program manual online. Biomonitoring data are available at **http://www.uwsp.edu/water/biomonitoring/index3.htm**.

Introduction

The DNR collects macroinvertebrate samples from hundreds of sites annually for the purpose of assessing stream water quality. Most DNR macroinvertebrate samples are processed under contract by UWSP. The data are used for watershed appraisals, basin assessments, stream classification, pre/post assessments of point sources, and various water quality assessments, and trends and research analysis.

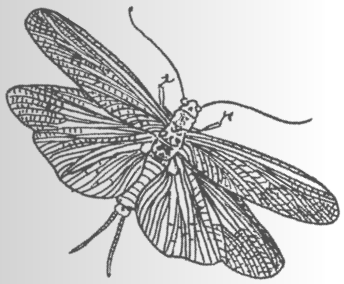
Although the data reports generated by the BUGPROGRAM include numerous community and trophic *metrics*, the DNR has almost exclusively used the Hilsenhoff Biotic Index (HBI) values in interpreting water quality. Other metrics may provide useful information but, because of a combination of uncertainty and unfamiliarity regarding interpretation, these metrics have not been fully utilized. *The term* ***metrics*** *as used in this report refers to any one of a number of various measurements or values that represent simple attributes of the macroinvertebrate community from the total number of different taxa found in a sample or percentages of certain taxa relative to the total number of organisms in a sample, to more complex measures based on tolerance values such as the Hilsenhoff Biotic Index (Hilsenhoff 1987).*

This guidance manual is designed to assist Department staff understand the biological significance of the metrics reported by the BUGPROGRAM and extract additional information from the macroinvertebrate data. The strengths and weaknesses of each metric are outlined, and the problems relating to interpretations of each metric are highlighted when known. The relative sensitivities of each metric to various environmental stresses or pollutants are described where known. Quality assurance and quality control issues are also outlined.

This manual is intended to serve as a primer on the subjects discussed. A list of references addressing macroinvertebrate community structure and analysis, biocriteria, bioassessment, and related issues are provided in Appendix A. This reference list includes a category of general review articles and several excellent books on subjects related to water quality monitoring and assessment.

A short history of the development and use of macroinvertebrate indices in Wisconsin is included (Appendix B) to provide a perspective of its evolution. A complete taxonomic listing of all aquatic insects known to occur in Wisconsin with corresponding life history traits and distribution information on each taxon is currently under development. It is intended that once completed the listing will be made available to Department staff electronically in the form of an intranet atlas compatible with ArcView and other GIS applications.

Stonefly adult.



Ten orders of insects found in Wisconsin have species with aquatic lifestages. Five (**Ephemeroptera, Odonata, Plecoptera, Megaloptera, and Trichoptera**) are aquatic orders in which almost all species have aquatic larvae. The remaining orders (**Heteroptera, Coleoptera, Diptera, Lepidoptera,** and **Neuroptera**) are partially aquatic orders in which most species are terrestrial, but in which certain families, genera, or one or more species have a lifestage adapted to the aquatic environment. The insect orders Collembola, Orthoptera, and Hymenoptera are primarily terrestrial orders that have some semi-aquatic species. However, these are not used in biomonitoring studies.

Table 1. *Individual sample metrics generated by the BUGPROGRAM. n.a. = not applicable*

Metric	Name or Description	Taxonomic Level	Range	Reference
Biotic Indices				
HBI	Hilsenhoff Biotic Index	Mixed (genus and species)	0-10	Hilsenhoff 1987
FBI	Family-Level Biotic Index	Families	0-10	Hilsenhoff 1988a
MTV	Mean Tolerance Value	Mixed (genus and species)	0-10	Lillie and Schlesser 1994
Max *x*	Max-*x* HBI	Mixed (genus and species)	0-10	Hilsenhoff 1998
Taxa Richness				
SR	Total Species Richness	Mixed (genus and species)[a]	0-∞	Plafkin et al. 1989
GR	Generic Richness	Genus	0-∞	Plafkin et al. 1989
EPTG[b]	Ephemeroptera-Plecoptera-Trichoptera Generic Richness	Genus	0-∞	Plafkin et al. 1989
Diversity				
d	Margalef's Diversity Index[c]	Genus	0-?	Margalef 1958
H'	Shannon's Index of Diversity[d]	Genus	0-?	Magurran 1988
Trophic Function				
Percent Scrapers	Percent of total represented by Scrapers	Individuals and genus [e]	0-100	Cummins and Merritt 1996
Percent Filterers	Percent of total represented by Filterers	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Shredders	Percent of total represented by Shredders	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Gatherers	Percent of total represented by Gatherers	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Collectors	Percent of total represented by Collectors	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Scrapers/ percent Filterers	Ratio percent Scrapers to percent Filterers	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Scrapers/ percent Gatherers	Ratio percent Scrapers to percent Gatherers	Individuals and genus	0-100	Cummins and Merritt 1996
Percent Scrapers/ percent Collectors	Ratio percent Scrapers to percent Collectors	Individuals and genus	0-100	Cummins and Merritt 1996
Dominance				
Dominant families	Percent of total count represented by top five families	Family	n.a.	Plafkin et al. 1989
Dominant genera	Percent of total count represented by top five genera	Genus	n.a.	Plafkin et al. 1989
Dominant species	Percent of total count represented by top five species	Species[f]	n.a.	Plafkin et al. 1989

[a] Note: Occasionally higher taxonomic levels used.

[b] This metric is also reported individually for each order (Ephemeroptera generic richness, Plecoptera generic richness, and Trichoptera generic richness) and also reports the ratio of numbers of individuals of EPT to total number of individuals of all taxa in a sample.

[c] Although not reported in BUGPROGRAM 6.01, Margalef's Diversity Index was reported in the first version of the BUGPROGRAM and it was thought that future comparative studies might evolve.

[d] This metric is reported in version 6.01 of BUGPROGRAM.

[e] This taxonomic level is computed separately on the basis of total individuals in sample and total genera in sample.

[f] Note: BUGPROGRAM may list as 'unidentified' because some taxonomic identification is only to genus.

Table 2. *Paired community comparison metrics available through the BUGPROGRAM.*

Metric	Name	Taxonomic Level	Range	Reference
CCL	Coefficient of Community Loss	Lowest taxa	0-∞	Courtemanch and Davies 1987
CS	Coefficient of Similarity	Genus	0-1	Pinkham and Pearson 1976
SIMI	Stander's Similarity Index	Lowest taxa	0-1	Stander 1970
PS	Percentage Similarity	Genus	0-1	Whittaker 1952, Rabeni and Gibbs 1980
B	Coefficient of Similarity	Genus	0-1	Pinkham and Pearson 1976
EDIS	Ecological Distance	Genus and species	0-1	Clark 1952, Rhodes et al. 1969

Metrics Generated by the BUGPROGRAM

Currently for each macroinvertebrate sample, version 6.01 of the BUGPROGRAM generates several measures of taxonomic richness, one diversity index, four biotic index values, eight trophic function ratio metrics at two taxonomic levels, and three sets of dominant taxa lists (Table 1). In addition, the BUGPROGRAM provides five metrics to compare macroinvertebrate communities between pairs of selected samples (Table 2).

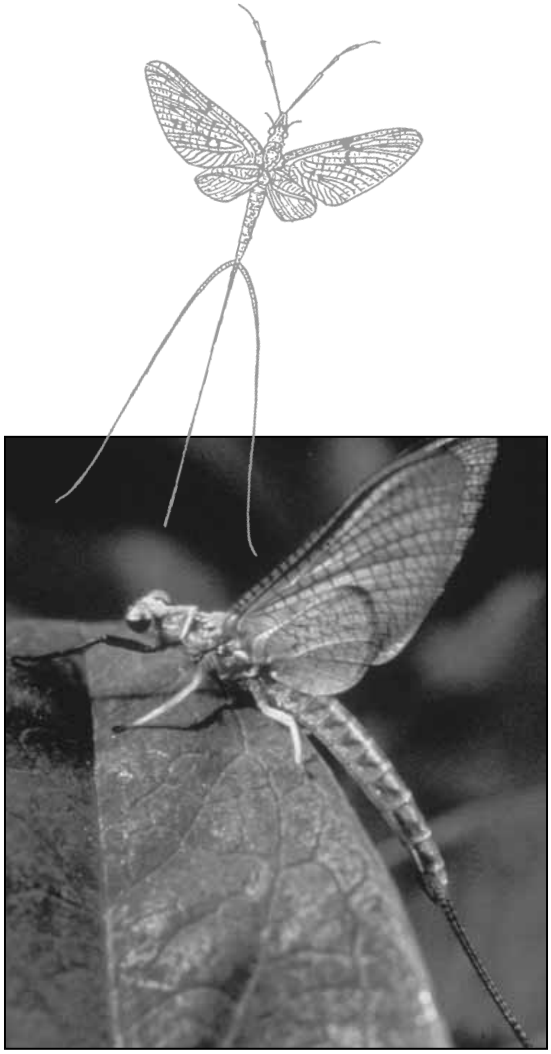
In addition to the previously named metrics (Tables 1 and 2), the BUGPROGRAM reports counts of individuals, percentages of individuals, or percent specific taxa present in the sample or used in computing the biotic indices. The various counts are used in computing some individual sample metrics and counts are useful in evaluating the validity of a particular sample. UWSP standard procedure is to subsample until 125 organisms are found (Hilsenhoff 1977, Hilsenhoff 1982, Hilsenhoff 1987, Hilsenhoff 1998) that have assigned HBI values. However, in some cases this goal is not achieved. This situation can result from a number of circumstances. First, not all arthropods present in a sample are currently assigned pollution tolerance values and not all specimens in a sample can be identified to a taxonomic level at which a tolerance value has been assigned (i.e., underdeveloped or damaged specimens lacking key structures necessary for identification to lower taxonomic level). Second, the following organisms are not included in the HBI count according to the UWSP and DNR protocols: hemipterans, non-dryopid coleopterans, insect pupae, insect adults, mollusks, annelids, decapods, nemata and nematamorphans, hydrocarinians, and tubellarians. The inclusion of underdeveloped or damaged specimens in the 125 count occasionally may result in reporting of HBI values based on less than the 100 specimen minimum recommended by Hilsenhoff (1987). For example, there may have been 165 organisms in a particular sample (total sample sort) but only 90 specimens may have had assigned HBI values.

Top: Cranefly adult (family Tipulidae).

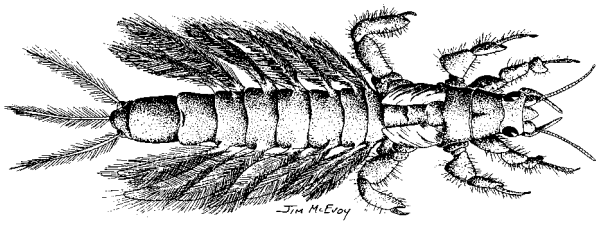
Bottom: Cranefly larva (family Tipulidae).

Interpretation of Metrics

In this section we explain how each macroinvertebrate metric is derived and how to interpret values. In many cases there are no strict rules available to assign qualitative designations to a sample metric. That is, it is difficult to say one value is "bad" while another is "good". Judgments under such circumstances remain subjective and open to debate. Nevertheless, the metric may be valuable in making *relative* comparisons of water resource quality among streams (or among stations within streams) *or* in identifying possible pollution sources. Some metric values may represent a continuum or gradient, while for other metric values a threshold may be established that links a specific value with a specific dose of an environmental stressor. For a more comprehensive review and coverage of the following topics we recommend the following three books: 1) *Biological Monitoring of Aquatic Systems* (Loeb and Spacie 1993) 2) *Biological Assessment and Criteria* (Davis and Simon 1994) and 3) *Freshwater Biomonitoring and Benthic Macroinvertebrates* (Rosenberg and Resh 1993).



Mayfly adult (family Ephemeridae).



Descriptive Metrics

Biotic Indices

HBI and FBI. The Hilsenhoff Biotic Index (HBI) and Family Level Biotic Index (FBI) represent the average weighted pollution tolerance value of all arthropods present in a sample (excluding organisms either too immature or damaged to allow for correct identification and organisms which have not been assigned a pollution tolerance value). The HBI is a well-tested metric that has been incorporated into national protocols for rapid bioassessment (Plafkin et al. 1989). The North Carolina Biotic Index (Lenat 1993) is a modified HBI index applicable for use in the ecoregions of the southeastern United States.

For HBI determinations, identification is carried to the lowest possible taxonomic level necessary to assign a pollution tolerance value (Hilsenhoff 1987). In many cases this means that an identification at the genus level is sufficient to assign a corresponding HBI value to that organism. All identifications for the FBI are made at the family level (Hilsenhoff 1988a). Anyone using the HBI or the FBI should be familiar with the field and laboratory procedures as described by Hilsenhoff (1987, 1988a). Field and laboratory procedures used for DNR can be found at the following intranet site: http://intranet.dnr.state.wi.us/int/es/science/ls/fpm/table.htm.

Sampling in sites not meeting established criteria (i.e., inadequate flow velocities, snags or pools rather than riffles or runs) seriously limit the use of the resulting data. In recent studies, comparing samples taken in close proximity within the same stream and that use a minimum of five replicates, snag data are more variable and generally give higher biotic index (BI) values than riffle samples (S. Szczytko, UW Stevens Point, pers. comm 2000). It is extremely important to emphasize that the HBI and FBI are indices of *organic* pollution and are based on a community's response to the combination of high organic loading and decreased dissolved oxygen levels. The HBI or the FBI was not intended for use outside the purpose of detecting or monitoring organic pollution. The relation of HBI values to water quality is presented in Table 3. Narf et al. (1984) provide a way to test for the detectable differences (DDs) in HBIs between samples. However, the subsequent range expansion of the HBI scale from 0-5 to 0-10 and assignment of new tolerance values makes the work of Narf et al. (1984) obsolete. To correct for the expansion of the HBI scale it is not statistically valid to multiply the DDs or HBIs by a factor of two (R. Narf and G. Lange, Wisconsin DNR, pers. comm 1997). Instead, historical BI data will have to be recalculated using the new BUGPROGRAM after changing

Table 3. *Water quality ratings for HBI values (from Hilsenhoff 1987).*

HBI Value	Water Quality Rating	Degree of Organic Pollution
≤ 3.50	Excellent	None Apparent
3.51-4.50	Very Good	Possible Slight
4.51-5.50	Good	Some
5.51-6.50	Fair	Fairly Significant
6.51-7.50	Fairly Poor	Significant
7.51-8.50	Poor	Very Significant
8.51-10.00	Very Poor	Severe

macroinvertebrate identification to correspond with changes in nomenclature and updating tolerance values. The current version of the BUGPROGRAM is able to update older databases including organism identification numbers, name changes, and tolerance values.

The HBI is seasonally dependent (Hilsenhoff 1988b). Higher HBI values occur in summer because organisms present during the summer months generally tend to be more tolerant than taxa inhabiting the same stream during the spring. To insure comparable HBI values, Hilsenhoff (1988b) recommends sampling warm water streams[2] in the spring before 440 degree days[3] accumulated and cold water streams[4] until 1050 degree days accumulated. Fall sampling can be resumed 60 days after the 440 degree day mark in warm-water streams, and 45 days after the 1050 degree day mark in cold-water streams. Please note, sampling after November 1st is not recommended. Sampling within these time windows should avoid exceeding the 95 percent confidence limits for the HBI. If sampling is conducted outside these time windows, Hilsenhoff (1988b) recommends subtracting 0.50 units from the reported HBI value to compensate for the bias.

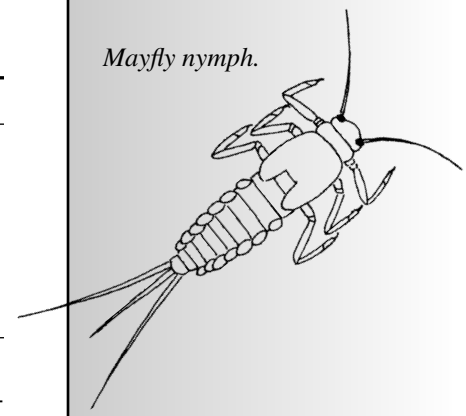
The FBI was designed as a rapid field assessment tool. As a consequence, the FBI can be less precise than the HBI. Generally the FBI underestimates the severity of pollution in highly polluted streams and overestimates the degree of impact in clean streams (Hilsenhoff 1988a, Szczytko 1988). To compensate for this bias Hilsenhoff (1988a) proposed a modified water quality rating system for the FBI (Table 4). Reporting FBI values may seem redundant when the more accurate HBI values are also available (i.e., lab-processed samples), in view of the fact that large disparities commonly occur between the two indices (Szczytko 1988). However, the HBI values generated by laboratory processed samples are potentially valuable for making direct comparisons with field monitoring assessments using the FBI technique. Irrespective of its limitations the FBI remains a valuable field tool for rapid bioassessments and screening of potential sources of impact.

[2] Defined as streams experiencing summer maxima above 20º C.

[3] Hilsenhoff (1988b) used a base of 4.5º C to calculate degree-days. Use mean daily air temperatures to compute degree-days.

[4] Defined as streams experiencing summer maxima below 20º C.

Mayfly nymph.



Ephemeroptera (Mayflies):

Presently, 115 species found within 52 genera and 19 families have been identified from Wisconsin. Larvae of all Ephemeroptera are aquatic. Most inhabit streams but several species inhabit a variety of permanent or temporary lentic habitats. Mayfly larvae are easily distinguished from other aquatic insects by having lateral or ventrolateral gills on most of the basal abdominal segments. Mayfly larvae mostly crawl about on aquatic substrates, and some burrow in soft sediment. Almost all larvae are herbivores or detritivores; only a few are know to prey on other invertebrates. Ephemeroptera ("ephemeral on the wing") do not feed as adults, and some species may live for as little as a few hours, but other species may live for as long as a few weeks.

Top and middle: Dragonfly nymph (Odonata). Bottom: Damselfly adult (suborder Zygoptera).

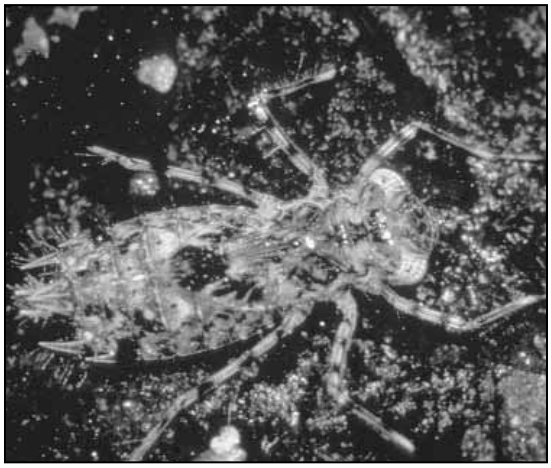
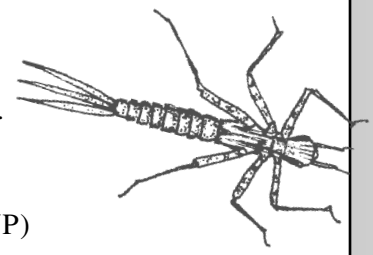


Table 4. *Water quality index for the family-level HBI (from Hilsenhoff 1988a).*

FBI Value	Water Quality Rating	Degree of Organic Pollution
≤ 3.75	Excellent	Unlikely
3.76-4.25	Very Good	Possible Slight
4.26-5.00	Good	Some Probable
5.01-5.75	Fair	Fairly Substantial
5.76-6.50	Fairly Poor	Substantial Likely
6.51-7.25	Poor	Very Substantial
7.26-10.00	Very Poor	Severe

THBI. The True HBI (THBI) was calculated by Hilsenhoff (1998) as the mean annual HBI excluding the three consecutive late spring or summer samples that had the highest HBI values and November samples. These dates are the times the HBI is not recommended by Hilsenhoff (1988b). Hilsenhoff (1998) viewed the THBI as the most accurate bioassessment of water quality for streams. Unfortunately this assumption is not supported by hard data and is open to debate. (i.e., the summer HBI values or MTV actually may be more closely correlated with trends or rankings in true water resource quality). Hilsenhoff (1998) found the metrics closest to the THBI were the annual mean HBI and the 25-Max HBI in clean and polluted streams respectively. In addition, Hilsenhoff (1998) also found that the annual mean HBI was always higher than the THBI in the streams in his study because the three summer sampling dates with the highest HBI were not included in the calculation of the THBI. Hilsenhoff (1998) recommends that diversity and species richness metrics not be used to supplement the THBI because factors other than pollution effects have a great impact on these metrics.

MTV. The Mean Tolerance Value (MTV) represents the average tolerance value of all taxa (as opposed to all individuals in the HBI) present in a sample. The MTV gives equal weight to rare and dominant taxa regardless of the abundance of the taxa (Lillie and Schlesser 1994) while the HBI places more weight on the dominant taxa. The MTV may be less susceptible to temporal changes and sample sizes than the HBI. The MTV is similar to other metrics that de-emphasize relative numerical abundance such as: the Macroinvertebrate Community Index (MCI) in New Zealand (Stark 1993), the Stream Invertebrate Grade Number Average Level biotic index (SIGNAL) in Australia (Chessman 1995), and the Average Score Per Taxon (ASPT) in Great Britain (Armitage et al. 1983, Wright et al. 1988). The ASPT is less sensitive to sampling effort and seasonal effects than its counterpart quantitative metric, the



Damselfly nymph.

Biological Monitoring Working Party (BMWP) score (Armitage et al. 1983).

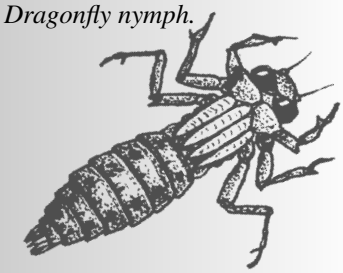
The MTV is intended to serve as a companion metric to the HBI and should not be used independently from HBI evaluations! In most cases the MTVs should exhibit a pattern similar to that presented by the HBI. Large discrepancies between the two metrics should indicate that further examination of the data is needed. For example, if large abundances of a relatively cosmopolitan species with high tolerance values are found in a sample otherwise dominated by less tolerant species, this may result in an inaccurate HBI value that is not reflective of the 'true' water quality of a particular stream. Likewise, the introduction of a few relatively intolerant taxa from a clean upstream tributary into a poor stream may substantially lower the MTV and bias the interpretation of the data. In general MTVs are similar to FBIs in that they have the same apparent bias relative to HBI values. The MTVs tend to be lower than the corresponding HBIs in highly polluted situations and higher than HBI scores in clean streams (Lenz and Miller 1996, Hilsenhoff 1998). This suggests that the HBI water quality rating system is not appropriate for use with MTV scores and that a separate rating system needs to be developed for MTVs.

After evaluating the effects of a range of maximum numbers (5, 10, and 25) on HBI values Hilsenhoff (1998) proposed a new index, the 10-Max BI. Hilsenhoff (1998) suggests limiting the maximum number of individuals of each taxon in an HBI sample to 10 as a means to reduce the effects of seasonal variability on the HBI. The 10-Max BI limits the effect of dominant organisms and does not elevate the importance of very rare taxa to the same level as the MTV. The use of the 10-Max BI may be superior to the HBI in evaluating polluted streams (Hilsenhoff 1998). The BUGPROGRAM allows the user flexibility in selecting a maximum number of individuals (i.e., 5, 10, 25, or any other number) to calculate a new HBI referred to as the Max-*x* HBI value. Setting the maximum value to 1 will produce the equivalent to the MTV. Setting the maximum value to 10 will produce the 10-Max BI recommended by Hilsenhoff (1998).

Richness Measures

Richness measures represent the number of distinctly different taxa found in a sample. A richness value does not represent the total number of taxa at a site, but rather it is a relative measure or index. Often it is only necessary to process a small fraction of a sample to compute an HBI value. In the case of a BUGPROGRAM report a richness metric is reported on the basis of the numbers of taxa present in a HBI subsample (i.e., modified fixed count subsample). The remainder of the sample is not included in the calculations and any information regarding additional taxa present at the site is lost. This is not intended to be a criticism of the HBI but is reflective of the established laboratory procedures and the need to keep processing costs down. The loss of information is an unfortunate by-product of the established fixed count laboratory procedure. This has significant ramifications

Dragonfly nymph.



Odonata (Damselflies and Dragonflies):

Presently, 154 species in the order Odonata have been found in Wisconsin, represented by 3 families, 19 genera, and 45 species in the sub-order Zygoptera (damselflies), and 6 families, 38 genera, and 109 species in the suborder Anisoptera (Dragonflies). The larvae of all species are aquatic with about two-thirds being lentic and one-third inhabiting lotic environments. Lotic-dwelling larvae occur in all types of permanent stream habitats, including gravel and rock riffles, debris along streambanks, bank vegetation, soft sediments and sand; occasionally they are found along the wind-swept shores of lakes. Lentic larvae inhabit permanent and temporary ponds, wetlands, and littoral zones and shoreline areas of lakes. Life-cycles are relatively long and range from one to four years. Most Odonate larvae found in Wisconsin can be identified to species.

Top: Stonefly nymph (Plecoptera).
Bottom: Stonefly adult (Plecoptera).

with respect to calculations and the use of other metrics derived from the sample. Consequently, the data derived from the HBI subsamples represent relative measures per total number of specimens examined.

Although high taxa richness is generally associated with good water quality, low taxa richness does not necessarily indicate poor water quality, nor does high richness always indicate good water quality. Some habitats (i.e., small cold headwater streams, or oligotrophic streams, or mineral poor waters) may naturally have low numbers of taxa density per unit area. Crunkilton and Duchrow (1991) demonstrated a peak in total taxa richness in middle order streams in Missouri. In some cases, intermediate or low levels of disturbance can cause an increase in taxa richness (Townsend et al. 1997). This could be obtained either through physical mechanisms (i.e., increased heterogeneity of substrate niches), biological interactions (i.e., addition of invasive or pioneering species without loss of native species), or increased nutrients and energy. While some water quality rating scales have been established (Szczytko 1988, Barbour et al. 1996) for various richness metrics in other regions, no scale has been established for Wisconsin streams. However, see discussion of La Liberte (unpub. data) in the Future Directions section of this document. Until a Wisconsin stream water quality scale is established relative comparisons of richness values with reference stations of a similar stream class and ecoregion is recommended.

There are three taxa richness metrics reported by BUGPROGRAM at two taxonomic levels: Species Richness (SR), Generic Richness (GR), and EPT Generic Richness (EPTG).

Species Richness (SR). SR is based on a count of the number of species identified in a HBI sample (Note: If an unidentified specimen is keyed to the genus level and not the species level the BUGPROGRAM will count the specimen as a species within the genus if no other specimens in that genus are identified. Therefore it is possible that unidentified specimens will count as only one species instead of multiple unidentified species within a genus). This count is not a true species count due to: 1) the lack of adequate keys for many aquatic larvae among some orders, 2) the condition and life stage of particular specimens, and 3) excellent taxonomic keys are available to species but identification beyond genus is not completed or required in calculating HBIs because all members of the genus have been assigned the same pollution tolerance value (i.e., elmid riffle beetles). As a result, SR may be based on a mixture of taxonomic levels and may result in an underestimation of true total taxa richness. If the influence discussed above is consistent across samples SR will continue to have merit in evaluating wide disparities among samples.

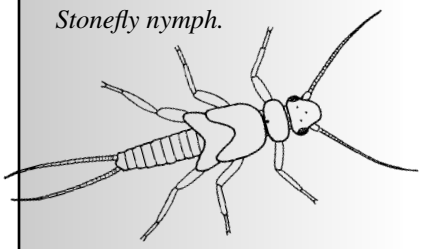
Generic Richness (GR). GR refers to the number of different genera represented in a biotic index subsample. As noted in the case of SR above, a single unidentified specimen that is identified to family but not to genus will be counted as a genus within that family if no other specimens in that family are identified. The points discussed above for SR also hold true for GR with the exception that most specimens are identified to the genus level.

EPT Generic Richness (EPTG). The third richness metric reported by the BUGPROGRAM is EPTG. This metric represents the number of distinct genera found only among the orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) in a biotic index subsample. These three orders are separated from other aquatic taxa because they *generally* represent the more organic pollution intolerant organisms present in rivers and streams. Consequently the EPTG is believed to be a more sensitive metric to organic pollution than SR or GR. The number of EPT taxa generally decreases with increasing perturbation (Barbour et al. 1996, Wallace et al. 1996, Growns et al. 1997). Use of the EPTG metric is somewhat redundant to the HBI (similar response characteristics) and has a higher degree of variability based on the Coefficients of Variation (CVs). This may be due to differences in the scales used in the two metrics (see highlight box below). Hilsenhoff (1998) reports that EPTG index values are seasonally variable and dependent on stream-order size. The EPTG metric has been applied successfully in numerous studies and has been incorporated into various state biomonitoring programs.

Metrics are often judged on the basis of their relative coefficients of variation (CVs) and recent studies have addressed the statistical properties of benthic macroinvertebrate metrics (Szczytko 1988, Resh 1994, Rheaume et al. 1996). One aspect of the metrics that has not been fully explored is that of scale. Many metrics are open-ended (i.e., scales range from zero to infinity) while others are restricted. The influence that restricted scales may have on CVs and other mathematical or statistical properties of the various metrics may be significant (author's conclusions). Direct comparisons between open ended richness metric CVs and other restricted metrics CVs may not be a fair method of estimating the significance of richness metrics (Diamond et al. 1996). Biological relevance is most important when determining value of richness metrics. For example, one metric may have higher CV than another but that same metric may display a wider range in values, thus having greater discriminatory power (Barbour et al. 1996). Likewise, while some metrics may be less significantly correlated with resource conditions than other metrics, those same metrics may be more effective in distinguishing between disturbed and undisturbed conditions (Fore et al. 1996). Highly variable metrics may exhibit a threshold response to human influence at a point in their range (Fore et al. 1996) which may make them useful in assessments. Consequently, direct comparisons of CVs among different metrics should not be the sole means of judging the merit of metrics. Rather an evaluation using a combination of metric CV and the discriminatory power of the metric to detect differences between test and reference sites should be applied (Diamond et al. 1996).

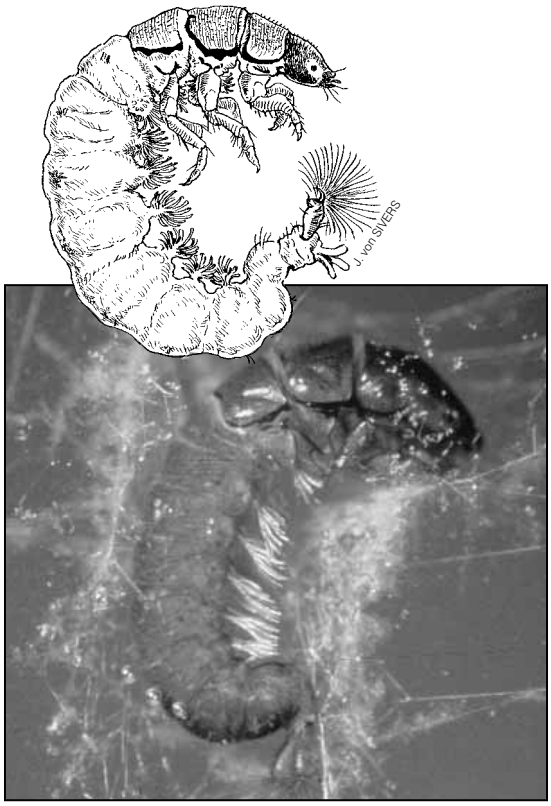
Caution should be taken when interpreting various richness metrics. Richness measurements derived from biotic index subsamples may be misleading. It is well documented that there are pros and cons associated with fixed count subsampling and problems related to interpretations of

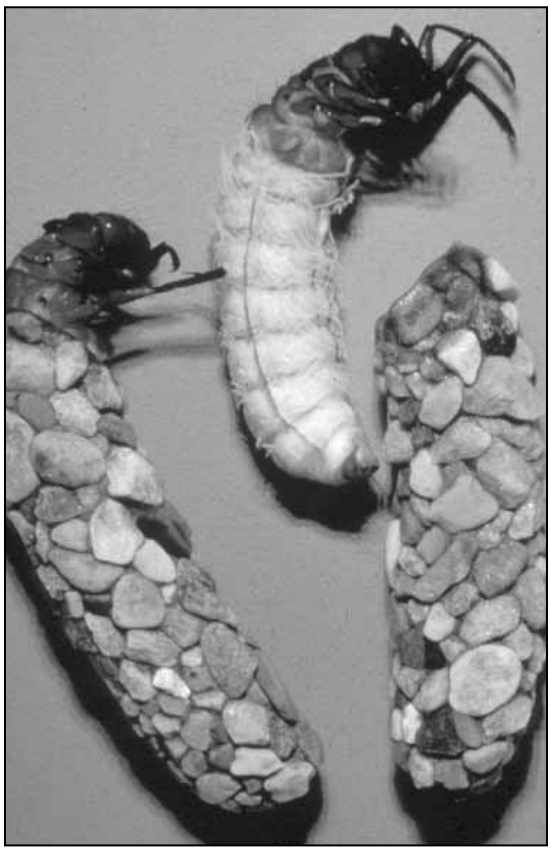
Stonefly nymph.



Plecoptera (Stoneflies):

Presently, 58 species of stoneflies in 8 families and 25 genera have been identified from Wisconsin. All larvae are aquatic, and almost all inhabit streams; larvae of a few species may live in cold oligotrophic lakes. Stonefly larvae differ from most aquatic insects by having two long filamentous caudal cerci (tail filaments) and an elongate or flattened appearance. Larvae obtain respiratory-oxygen from water through their cuticle or primitive gill structures, and as a result are relegated to cold, fast-flowing, highly oxygenated streams. Feeding habits vary among families with most being herbivore-detritivores, although several families have predatory species. Lifecycles are relatively long with some taxa living 3 years or more in the nymphal stage. Some species of stoneflies have some of the earliest emergences in the year, hatching as early as January and February, and the adults are often seen crawling on the snow along streambanks.





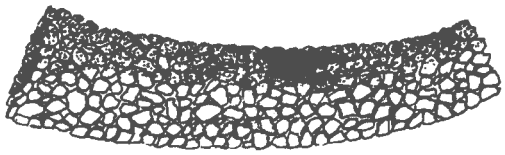
Top: Net-spinning caddisfly larvae (family Hydropsychidae).
Bottom: Case-making caddisfly nymphs and cases (family Limnephilidae).

richness metrics derived from such counts (Courtemanch 1996, Barbour and Gerritsen 1996, Vinson and Hawkins 1996, Somers et al. 1998, Growns et al. 1997, Larsen and Herlithy 1998, Cao et al. 1998). The standard protocol used in sorting HBI samples outlines counting a minimum of 100 organisms (Hilsenhoff 1987; note the UWSP Benthic Macroinvertebrate Laboratory counts a minimum of 125 organisms). Thus, in most samples only a subsample of the organisms present are counted while noting what percent of the total sample is examined.

For example, in one sample only ten percent of the total number of organisms present is counted and included in the taxa richness determination. In a different sample the entire sample is searched meticulously to reach the minimum number of organisms required to include in a taxa richness evaluation. If the SR is 15 in the first sample (ten percent examined), and the SR is 25 in the second sample (100 percent examined), one should note that these values represent the number of distinctly different taxa present in the *portion* of the sample that was enumerated. Because the percent of the sample that was examined to reach a 'fixed-count' is different between the two samples, it is quite possible that both samples contained a true total of 25 taxa. Barbour and Gerritsen (1996) propose to define this value as taxa density (as opposed to taxa richness). They suggest that while taxa richness may be a desirable metric in some circumstances (assuming equal areas sampled) relative taxa density is more economical and has greater discriminatory power than taxa richness. Others argue that comparing relative taxa richness (or any other relative richness metric) between any two samples may be questionable because one sample may have an inordinate abundance of a particular taxon that may skew the count and result in missing the rarer taxa.

Richness metrics are strongly dependent upon a correlation between the area sampled and the number of individuals examined (Vinson and Hawkins 1996). It is important that field sampling efforts in collecting biotic index samples be standardized to the greatest extent possible (i.e., choose equal sample areas of representative substrates[5]). As outlined earlier, fixed count subsampling may underestimate the true or absolute taxa richness of a sample (or stream). However, Vinson and Hawkins (1996) found that estimated taxa richness (using a rarefaction technique presented by Hurlbert 1971 that "normalizes" taxa counts to a standard

[5] It is the opinion of the authors that the DNR Field Manual should be amended to reflect the fact that restricting kick sampling to too small of an area within a riffle may seriously bias a sample due to the tendency for aquatic organisms to be highly aggregated. It is preferable to collect a composite sample representing a wider area of a riffle to gain a better representation of the entire macroinvertebrate community present.

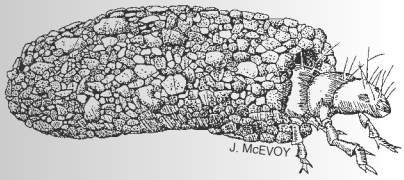


Example of a case-building caddisfly case.

number of organisms examined) in samples containing as few as 200 individuals, were equally sensitive in defining differences in richness among streams from several ecoregions (Omernik 1987). Vinson and Hawkins (1996) also suggest that fixed-count subsampling is a mechanical form of rarefaction and that data derived from counts ranging between 100-200 individuals may be sufficient to detect differences among samples. Barbour and Gerritsen (1996) suggest that fixed count subsampling provides a repeatable estimate of taxa richness (i.e., relative number of taxa per standardized number of organisms).

If the objective of estimating taxa richness is to compare differences in relative taxa richness among samples, then data from fixed count subsamples may be adequate. However, it should be emphasized that this metric does not equate to true total taxa richness but rather to area density (taxa density per unit area). Larsen and Herlithy (1998) suggest that a fixed count of 300-500 individuals will produce a richness value that will approximate area density. Somers et al. (1998) found that counts of 200 or 300 produced very little gain in discriminatory power over counts of 100 among lakes using a variety of indices *except* for richness metrics where larger counts were deemed better! Doberstein et al. (2000) show that subsampling severely limits the discriminatory power of most metrics and the ability to accurately assess biological condition. Courtemanch (1996) explains that expression of richness metrics based on fixed counts lacks ecological interpretative value. All in all, interpretation of BUGPROGRAM taxa richness values should be made with care.

Richness measurements derived from quantitative samples (i.e., Hess stovepipe samples, cores, and some artificial substrate samplers) are less controversial when the entire sample is examined (see precautions listed by Palomaki and Paasivirta 1993). However, there are other alternatives to processing the entire sample. Vinson and Hawkins (1996) suggest that implementation of a two phase subsampling technique (Cuffney et al. 1993) with more extensive habitat sampling (and pooling of samples) would represent a compromise to expand the taxa lists for particular sites. The two phase subsampling includes searching the entire sample for large or rare taxa present. Courtemanch (1996) suggests a method which consists of serial processing of additional subsample cells until the recovery of new taxa levels off, or reaches an asymptote. Both of these alternatives have a common weakness associated with the visual recognition of new taxa. While a larger rare taxon may be immediately recognized as representing a new taxon, scanning additional cells may overlook numerous small, nondescript species, which normally would only be identified as a new species after examination at a higher magnification. Barbour and Gerritsen (1996) clearly point out that subsampling is intended to provide an unbiased estimate of a larger sample. Interpretations of data and comparisons based on relative compositions of randomly selected subsamples are indeed valid if standardized and rationalized appropriately. Much of the controversy regarding richness metrics appears to be clouded by semantics and lack of clear definitions. The issue is certainly not resolved and further thought and discussion are warranted.

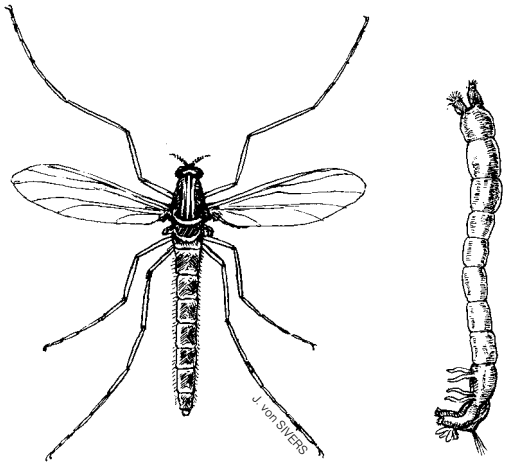


Trichoptera (Caddisflies):

Presently, 245 species of caddisflies in 19 families and 72 genera have been identified from Wisconsin. Caddisfly larvae are a very important faunal component of most streams, but half of the families also have species occurring in lentic environments as well, some even in temporary ponds. Caddis larvae use silk to spin nets for food collection or to construct tubular shelters. The structures vary by size, shape, and use of streambed organic and inorganic materials for construction. For some taxa the cases can be used to identify the animal to genus and sometimes species. Cryptic cases protect some species from predation, while certain case-forms allow other taxa to inhabit fast currents, or provide protection from abrasive scour. All taxa pupate in closed silken cocoons. The adults tend to be drab-colored and moth-like in their shape and erratic flight behavior.

Diversity Measures

Diversity indices represent a measure of the distribution of individuals among different taxa present in a sample. In theory, a macroinvertebrate community consisting of many taxa of even distribution (i.e., relative abundance) is considered more natural than a simple community dominated by one or few taxa (see review in Winget and Mangum 1979). However, it should be noted that a simple community can result from natural conditions (Winget and Mangum 1979).



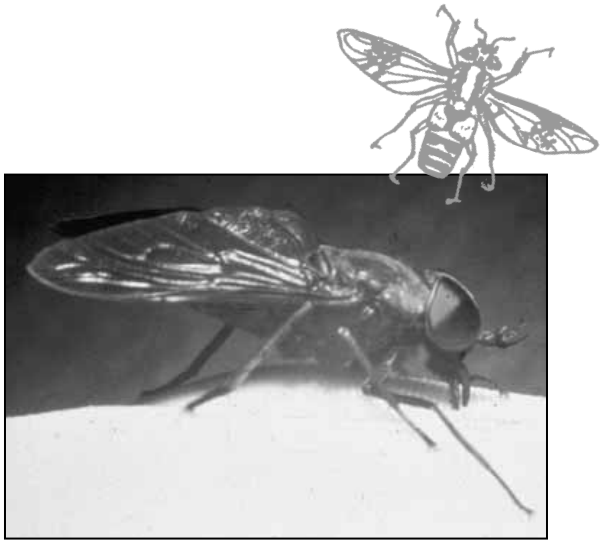
Midge adult and larva.

Diversity Index *(d)*. Former versions of the BUGPROGRAM reported values for Diversity Index (*d*) which was calculated at the generic level based on the logarithmic relationship between the number of individuals in each genus and the total number of organisms in the sample (see equation below).

$$d = (N \log_2 N - \Sigma n_i \log_2 n_i)1/N$$

Where 'N' = total of all arthropods in sample,
and 'n_i' = number of individuals in each genus.

The formula used to compute the Diversity index was actually a variant of the Margalef index as modified for stream arthropod analysis by Hilsenhoff 1977 (see Washington 1984 for further explanation). Consequently, when the BUGPROGRAM was developed, the Diversity index was referred to as the "Margalef Diversity index" despite the fact that technically it was not the same.



Deerfly (family Tabanidae).

The Margalef index is seldom used today (Washington 1984). Boyle et al. (1990) showed that the response of Margalef's diversity value to theoretical perturbations was erratic. Resh and McElravy (1993) conclude that less than five percent of macroinvertebrate studies surveyed used the Margalef's Diversity index. Despite the low usage, Resh and Jackson (1993) report that Margalef's Index proved superior over other diversity indices in detecting impacts of acid and toxic metals inputs. Magurran (1988), in her review of ecological diversity measures, considers the Margalef's index advantageous to use due to its simplicity. She admits that Margalef's index is applied infrequently but concludes that the index could be an important tool because it is easy to interpret and statistically and ecologically sound (Magurran 1988).

Shannon's Index Of Diversity (H'). A recent modification to the BUGPROGRAM included replacing Margalef's Index (as modified by Hilsenhoff 1977) with the Shannon Diversity index (H'). Resh and McElravy (1993) found that H' is the most commonly applied index in a survey of 90 lentic and lotic benthos field studies. The Shannon Diversity

index, which often is called mistakenly the Shannon-Wiener index (see Magurran 1988 for further discussion), is represented by the formula:

$$H' = -\Sigma\, p_i \ln p_i$$

Where p_i = the proportion of individuals represented by each taxon.

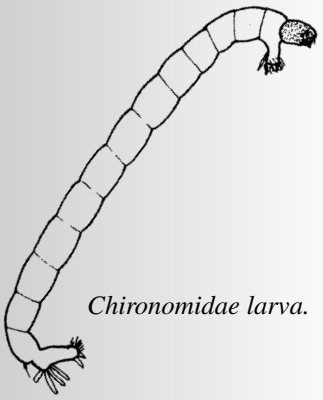
H' values range between 1.5 and 3.5 with higher values representing higher diversity.

The Shannon Diversity index is based on two assumptions that may be violated in BI sampling (Magurran 1988). These assumptions are: 1) random sampling of an infinite population occurs, and 2) that all species present are sampled. It becomes important to define adequately and sample the particular areas that we wish to compare. Are we interested in comparing diversity among microhabitats, riffles, reaches, or rivers? It is important to randomly sample an area adequate enough to provide a representative picture of what taxa are present and the relative proportions that are represented by these taxa. Samples not collected randomly (i.e., artificial substrate samplers or light trapping) promote the use of an alternative diversity index such as the Brillouin index (see Magurran 1988).

DNR HBI samples may not be conducive to interpretation with the Shannon Diversity index. During HBI lab processing some individuals are identified to species level, some to genus level, and some to even coarser taxonomic levels. During field collections riffle kick sampling may include taxa from fine, depositional sediment areas, while other riffle kick samples may be restricted to coarser sediments. It is not clear how these differences influence H' values.

Diversity values, irrespective of the index chosen, may vary directly with water quality and low diversity may indicate an unstable community. However, cold, clean, headwater streams may have low diversity and still represent excellent water quality. In some cases the diversity index value does not respond as expected due to the replacement of intolerant taxa by an equal number of tolerant taxa. Hilsenhoff (1977) found that the best stream (ranked by the lowest mean HBI value) in his study was only ranked 32 out of a total of 53 by Margalef's diversity index (using Hilsenhoff's formula) and the second best stream ranked only 45 out of 53.

Diversity indices have undergone severe criticism (Hurlbert 1971, Norris and Georges 1993). Many diversity indices are dependent upon sample size, which may not be controlled in BI sample analysis (i.e., while a minimum of 125 organisms are counted, the actual number counted and included in the diversity index computation may exceed this number by more than 50 percent). In addition, no consensus has been reached as to what levels of change in diversity index values when shown to be statistically significant are *biologically* significant. As a result, little value can be placed on Margalef's Diversity index or Shannon Diversity index values as computed by the BUG-PROGRAM. Perkins (1983) reviewed the performance of eleven diversity indices and concluded that Shannon's index should not be used alone, however, Florida currently uses the Shannon-Wiener Index as a biocriterion (Barbour et al. 1996). Although Norris and Georges 1993 suggest that diversity indices may be of some value if used with caution, interpretation of results remains open to debate and is largely inconclusive.

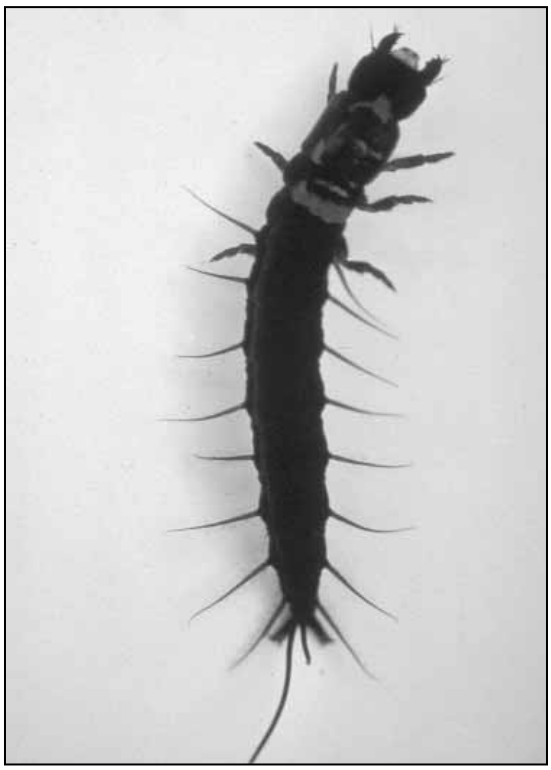


Chironomidae larva.

Diptera (Aquatic Flies and Midges):

While primarily a terrestrial order Diptera is the dominant order of aquatic insects in Wisconsin, with 660 species estimated to occur in the state distributed among 19 families and at least 185 genera. Larvae and pupae of many species are aquatic, and account for more than one-third of all aquatic insect larvae found in Wisconsin. More than one-third of aquatic Diptera are in the family Chironomidae. Larvae inhabit all types of aquatic environments, and often dominate the invertebrate fauna in lentic environments. Diptera larvae are easily recognized by their lack of segmented thoracic legs. Taxonomic identification to the species level is not possible for the larvae of most families.

Top: Dobsonfly larva (Megaloptera).
Bottom: Alderfly adult (Megaloptera).



Functional Feeding Classes

Several metrics generated by the BUGPROGRAM are based on a trophic functional feeding classification (Cummins 1973, Cummins 1974, Cummins and Klug 1979, Merritt and Cummins 1984). Functional feeding groups represent general modes of food acquisition based on an organism's principal feeding mechanism. Cummins and Merritt (1996) assign functional feeding class designations for most aquatic insects based on research conducted by themselves and others on food habits, diet, and mouth part morphology. Wallace and Webster (1996) provide a current review of the role of macroinvertebrate functional feeding groups in stream ecosystems.

The trophic structure of the community reveals much about the character of a stream. Food sources, substrates, and contaminants play important roles in shaping (i.e., establishing a templet for) the invertebrate community. The effects of riparian land use and stream side vegetation potentially have strong influence on trophic metrics (Sweeney 1993, Weigel et al. 2000). The classic river continuum concept states that trophic functional feeding classes reflect a combination of external (allochthonous) and internal (autochthonous) energy inputs to the stream. As a result, location in the stream relative to stream order or size (i.e., headwaters versus large, high order rivers), dams, lake outlets, tributary streams, riparian or general land use in the watershed, and ecoregion or climatic influences each impact upon the character of the invertebrate community inhabiting any reach of stream (Vannote et al. 1980).

Biotic indices are designed to detect organic impacts whereas functional feeding group metrics are potentially useful for detecting the impact of both organics and toxicants (Plafkin et al. 1989). Fore et al. (1996) found that feeding ecology metrics were not able to distinguish between most and least disturbed sites in evaluating the effects of logging operations in Oregon streams. Similarly, James Karr (University of Washington, pers. comm. 1988) concluded that trophic measures are not especially useful in examining the effects of human actions on invertebrate assemblages.

The BUGPROGRAM reports relative composition represented by five functional feeding classes among total individuals and total genera in a sample. The five functional feeding classes reported are: scrapers, filterers, shredders, gatherers, and collectors (note: filterers and gatherers are subcategories of collectors). The ratios of scrapers to filterers, gatherers, and collectors are also reported. Predators, parasites, and piercers are not reported in the BUGPROGRAM report. Assignment of Wisconsin taxa to these classes is based on a current literature review and classification by Merritt and Cummins (1984).

These metrics are useful in characterizing the food base of a community. However, effects of toxicants adsorbed onto particulate food items often confound interpretations. Stream size and the concepts of river continuum theory are also important considerations in interpreting the metrics derived from functional feeding classes. It should be added that many organisms shift from one feeding class to another as they advance through their respective life stages from early instar to adult.

Scrapers. The class scrapers include various herbivores and detritivores that graze periphyton (in particular diatoms) and attached microflora and fauna on mineral or organic surfaces. Examples include many mayflies (especially Heptageniidae) and caddisflies (including Glossosomatidae). A high proportion of scrapers is indicative of an abundant supply of periphyton. Filamentous algae and mosses can interfere with feeding by scrapers. Kerans et al. (1992) and Barbour et al. (1996) report that the proportion of grazers to scrapers decreases with increased human impact. However, Fore et al. (1996) reports a variable response to human impact.

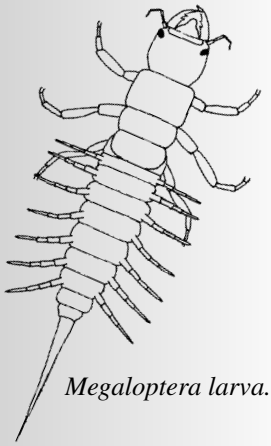
In a California study, Hannaford and Resh (1995) found that scraper metrics were not useful in discriminating among restored, unrestored, and reference sites. Conversely, in another California study, Resh (1994) found that percent scrapers was the only functional measure to indicate a significant change following an oil spill.

Filterers. Filterers are a subcategory of collectors. The class filterers include various suspension feeders consisting of a combination of detritivores, herbivores, and carnivores. Examples include the simuliids and net spinning caddisflies. Because filter feeders feed on suspended fine particulate organic material (FPOM) and often use filamentous algae for attachment sites, a high preponderance of filterers may suggest increased organic enrichment. Filterers are sensitive to toxicants often associated with fine particles, and as a result, large inputs of toxicant laden particles may cause a reduction in filterers under certain circumstances. The presence of rich deposits of FPOM and filamentous algae in combination with low counts of filterers may be a good indication of such a condition.

In one selected study (Kerans et al. 1992) percent filterers was one of only six metrics that responded in a similar direction in both pools and riffles. Kerans and Karr (1994) include percent filterers as one of 13 community attributes in their benthic index of biotic integrity. According to Barbour et al. (1996) percent filterers decreases with increasing perturbation. However, Fore et al. (1996) indicate a variable response to human disturbance.

Shredders. The class shredders includes a large group of detritivores and herbivores that feed on both live and dead matter. Examples include many stoneflies especially Pteronarcyidae, Peltoperlidae, Leuctridae, Taeniopterygidae, Nemoridae, and Capniidae. Among the Diptera, Tipulidae are the representative shredders. Shredders feed primarily on the coarse particulate organic matter (CPOM) and subsequently Resh and Jackson (1993) suggest that percent shredders should be assessed using leaf pack samples.

This class is particularly sensitive to riparian zone influences and land use. Barbour et al. (1996) found that percent shredders decreases with



Megaloptera larva.

Megaloptera (Fishflies and Alderflies):

This relatively small order of insects has two families Corydalidae (fishflies and dobsonflies), and Sialidae (alderflies), four genera and 11 species with aquatic larvae that have been identified from Wisconsin. The larvae are elongate and slightly flattened and may reach 90mm in total length. The larvae have large mandibles, and are voracious predators and can have strong predatory influences on the invertebrate community. Fishflies and dobsonflies inhabit coarse rocky substrate in well-oxygenated streams, whereas alderflies occur in both lotic and lentic environments, usually in silt deposits. The adults are weak fliers and tend to hide in streamside vegetation upon emergence.

increasing perturbations. The lack of allochthonous inputs of leaves and associated microbial colonizers due to the lack of wooded areas adjacent to and upstream from a stream site will be reflected in a decreased importance of shredders. Hannaford and Resh (1995) reported that percent shredders discriminated among restored, non restored, and reference streams despite a relatively low level of precision. The accumulation of terrestrial toxicants (i.e., pesticides and herbicides) on leaf surfaces can contribute to a substantial loss of shredders (Plafkin et al. 1989), but Fore et al. (1996) showed a variable response in this metric to human disturbance. Kerans and Karr (1994) excluded shredders from their benthic index of biotic integrity because the metric misclassified sites in comparison with the fish IBI assessments.

Top: Giant water bug adult (family Belostomatidae).
Bottom: Riffle beetle adult (family Elmidae).

Gatherers. The class gatherers include detritivores and herbivores that are deposit feeders (taxa living in and feeding upon items found in or on the surface of surficial deposits). Representatives include many dipterans (particularly chironomids), some trichopterans, and mayflies.

Kerans et al. (1992) suggest that the proportion of gatherers increases with increases in human impacts, while Barbour et al. (1996) and Fore et al. (1996) indicate a variable response for this metric. Barbour et al. (1996) point out that gatherers may dominate under conditions of organic enrichment, but they can be severely reduced if the stressor is a form of toxicant. Kerans and Karr (1994) tested but did not use gatherers in their benthic index of biotic integrity because the metric misclassified sites as compared with fish IBI assessments.

Collectors. This class is the sum of gatherers and filterers. Because the response of gatherers may be variable (Barbour et al. 1996, Fore et al. 1996), the response of collectors may also be indeterminant. Barbour et al. (1996) suggest that because collectors and filterers are generalists, they may be more tolerant to certain forms of pollution. Consequently, the relative contribution of collectors in a sample may increase with more severe perturbations due to the loss of more intolerant taxa.

Ratio Measures

Metrics derived from ratios generally exhibit wider fluctuations than non-ratio metrics (Plafkin et al. 1989, Barbour et al. 1992), are difficult to interpret biologically (Kerans et al. 1992) and may have serious statistical shortcomings (Jackson et al. 1990, Berges 1997).

There are three ratio metrics computed by the BUG-PROGRAM. The ratio of percent scrapers to percent filterers has the clearest meaning. A high percentage of scrapers

to filterers generally reflects an unbalanced community and points to the relative abundance ratio of CPOM to FPOM. Generally stream orders four through six (Cummins 1974) contain organisms that are dependent upon light reaching the stream bottom to support attached algae (diatoms and filamentous forms). Increased algae growth will in turn support grazers and scrapers. However in deep high order streams (orders seven through twelve) shading and increased concentrations of suspended organic and inorganic particles reduce light penetration and reduces attached algae growth. A reduction of algae contributes to an increase in filterers relative to scrapers.

Dominance Measures

The contribution a particular taxon (family, genus, or species) to the total number of individuals represented in a sample provides an indication of relative dominance, and is a simple estimator of evenness (Plafkin et al. 1989). A healthy and stable community should contain a diverse group of organisms with few dominant taxa. The percentage of a dominant organism (irrespective of the identity) increases with increasing perturbation (Barbour et al. 1996).

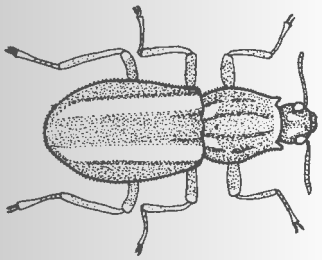
The BUGPROGRAM reports the five most dominant families, genera, and species in a sample. Communities dominated by a few taxa with high combined percentages generally reflects a disturbed situation, whereas communities dominated by a good mixture of taxa with lower combined percentages indicates a more balanced and healthy condition. It is difficult to provide guidelines for interpreting community structure based on dominants because a myriad of natural communities exist.

Aside from dominance by functional feeding groups (see section on functional feeding classes), certain taxa may indicate that particular conditions exist. For example, percent Ephemeroptera, percent Plecoptera, percent Trichoptera, and percent Amphipoda all decrease with increasing perturbation; however, percent Odonata, percent Diptera, and percent Isopoda all increase with increasing perturbation (Barbour et al. 1996). The percent contribution of dominant taxon metric has been shown to be sensitive to sub-sampling and collector bias (Hannaford and Resh 1995). Additionally, Plafkin et al.(1989) found that dominance metrics are not particularly sensitive to moderate amounts of organic or toxic loadings. Impairment (or nonimpairment) response is most evident at the extremes.

Comparison Metrics

There are a number of ways by which macroinvertebrate communities may be compared. Various non-parametric and parametric tests may be used to compare single test metrics between pairs of stations or among multiple stations. These comparisons may represent upstream-downstream studies (i.e., above and below discharge point), before-after (i.e., pre-post treatment or spill), or control-impact (i.e., treated versus untreated). Bioassessment of biological condition is best conducted by comparison with a reference site of a similar position or class (Plafkin et al. 1989). The current emphasis is on comparison with reference or least-disturbed sites by ecoregion and stream type. Staff may best be advised to consult with a biostatistician prior to beginning any detailed investigation or analyzing existing data.

Riffle beetle larva.



Riffle beetle adult.

Coleoptera (Beetles):

Coleoptera is the largest order of both terrestrial and aquatic insects. Presently, 369 species found in 14 families and 82 genera have been identified from Wisconsin. Size, structure, distribution, habitats, lifecycles and feeding habitats vary widely among aquatic beetle families. Almost all adult beetle species can be identified to species, whereas the taxonomy of the larval stages is poorly understood.

Neuroptera larva.

Neuroptera (Spongillaflies):

While Neuroptera is a large terrestrial order, larvae in only one family found in Wisconsin are aquatic. The larvae of aquatic Neuroptera are parasitic on freshwater sponges. Eggs are laid on terrestrial vegetation above aquatic habitats inhabited by sponges. Larvae hatch and drop into the water and drift with the current or swim until a sponge host is found. After completing development the larvae crawl to shore spin a cocoon for pupation. The emerging adults will live for a few weeks feeding on nectar. Most aquatic Neuroptera overwinter as larvae in sponges.

Paired Comparison Metrics

In addition to descriptive characteristics, six paired community comparison metrics are available through the DNR-BUG program. These include both similarity and dissimilarity indices (Table 2). Despite the high degree of variability[6] associated with similarity indices (Szczytko 1988) their application to bioassessments is not entirely without merit. In fact, community similarity indices are listed among recommended rapid bioassessment protocols (Plafkin et al. 1989) and continue to be applied successfully in many studies (Pearson and Pinkham 1992, Lillie and Isenring 1996). Interpretation of similarity index values with respect to condition or biological impairment is difficult. Determinations of similarity or dissimilarity are often obscure and arbitrary with discriminatory threshold values differing among indices. However, in rapid bioassessments, Plafkin (1989) and Hannaford and Resh (1995) recommend percent similarity thresholds of 83 percent and 65 percent respectively. Comparisons with a reference stream (or historical reference condition on the same stream) and other known impacted sites are recommended. As is the case with most metrics replication in terms of either samples, closely spaced stations, or different dates is recommended if time and funding permits.

Ratio Metric Comparisons

The Coefficient of Community Loss (CCL) may be used to compare changes (before-after) or differences (control-impact) in communities based on simple taxa presence or absence. The CCL represents a ratio metric that corresponds to loss of taxa at an impacted site relative to a control site. The CCL differs from other similarity metrics in that the values range from 0 to infinity. Low values suggest little change or difference between samples while high values indicate greater impacts. Courtemanch and Davies (1987) discuss the range in CCL values relative to different levels of impacts. Barbour et al. (1992) reported finding the CCL to be unacceptable in discriminating differences among macroinvertebrate data in eight ecoregions.

[6] Judging the relative value of individual metrics purely on the basis of CV comparisons is not recommended. Metrics with open-ended scales may have higher CVs than metrics with restricted or limited scales purely as an artifact of their mathematical properties. Likewise, caution should be taken in evaluating the utility of similarity indices based on CVs. By the very nature of the paired comparisons, variation is magnified. The use of replicates actually introduces greater opportunity for increased variation because the weight of outliers is twice that of its representation.

Presence/Absence and Percentage Comparison Metrics

The BUGPROGRAM computes five other similarity-dissimilarity metrics with values ranging from 0 to 1. These metrics include: coefficient of similarity (*CS* and *B*), Stander's similarity index (SIMI), percentage similarity (*PS*), and ecological distance (EDIS). With the exception of the EDIS, which is a dissimilarity index, low values represent dissimilar communities while high values represent more similar communities. The calculation of each of these metrics differs slightly (see Szczytko 1988 for formulas). The Coefficient of Similarity (*CS*) is based on simple presence or absence of taxa which allows application to both quantitative and semi-quantitative data. The remaining similarity metrics are based on relative taxa abundance in paired samples.

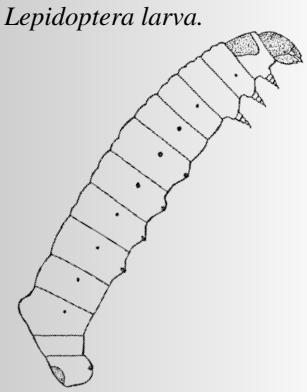
Each metric has its own set of strengths and weaknesses which derive from their mathematical calculation. Some are subjective in their interpretation (i.e., SIMI and EDIS) while other indices are more sensitive to dominant (i.e., SIMI) or rare taxa (Coefficient *B*; Brock 1977). Currently the CCL, *B* similarity, and the Jaccard Coefficient of Community are recommended in rapid bioassessment protocols (Plafkin et al. 1989). The Bray-Curtis Index is favored by some investigators because it varies linearly relative to changes in species numbers and abundance (Norris and Georges 1993) and is said to reflect 'true' similarity (Bloom 1981). While similarity-dissimilarity indices have great potential in comparing invertebrate communities (Plafkin et al. 1989), no single index can be recommended over another. It is recommended that more than one metric be applied whenever possible.

Coefficient of Similarity (*CS*) And Pinkham Pearson *B* Similarity Index (*B*). Brock (1977) argues that the Pinkham Pearson *B* similarity index (Pinkham and Pearson 1976) is too sensitive with rare species, not sensitive enough with dominant forms, and may be subject to bias or sampling error. Other studies consider the sensitivity of the *B* similarity index to be an asset. Boyle et al. (1990) reported the *B* similarity index was preferred over the Jaccard Coefficient for detecting impairment. Barbour et al. (1992) recommends the *B* similarity index as a community similarity metric. Lillie and Isenring (1996) found the *B* similarity index useful when comparing EPTG community structure among streams in the region of Baraboo Hills, WI. Note, *CS* and *B* similarity (Pinkham and Pearson 1976, Pearson and Pinkham 1992) can be calculated using the software BIOSIM1 (Gonzales et al. 1993).

Stander's Similarity Index (SIMI). This index is based on the relative proportion of individuals in a community. Values range from 0 (no taxa in common) to 1 (the relative abundance of all taxa are equal at both sites) (Stander 1970). The interpretation of SIMI values, and assigning categorical ranks to the degree of similarity observed (i.e., low, medium, high) is difficult because the SIMI is a correlation measurement (Johnson and Millie 1982). Based on this fact SIMI is best used in making general comparisons between two or more sites with a common reference site.

Various ranking scales have been proposed for the SIMI (Rohr 1977, Tuchman and Blinn 1979). Johnson and Millie (1982) recommend the

Lepidoptera larva.



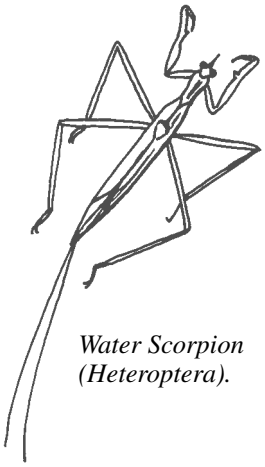
Lepidoptera (Aquatic Caterpillars):

Nine species and six genera in the family Pyralidae are the taxa of moths found in Wisconsin that have aquatic larvae. The larval stages inhabit rooted aquatic macrophytes and feed by mining the plant stems or by feeding on the attached algae. Adults are nondescript moths that hold their wings "tent-like" over their bodies.

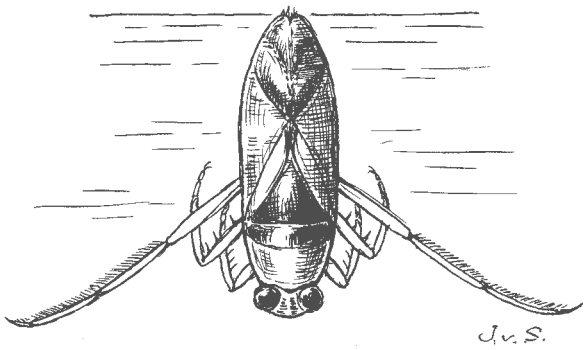
most appropriate application of SIMI is based on calculating confidence intervals using replication and computer simulation. However, Smith et al. (1986) suggest that the jackknife or bootstrap methods are better for estimating confidence intervals.

Czarnecki (1979) believed that a weakness of the SIMI is in that it is more sensitive to changes in dominant taxa than in the rare or uncommon taxa. But Johnson and Millie (1982) suggest that the occurrence of rare taxa in low abundance would not have a great effect on the overall structure of the community. For additional insight regarding the ecological significance and importance of intolerant but rare taxa please see Fausch (1990).

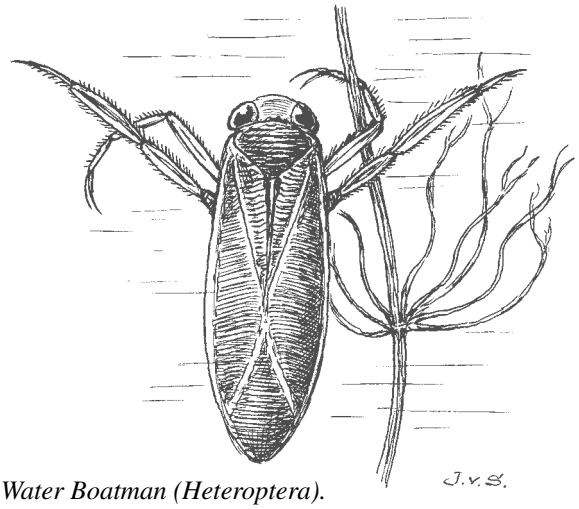
The SIMI has been found to be relatively insensitive to low and moderate changes in community structure (Boyle et al.1990). Despite the fact that the SIMI has probably been used more frequently (particularly in algae studies) than any other single similarity index, there is considerable confusion and lack of accord in the literature with respect to the interpretation of SIMI values. Most investigators agree that more than one similarity index should be used when making community comparisons.



Water Scorpion (Heteroptera).



Backswimmer (Heteroptera).



Water Boatman (Heteroptera).

Percentage Similarity (*PS*). The *PS* is based on the comparison of percentages of organisms at the genus level in two communities (Whittaker 1952). Values for this index can range from 0 (no taxa in common) to 1 (all taxa in common and in same percentages). The Vermont Department of Environmental Conservation has developed guidelines for interpreting *PS* values: $PS > 0.75$ represents no significant biological alterations (typical for comparisons between replicates), *PS* 0.25 – 0.75 indicates a probable change has occurred, and $PS < 0.25$ represents a significant change has occurred (S. L. Fisk, Ohio EPA, pers. comm. 1987).

There has been much debate on the value of *PS* as similarity index. Whittaker (1952) reports that the *PS* was inaccurate when the relative proportions of the taxa were equal but the overall abundance was great. However, Brock (1977) points out that this inaccuracy may not be important in pollution studies where severe effects usually change dominance; if dominance shifts, and the taxa are in balance between the two communities, then they may be functioning the same. Boyle et al. (1990) found that the *PS* was less sensitive to changes in community structure than Jaccard's Index. Additionally, Brock (1977) found that for separating structural and functional differences between communities the *PS* might be better than the *B* similarity index. Cao et al. (1997a) demonstrated that *PS* (along with several other similarity measures) are sensitive to sample size. He argues that because *PS* is more sensitive to sample size, it should therefore be

more sensitive to community changes and thus a good similarity measure. In a companion paper Cao et al. (1997b) berates *PS* for overweighing abundant species and ignoring 'sample-specific' species. Apparently some people make a living at this sort of thing!

Ecological Distance (EDIS). EDIS measurements have largely been used by terrestrial ecologists to measure dissimilarity (Clark 1952, Rhodes et al. 1969). The EDIS is based on the number of different genera (or species) in a set of paired samples and the differences in abundance of each taxa between the samples. Values can range from 0 (least dissimilar = identical) to 1 (greatest dissimilarity = no genera or species in common). The similarity between sites is estimated by subtracting the EDIS value from 1. The assignment of discrete values or ranges of values representing different degrees of dissimilarity has been arbitrary and there is no general consensus in the literature of what these values or ranges should be. Values also may vary widely depending on the design of studies and the specific organisms used in the analysis of community structure.

Recommendations

Macroinvertebrate data can be used in a variety of ways for making bioassessments. The method of analysis differs based on the project objectives (i.e., are the data to be used for baseline monitoring, trends analysis, or detecting unknown sources of pollution?). Whatever the case, one first must define a question. This determines a path to follow, allows determination of data collection issues (i.e., sampling or monitoring plan or design), and helps focus on the metrics that should be examined. Evaluations may be inconclusive using existing data; it is ok to design a new sampling strategy tailored to answer a specific question.

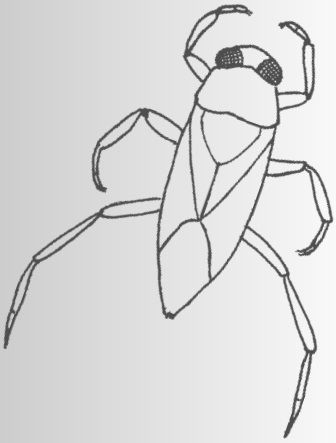
For additional recommendations, there is material available at the US EPA Rapid Bioassessment Protocols website. **http://www.epa.gov/owow/monitoring/rbp/**

Macroinvertebrate data, as with all biological data, exhibit a considerable amount of natural variability both temporally (i.e., seasonal) and spatially (i.e., erosional versus depositional areas). However, understanding what an increase or decrease in a metric means and which direction indicates a condition of improvement or deterioration is vital. It is important to recognize an impact when one occurs and one should have an idea of how much change must occur in order to represent a significant impact (keeping in mind that some changes are natural and may not be related to human impacts). If the sampling design is carefully planned to answer a question, the results will be able to separate and discriminate between human induced impacts and background variation or noise.

Objectives

The first step in making a bioassessment is to define the objectives. It is important to clearly define what do you want to know or examine. For example, you are interested in measuring the biological condition at a particular

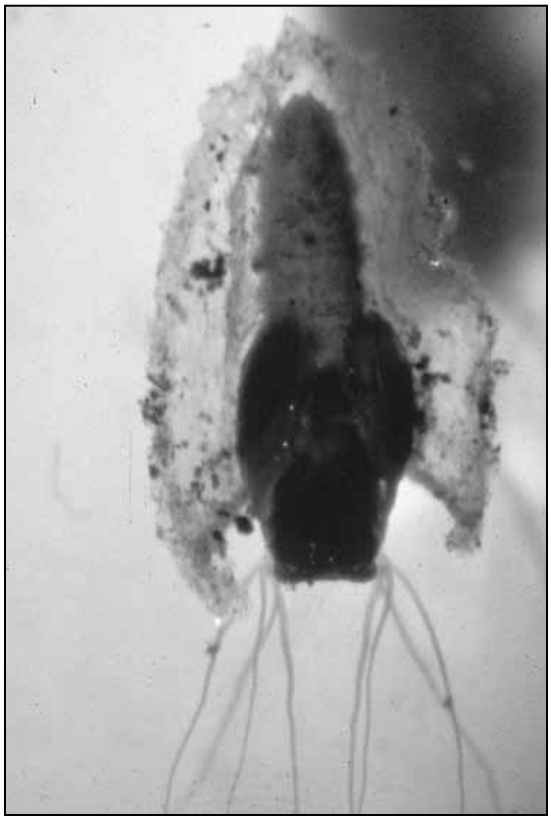
Heteroptera adult.



Heteroptera (Aquatic and Semi-Aquatic Bugs):

Heteroptera is primarily a terrestrial order. Presently 67 aquatic and 26 semi-aquatic species in 30 genera and 11 families have been identified from Wisconsin. The adults of Heteroptera can be distinguished from other aquatic insects by their sucking mouthparts that are formed into a broad tube or rostrum, except Corixidae. Adults of all Heteroptera found in Wisconsin can be identified to species. Almost all are predators feeding on invertebrates and occasionally on small vertebrates such as tadpoles and minnows.

Top: Blackfly larvae (family Simulidae).
Bottom: Pupating blackfly (family Simulidae).



site on a stream and have reason to believe (or are certain) that some form of impact has occurred at or above the sampling site. You would like to determine whether that impact has had a significant effect on the macroinvertebrate community at that site.

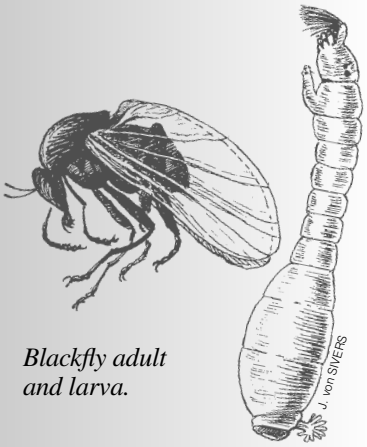
If you have access to historical data recorded at the sites of interest, it is possible to attempt a Before-After, Control-Impact (BACI) sampling design[7]. You could then collect a new set of samples (preferably three or more replicates) from the site and compare the results with the historical data using any of the various statistical procedures. However, this simple method of before-after comparison may allow you to detect a difference or change but the comparison may not necessarily indicate a direct cause-effect relationship between the change and the suspected factor. Without sampling other sites in the watershed you cannot be sure that some other factor (i.e., drought or flooding) may have caused the significant change in the macroinvertebrate community at your sampling sites. Additionally, the differences seen between the new set of data and the historical data may be the result of slight differences in emergence patterns between the two data sets (i.e., climatic noise). If you are in the position of gathering new data it may be beneficial to sample comparable sites in adjoining basins or upstream of the suspected point of impact. These sites also should have historical data available along the same time scale as at the 'impacted' sites.

In other circumstances, historical data do not exist and you may be sampling a site for the first time. In this case the best approach is to compare the new data (a single sample may be adequate, but always take replicates if budget and time allow) with existing data representing least-disturbed *reference sites* of a similar stream order, ecoregion, watershed (Omernik et al. 2000), or Relatively Homogeneous Units (RHU) (Rheaume et al. 1996, Robertson and Saad 1996). An analysis of the FBI value may be a good start. For comparisons, it is important to use data collected during the same season**,** within the same habitat, and using the same sampling and laboratory protocols. While this type of analysis can not determine if or when a change has occurred, it can quantify the relative difference between the macroinvertebrate community at the site in question and the RHU. The reference site could be a 'least-disturbed' site or even represent an upstream (possibly unimpacted?) or downstream (recovery?) site. Careful examination of the differences in community structure among sites may help form a hypothesis as to what caused the differences. Keep in mind that some differences among sampling sites in community structure may result from subtle differences in substrate (particle size, texture, composition, algae, macrophytes), flow (velocity, depth), temperature (springs, shading), or channel position (head versus bottom of riffle or snag).

Microhabitat Considerations

Most lotic macroinvertebrate populations exhibit a contagious or clumped distribution pattern in a stream matrix. Because of this, most macroinvertebrate data exhibit a non-normal distribution pattern and require the use of non-parametric statistics. Macroinvertebrate population data can vary depending on where and when samples are taken. Similar microhabitats must be sampled both temporally and spatially to insure comparable data. In some cases streams do not have suitable riffle substrates to sample (i.e. many Wisconsin streams in the central sands area). In these situations it is acceptable to sample snag habitat in place of riffle habitat.

In a recent study of central Wisconsin trout streams that included comparisons of 82 sets of 5 replicate samples (total of 410 samples), snag samples exhibited greater variability than riffle samples using the HBI, 5-25 Max HBI, FBI, and MTV (S. Szczytko, UWSP, unpublished data). The yearly mean HBI value from snag samples was always higher and at least one water quality classification poorer than the THBI determined from riffle samples. Annual mean values of 5-25 Max HBI, FBI and MTV were also higher using snag samples than riffle samples (S. Szczytko, UW Stevens Point, unpublished data). These results indicate that if you are relegated to sample snag habitats instead of the recommended riffles, the resulting metric values are likely to be higher than the THBI.



Blackfly adult and larva.

Choosing the Appropriate Metrics

There are a number of choices available to analyze the metric data. Depending on the quantity, quality, distribution characteristics of the data, and the objectives any number of statistical tools can be applied. These tools may range from simple graphic analysis techniques (i.e., univariate plots or bivariate regression plots) to more complex procedures (i.e., ANOVA, t-tests, chi-square, etc.) and a wide range of other parametric and non-parametric analyses.

The various similarity (or dissimilarity) indices are particularly useful for making relative comparisons among streams or when searching for trends in water quality at a particular site. Each metric comes with its own set of strengths and weaknesses and each may be more sensitive to different forms of impacts. Table 5 provides a general overview of which metrics are useful for selected impacts. To select what statistical analysis to use you should be aware of the data assumptions that affect the correct application (i.e., data normally distributed, equal variance, etc.) and be prepared to make the necessary data transformations if required. When transformations fail to 'normalize' the data you may need to use non-parametric statistics. If you are in doubt, please consult the computer software help advisor or speak to a Department statistician[8].

Blackfly adult (family Simulidae).

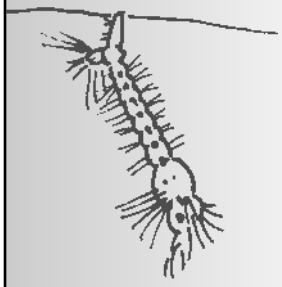
[7] When using historical data it is best to: determine who collected the data, what type of data they collected, how they collected the data, where exactly in the stream reach the samples were collected, and why the data were collected in the first place.

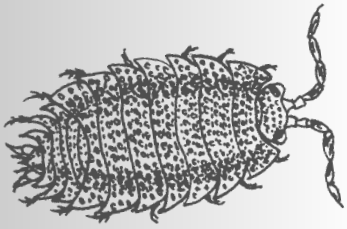
[8] See also: NABS (1993) workshop on *Use of Biostatistics in Benthic Ecological Studies*; Fore et al. (1994). Statistical properties of the IBI are discussed including inherent bias related to the way metric values are scored.

Table 5. *Anticipated metric response to stressors. n.r.= not recommended*

METRIC	NAME or DESCRIPTION	STRESSOR	STRESSOR
		Organics, Nutrients, Low DO	**Toxics, Contaminants, Heavy Metals**
Biotic Indices			
HBI	Hilsenhoff Biotic Index	Increases	n.r.
FBI	Family Biotic Index	Increases	n.r.
MTV	Mean Tolerance Value	Increases	n.r.
MAX-10	Max-10 HBI	Increases	n.r.
Taxa Richness			
SR	Total Species Richness	Decreases	Decreases
GR	Generic Richness	Decreases	Decreases
EPTG	Ephemeroptera-Plecoptera-Trichoptera Generic Richness	Decreases	Decreases
Diversity			
d	Margalef's Diversity Index	Increase or Decrease	Decreases
Trophic Function			
Percent Scrapers	Percent of total represented by Scrapers	Decreases	Decreases
Percent Filterers	Percent of total represented by Filterers	?	Decreases?
Percent Shredders	Percent of total represented by Shredders	Decreases	Decreases
Percent Collectors	Percent of total represented by Collectors	Increases	Increases
Percent Scrapers/ percent Filterers	Ratio percent Scrapers to percent Filterers	n.r.	n.r.
Percent Scrapers/ percent Gatherers	Ratio percent Scrapers to percent Gatherers	n.r.	n.r.
Percent Scrapers/ percent Collectors	Ratio percent Scrapers to percent Collectors	n.r.	n.r.
Dominance			
Dominant Families	Percent of total count represented by top five families	Response at extremes	Some chironomids tolerant to metals
Dominant Genera	Percent of total count represented by top five genera	Response at extremes	
Dominant Species	Percent of total count represented by top five species	Response at extremes	
Comparison Metrics			
CCL	Coefficient of Community Loss	Increases	Increases
CS	Coefficient of Similarity	Decreases	Decreases
SIMI	Stander's Similarity Index	Decreases	Decreases
PS	Percentage Similarity	Decreases	Decreases
B	B Similarity Index	Decreases	Decreases
EDIS	Ecological Distance	Increases	Increases

STRESSOR	STRESSOR	STRESSOR
Inorganics, Sediments	**Flow Disruption**	**Thermal**
n.r.	n.r.	n.r.
n.r.	n.r.	n.r.
n.r.	n.r.	n.r.
n.r.	n.r.	n.r.
?	Decreases	Decreases
?	Decreases	Decreases
?	Decreases	Decreases
Increase or Decrease	Increase or Decrease	Decreases
Decreases	?	Increases if periphyton production enhanced?
Increases?	?	?
Decreases	?	?
Increases	?	?
n.r.	n.r.	n.r.
n.r.	n.r.	n.r.
n.r.	n.r.	n.r.
Loss of many filterers, scrapers, shredders with increased imbeddedness.	Favors mobile forms over sessile forms above line of low water levels.	Interference of life cycles difficult to predict, but some insects may be eliminated while others fill vacated niches (try before/after examinations).
Increases	Increases	Increases
Decreases	Decreases	Decreases
Decreases	Decreases	Decreases
Decreases	Decreases	Decreases
Decreases	Decreases	Decreases
Increases	Increases	Increases





We can compare the percent change (before-after) or differences (control-impact) using a variety of methods. These include upstream-downstream site analysis, treatment versus non-treatment areas; or, using comparisons with 'reference' streams that represent undisturbed, least-disturbed, or 'typical' conditions. Statistical significance of percent change may be accomplished through repeated measurements (i.e., replication among sites or among similar stations) and the subsequent determination of change associated with varying levels of disturbance. Qualitative ratings (i.e., no impact, moderate impact, severe impact) can be assigned only after extensive sampling and analysis. If categorical ratings have been assigned to the metric we can assign a water quality rating score based on the data (i.e., good, fair, poor, etc.) and examine changes or differences in ratings between pairs of data or multiple observations. Comparisons to reference streams (Lenat and Barbour 1994, Hughes 1995, Bailey 1995, Reynoldson et al. 1997) may take into account a variety of factors that are known to influence community composition. These factors may include: ecoregions, habitat types, stream order, longitudinal position, coldwater, warmwater, and many others. Regardless of the methods chosen, you should always identify and account for the expected seasonal influences on the involved metrics.

Future Directions

Do not overlook new developments in the field of macroinvertebrate community analysis. The current literature is exploding with articles incorporating new and innovative approaches to interpret macroinvertebrate data. The following recommendations refer only to typical kick-net sampling assessments. Other macroinvertebrate assessment techniques come with their own set of metrics and recommendations. For example, in the assessment of sediment contaminated sites, oligochaetes and chironomids become more important and separate indices have been formulated for sediment sample analysis (Krieger and Ross 1993, Reynoldson et al. 1995, Hoke et al. 1995, Canfield et al. 1996). In addition, sampling methods, laboratory analysis, and data interpretation of sediment samples warrant a different set of recommendations (Aartila 1996, North American Benthological Society 1995). The following summaries present only a few of the various approaches that are being used.

Faunal Typology. To describe ecosystem function Castella and Amoros (1983) used multivariate analysis to illustrate the faunal affinities of 26 macroinvertebrate communities in the Rhone River. Cluster analysis using different sets of aquatic orders, aquatic vegetation, sediment, and water quality represent an innovative approach to search for faunal descriptors and group associations.

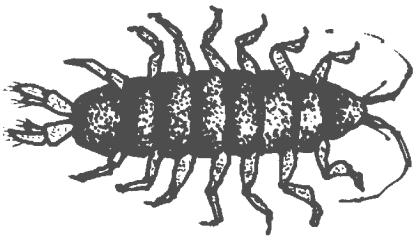
Bioequivalence. McDonald and Erickson (1994) present a novel approach to determine whether two sites are 'bioequivalent'. The authors discuss an alternative procedure to apply in cases where use of the classical null and alternative hypotheses is inappropriate. Although too lengthy to describe here, the method is founded in the medical field (i.e., drug-testing) and may be used to assign confidence limits for regulatory purposes.

Percent Modal Affinity. Novak and Bode (1992) developed the Percent Model Affinity (PMA) index to compare the percent similarity (Whittaker and Fairbanks 1958) of macroinvertebrate communities in selected streams to typical communities representative of unpolluted sites (reference sites). The degree of impact was judged by the ranges of similarity to the 'model' community. Barton (1996) found the PMA useful in detecting land use impacts on 213 Ontario streams. The sensitivity of the PMA index increases with the level of taxonomic resolution used (Barton 1996).

Other Dominance Indices. Beisel et al. (1996) compares the performance of four dominance indices: the Simpson Index, McIntosh dominance index, Berger-Parker index, and the Camargo dominance measure. They favor the McIntosh dominance index for its ability to detect small differences in the abundant taxa in communities and its relative insensitivity to rare taxa. However, Camargo (1997) refutes Beisel's conclusions and provides evidence to suggest that the Camargo index is the best dominance and diversity metric available.

Other Biological Indices. Cairns et al. (1968) present the Sequential Comparison Index (SCI) (Resh and Price 1984). Death (1996) discusses species abundance patterns as indicative of habitat stability. A number of new indices have been proposed based on the relationship between morphological deformities in

Isopod or aquatic sowbug.



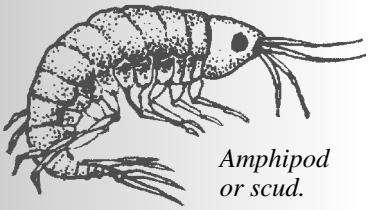
aquatic larvae and various contaminants (Warwick and Tisdale 1988, Vermeulen 1995). A number of deformities in Odonate larvae have been discovered in Wisconsin streams (B. Smith, Wisconsin DNR, pers. comm. 1998). Done and Reichelt (1998) suggest an innovative approach to evaluate the effectiveness of management decisions using four new indices based on biomass and trophic structure, endangered species, representativeness or uniqueness of species composition, and successional stage. They introduce the concept of 'desired' and 'degraded' states and discuss disturbance regimes in reference to management goals. Although the paper is directed at fisheries management it is clearly applicable to any resource.

Multimetric Indices. The development of multimetric indices for use in evaluating the biological integrity of freshwater ecosystems is reviewed extensively by Simon and Lyons (1995). Multimetric indices offer an advantage over individual metrics in that they integrate effects of multiple stressors (Barbour et al. 1995). Karr (1981) presented the first Index of Biotic Integrity (IBI) based on fish community attributes in Illinois and Indiana streams. Since its introduction, the IBI and its many modifications have become very popular and incorporated into many state and agency management protocols. Currently multimetric indices have been developed or proposed for: Ohio (DeShon 1995), Florida (Barbour et al. 1996), the Tennessee Valley (Kerans and Karr 1994), Oregon (Fore et al. 1996), the Chesapeake Bay (Weisberg et al. 1997), Washington (Kleidal 1995), Arkansas (Shackleford 1988), Michigan (Grant and Thorpe 1993), Texas (Twidwell and Davis 1989), and several developing countries (Thorne and Williams 1997). La Liberte (1996) developed a macroinvertebrate multimetric for use in Wisconsin using data from 14 West Central Region counties with restricted application in the West Central Region. The macroinvertebrate multimetric is composed of six metrics, some adjusted to compensate for effects of stream width (La Liberte 1996).

Other Multivariate Methods. An increasing number of studies are using multivariate analyses (ordinations, canonical correspondence analysis) to examine macroinvertebrate communities (Culp and Davies 1980, Cortes 1992, Jackson 1993). Reynoldson et al. (1997) compared the performance characteristics of selected multi*variate* methods with selected multi*metric* methods. They concluded that multivariate approaches had higher precision and accuracy than the multimetric approaches. Fore et al. (1996) found that a multimetric index performed better than a principal components analysis in detecting impacts of disturbance on Oregon streams. Other studies use a combination of both methods to make water quality assessments (Zamora-Munoz and Alba-Tercedor 1996). It is evident that more studies are applying multivariate procedures to understand and explain macroinvertebrate community structure.

Isopoda (Aquatic Sowbugs):

Members of the class Crustacea (including isopods, amphipods, mysids, and decapods) are very important components of the invertebrate community found in both lentic and lotic habitats in Wisconsin. Aquatic sowbugs are a significant group of crustaceans occurring throughout North America. Of the four families, Asellidae is the most important in terms of taxa numbers. Asellus (= caecidotea; Jass and Klausmeier 1997) commonly are omnivore-detritivores found living in leaf detritus. Some taxa are relatively tolerant of organic pollution, whereas others are relegated to springheads.

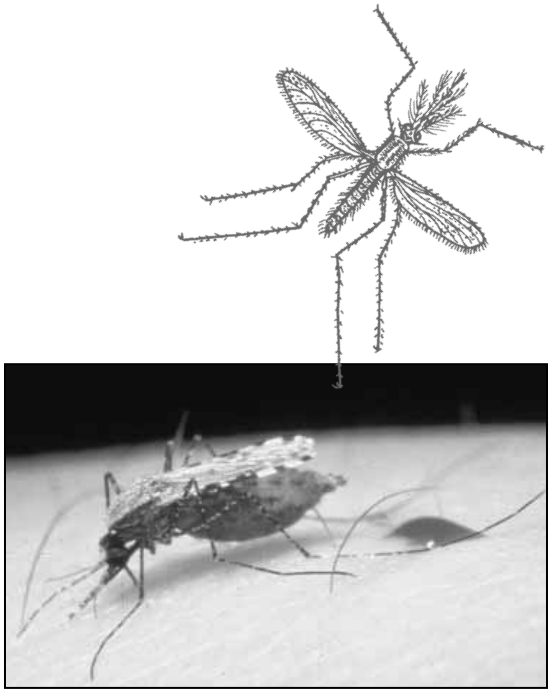


Amphipod or scud.

Amphipoda (Aquatic Amphipods):

Approximately 90 species found in 3 families and 8 genera occur in North America. The families Gamaridae and Talitridae are common in Wisconsin. Scuds are benthic, primarily occur in shallow waters, feed upon detritus, and in-turn are important prey of a number of fish species. Scuds are laterally compressed and often swim on their sides, hence the common name sideswimmers.

Predictive Models. An alternative approach to using multimetric indices is predictive modeling. Developed primarily in England and Australia, predictive modeling compares measured and predicted communities based on selected environmental attributes (Winget and Magnum 1979, Wright et al. 1984, Armitage et al. 1987, Ormerod and Edwards 1987, Wright 1995, Norris 1995). The degree of impact can be measured by comparing predicted assemblages in the absence of impact with observed communities. Investigators have used different taxonomic levels from species to the family level with generally good success. Predictive models in England helped create the River Invertebrate Prediction and Classification System (RIVPACS). This system, which has been tested successfully in a number of European countries, is based on the application of TWINSPAN computer technology. For information on the future of this application in the United States see the EPA web site at **http://www.epa.gov/OWOW/monitoring/techmon.html**.



Mosquito (family Culicidae).

Species Trait Modeling. Many studies include elaborate examinations of species traits characteristics which appear to show exciting possibilities for the future (Richoux 1994, Cellot et al. 1994, Tachet et al. 1994, Townsend and Hildrew 1994, Usseglio-Polatera 1994, Townsend et al. 1997, Poff 1997, and Richards et al. 1997). This approach is based on the premise that habitat, in the form of both abiotic and biotic features, forms a templet from which aquatic insect species traits (i.e., age, life history characteristics, body form, etc.) evolve. As a result, this templet may predict what type of species may occur in any particular habitat. This approach is often used in combination with 'fuzzy coding' procedures (Chevenet et al. 1994, Roberts 1986) to graphically present relationships between species traits and environmental variables.

Landscape Classification In Macroinvertebrate Bioassessments. The ability to predict and classify macroinvertebrate community composition is of interest to water resources managers. The ability to detect human impacts often depends on our knowledge of what the biota was like prior to human influence. To assist in this effort, various ecological frameworks have been developed to characterize the biological properties of various geographic regions (i.e., ecoregions or provinces) (Omernik 1987, Bailey 1995). A recent volume of the *Journal of the North American Benthological Society* (Vol. 19, No. 3, September 2000) is devoted to this topic and should be reviewed in its entirety.

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Dobsonfly larva and adult.

Appendix A. Selected Literature Review[9]

General Reviews

Boyle et al. 1990. Diversity and similarity indices.
Cairns 1977. Biological integrity; diversity indices; functional integrity.
Cairns and Pratt 1993. Historical perspective of biological monitoring.
Cummins 1973. Early paper on functional feeding classes.
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Guhl 1987. Biotic, diversity, and similarity indices in aquatic ecosystems.
Hughes 1995. Review on use of reference stations for biological assessments.
Karr 1991. Review of IBI use in water resource management.
Lenat and Barbour 1994. Macroinvertebrates in rapid bioassessments.
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Norris and Georges 1993. Interpretation of invertebrate data.
Noss 1990. Biodiversity; definitions, indicators, and scales.
Resh and Jackson 1993. Review of rapid bioassessments; macroinvertebrates.
Resh and McElravy 1993. Contemporary quantitative biomonitoring; review of lotic and lentic studies.
Resh et al. 1995. Rapid bioassessment metrics.
Rosenberg and Resh 1993. Biomonitoring; statistical considerations (pp. 252-253).
Somers et al. 1998. Lakes; ordination, metrics, sample sizes.
Steinhart et al. 1981. Water quality; biological and related indices.
Szczytko 1988. Interpretative techniques in Wisconsin bioassessments.
Wallace and Webster 196. Review of macroinvertebrate functional groups.
Winget and Mangum 1979. Biotic Condition Index (USFS); integrates aspects of habitat, water quality, and macroinvertebrate community.
Washington 1984. Diversity, biotic, and similarity indices.

Evaluations of Rapid Bioassessment Protocols

Barbour and Gerritsen 1996. Subsampling; defense of fixed count method.
Barbour et al. 1992. Ecoregion study in Oregon, Kentucky, Colorado; test of eleven metrics.
Courtemanch 1996. Subsampling; effect on richness metrics.
Diamond et al. 1996. Comparisons CVs, discriminating power, ecoregions.
Gaunt and Thorpe 1993. Non-wadable streams in Michigan; test of six metrics.
Growns et al. 1997. Effect of subsampling, family level, SIGNAL biotic index, EPT, taxa richness, discriminatory power.
Hannaford and Resh 1995. California; habitat, metric evaluations.
Plafkin et al. 1989. Evaluation of rapid bioassessment protocols.
Rabeni et al. 1999. Metric sensitivity to sample size; similarity comparisons.

[9] Copies of all references listed in the selected literature review are available upon request from the DNR library. It is anticipated that this section of the guidance document will be updated periodically in an attempt to incorporate new and relevant literature on specific issues important to the Department. Any assistance with this effort will be greatly appreciated. We hope to make this reference collection searchable on the web and to create a more thorough index of the content of the included articles. Please note that this list of references is by no way complete, but is intended to serve as a starting point to find specific references that may support you in your work.

Resh 1994. Evaluation of richness, diversity, biotic indices, functional metrics, taxonomic resolution.
Resh 1995. Good review article on design issues.
Vinson and Hawkins 1996. Effect of sampling area and subsampling on taxa richness metrics.

Biotic Indices

Cao et al. 1997a-c. Modified Chandler; ASPT Index, Great Britain.
Chandler 1970. Precursor to HBI in Britain.
Chessman 1995. Family level 'SIGNAL' rapid bioassessments in Australia.
Chessman and McEvoy 1998. Three disturbance sensitivity indices; family level.
Chessman et al. 1997. Australia; 'SIGNAL' biotic index, family level, average score per family index.
DeShon 1995 (also Ohio EPA 1987 and 1988). Invertebrate Community Index, Ohio.
Grotheer et al. 1994. Colorado system of "biologically determined values" based on environmental preferences of taxa present.
Hilsenhoff 1977. Original HBI publication.
Hilsenhoff 1982. Further HBI advancements with taxonomic keys.
Hilsenhoff 1987. Revised Biotic Index.
Hilsenhoff 1988a. Family level biotic index.
Hilsenhoff 1988b. Seasonal corrections to biotic index.
Hilsenhoff 1998. Further modifications to HBI to account for seasonality; MAX-10 HBI.
Illinois EPA 1988. Modified HBI; biotic resources, Pecatonica River study.
Lang and Reymond 1995. RIVAUD 95 Index, Switzerland.
Lenat 1993. Modified HBI for use in North Carolina.
Lenat and Penrose 1996. Discussion of history of EPT metric.
Lillie and Schlesser 1994 Mean tolerance value biotic index.
McDonald et al. 1991. Use of Stream Quality Monitoring (SQM) in Illinois and Ohio.
Murphy 1978. Temporal variability in six BIs and diversity indices, Wales.
Narf et al. 1984. Statistical procedures; HBIs in Wisconsin (out of date).
Novak and Bode 1992. Percent Model Affinity Index, New York.
Shackleford 1988. Arkansas; Rapid bioassessments; Indicator Assemblage Index.
Stark 1993. MCI; mean tolerance value, New Zealand.
Winget and Mangum 1979. Biotic Condition Index for US Forest Service. Integrates aspects of habitat, water quality, and macroinvertebrates.
Wright 1995. United Kingdom RIVPACS. Review of the development of prediction and classification of invertebrates based on selected environmental attributes.
Wright et al. 1988. Family level; environmental data, RIVPACS predictive modeling.

New Diversity Indices

Camargo 1992. Species richness, species evenness diversity, community structure.
Camargo 1997. Dominance Index, importance of rare taxa.
Docampo and de Bikuna 1994. Complex paper dealing with development and testing of yet another diversity index.
Small et al. 1996. Macrophyte based diversity index for stream water quality assessment.

Taxa/Species Richness

Cao et al. 1998. Importance of rare species in bioassessments, species richness.
Clements 1991. Species richness better than abundance (except heavy metals).
Death and Winterbourn 1995. Species richness and evenness influenced by disturbance; habitat stability.
Douglas and Lake 1994. Stone sizes and areas; species richness, indicators.
Fausch et al. 1990. Fish communities, but discussion applies to inverts.
Gray 1981. Log normal distribution individuals among species.
Kerans et al. 1992. Influence of sampling protocol taxa richness; other metrics; pool versus riffles.
Larsen and Herlihy 1998. Richness; area versus fixed count.
Lenat 1988. Multihabitat sampling; EPT, taxa richness by ecoregions.
Lenz and Miller 1996. Wisconsin; Invertebrate comparisons using different collection methods.

Minshall et al. 1985. Seasonality; stream size.
Palomaki and Kosenniemi 1993. Effects of freezing (bottom) on taxa richness.
Palomaki and Paasivirta 1993. Species richness influenced by area and sampling sizes in Finland.
Rae 1990. Species richness; depth and discharge.
Siegel and Gegrman 1982. Rarefaction, taxonomic diversity and richness.
Thorp 1992. Ohio River. Influence of plants and location on metrics.
Townsend et al. 1997. Intermediate disturbance hypothesis; taxonomic richness.
Vinson and Hawkins 1996. Effect of sample area and subsampling on taxa richness.

Indicator Species

Fausch et al. 1990. Fish, but good discussion of indicator taxa.
Johnson et al. 1993. Review article
Muck and Newman 1992. Amphipods as indicators of land use in Minnesota.
Nelson and Roline 1993. Mayflies and metal pollution in Arkansas River.
Patrick and Palavage 1994. General review species as indicators.
Ribera and Foster 1992. Beetles as indicators.

Specific Effects: Sediment/Suspended Sediment

Angradi 1999. Fine sediment, metrics, ordinations.
Angradi 1999. Forestry management and sediments, macroinvertebrate metric response.
Armitage 1989. Substrate; community associations, predictive RIVPACS system used in Great Britain.
Farnsworth et al. 1979. EPA review article on sediments and nutrients.
Kohlhepp and Hellenthal 1992. Sediment deposition, effects on insects, functional feeding classes in Indiana streams.
Krieger and Ross 1993. Great Lakes sediments; changes in oligochaetes and chironomids
Lemly 1982. Sedimentation, interstitial spaces, filter feeders.
Luedtke and Brusven 1976. Sand; stream insects, drift and colonization issues in Idaho.
Newcombe and MacDonald 1991. Effects of sediments on fish and invertebrates.
Quinn and Hickey 1990b. Substrate index, invertebrates, floods, land use.
Reynoldson et al. 1995. BEAST index, multivariate tests, invertebrates, sediments.
Roback 1978. Chironomids and Mayflies; sediments and thermal effects.
Schrank 1982. Wisconsin review article on the effects of erosional sediment on streams.
Williams 1980. Substrate heterogeneity, invertebrate associations.

Specific Effects: Physical Disturbance Including Flows, Floods, and Drought

Anderson 1992. Species richness and diversity by age of forested watershed.
Boulton et al. 1992. Effect of spates on invertebrate assemblage composition.
Chessman and Robinson 1987. Effect of drought on macroinvertebrate richness; wastewater.
Cobb et al. 1992. Discharge and substrate stability; stream aquatic insects in Canada.
Death 1996. Habitat stability, shape of distribution curve.
Death and Winterbourn 1995. Habitat stability, invertebrate richness in New Zealand.
Doeg et al. 1989. Australian study; invertebrate richness and densities; recovery after disturbance.
Duchrow 1983. Study done in the Ozarks; stonefly and mayfly response to disturbance.
Gurtz and Wallace 1984. Watershed disturbance, densities of aquatic insects.
Hendricks et al. 1995. Impact of flooding on invertebrate densities.
Mackay 1992. Colonization patterns.
Matthaei and Townsend 2000. Effects of disturbance on stream invertebrates in New Zealand.
Quinn and Hickey 1990a, 1990b. Flooding and substrate size; impact on species richness in New Zealand.
Palomaki and Koskenniemi 1993. Impact of freezing on invertebrates.
Poff and Ward 1989. Impacts of stream flow variability or stability on community structure.
Rader 1997. Species traits for aquatic insects; drift, availability to salmonids.
Rae 1990. Effect of discharge on species richness.
Reice 1991. Impact of spates on community structure in North Carolina.

Richards and Minshall 1992. Impact of fire in watershed on macroinvertebrates, PCA analysis.

Rosillon 1989. Belgium streams; yearly changes in invertebrates related to floods.

Townsend et al. 1997. New Zealand study; stream bed disturbance, intermediate disturbance hypothesis, taxa richness.

Specific Effects: Land Use

Berkman et al. 1986. Agricultural impacts on fish and invertebrates.

Cingolani and Morosi 1992. Landfill; impacts on invertebrate community.

Collins and Pess 1997a and 1997b. Forestry practices, riparian buffers, large woody debris. Not an invertebrate paper, but useful for management issues.

Corkum 1990. Invertebrate distributions and assemblages as influenced by land use.

Fore et al. 1996. Multimetric evaluation of road building and timber harvest.

Hachmoller et al. 1991. Invertebrate response to riparian alterations in Washington.

Johnson and Gage 1997. Landscape approaches using GIS technology; review of multivariate techniques used in watershed analysis.

Jones and Clark 1987.Urbanization, stream insect communities, PCA analysis.

Lenat 1984. North Carolinan study dealing with agricultural runoff, taxa richness, EPT metric response.

Lenat and Crawford 1994. Land use; effects on taxa richness and biotic index in North Carolina.

Matthews et al. 1991. Changes in community structure with land use, correspondence analysis.

May et al. 1997. Urbanization, riparian buffers, LWD, and biotic integrity.

Muck and Newman 1992. Use of amphipods to detect impacts of land use on water quality.

Pedersen and Perkins 1986. Impact of urban runoff on invertebrates.

Richards and Host 1994.GIS study of relation between land use and macroinvertebrates.

Richards et al. 1993. Michigan studies; multivariate analysis CCA ordination; EPT.

Richards et al. 1997. GIS catchment scales, geology, species traits of insects.

Sweeney 1993. Impacts of riparian vegetation on macroinvertebrate production.

Trimble 1997. Classic Coon Creek study in Wisconsin, riparian land use, grazing; physical effects (no invertebrates).

Wang et al. 1997. Wisconsin land use, habitat quality, and fish IBI.

Wang et al. 2000. Watershed BMPs, macroinvertebrate response.

Weigel et al. 1995. Biotic index response to canopy cover in Wisconsin.

Weigel et al. 2000. Wisconsin riparian buffers, land use, insect metric performance.

Specific Effects: Herbivores/Grazing

Gilliam et al. 1989. Fish interactions, invertebrate distributions and community structure.

Hart and Robinson 1990. Periphyton, phosphorus enrichment and grazing, caddisflies.

Jacoby 1987. Algae and diatoms, Cascade stream, caddisflies.

Peterson and Tuchman 1994. Substrates, associations with microbiota; a review article.

Peterson et al. 1993. Fertilization; responses in a tundra river.

Stewart 1987. Fish grazing and algae, fertilization.

Specific Effects: Gasoline and Road Runoff

Pontasch and Brusven 1988. Macroinvertebrate comparisons using 7 similarity indices in Idaho.

Smith and Kaster 1983. Wisconsin highway runoff impacts on invertebrate abundance and biomass.

Specific Effects: Climate

Eyre et al. 1993. Effect on beetles in Great Britain, climate index.

Hann et al. 1992. Climate effects on aquatic insects; debate article.

Sweeney et al. 1991. Climate change effects on aquatic insect distribution patterns.

Walker et al. 1991. Climate, distribution of aquatic insects, chironomids.

Walker et al. 1992. Further discussion on climate effects; debate article.

Specific Effects: Chlorine/Chlorides

Camargo 1991. Effects on Hydropsychids.

Newman et al. 1987. Chronic chlorine exposure.

Williams et al. 1997. Earlier work on chlorine and urbanization impacts.

Williams et al. 2000. Ontario springs; Chloride Contamination Index.

Specific Effects: Trace or Heavy Metals/Contaminants

Aartila 1996. Sheboygan River sediments in Wisconsin.

Canfield et al. 1996. Great Lakes, sediments, TRIAD approach, chemistry, toxicity, benthos.

Clements et al. 1988. Copper; taxa, dominants, and abundance.

Diamond et al. 1993. Acid mine drainage; metal toxicity; EPT, Bray-Curtis ordination.

Diggins and Stewart 1998. Chironomids, community composition, metals.

Duchrow 1983. Ozark study; sensitivities; stoneflies and mayflies, lead tailings.

Faith et al. 1995. Mining aspects in Australia; BACI, family level, Bray-Curtis analysis.

Garie and McIntosh 1986. Urban runoff, heavy metals in New Jersey.

Hoke et al. 1995. Macroinvertebrate community structure; contaminants in sediments.

Humphrey et al. 1995. Mining impacts; biological monitoring.

Ingersoll et al. 1995. Review of methods and applications, toxicity, bioaccumulation, invertebrates.

Kiffney and Clements 1994. Zinc; taxa richness in Colorado streams.

LaPoint et al. 1984. Number of taxa; community structure in 15 United States streams.

Lynch et al. 1988. Mine drainage, trace metals, aquatic insects.

Nelson and Roline 1993. Mayflies as indicators of metals in the Arkansas River.

North American Benthological Society 1995. Workshop on benthic ecology in assessing sediment contamination.

Perkins 1983. Effects of copper on aquatic insects; community comparison index.

Phipps et al. 1995. Contaminants; amphipods, chironomids, oligochaetes.

Roline 1988. Diversity; Baetis tolerant, heavy metals in the Arkansas River.

Waterhouse and Farrell 1985. Chironomid communities, heavy metal gradient, 13 similarity coefficients tested.

Specific Effects: Acid Rain

France 1992. Lakes in Canada; amphipods as biotic index to acidification.

Hall 1994. Ontario study; pH, drift, macroinvertebrate densities.

Stephenson et al. 1994. Littoral benthic macroinvertebrates; lake acidification, NMDS ordination, taxonomic importance index.

Weatherley and Ormerod 1989. Taxa richness associated with acidification.

Specific Effects: Phosphorus and Other Nutrients

Clarke and Ainsworth 1993. Study dealing with Diatoms, but approach is applicable to other organisms; CANOCO, diatoms and environment.

Monda et al. 1995. Euclidean similarity index to detect effects of ammonia.

Peterson et al. 1993. Experimental phosphorus fertilization; impacts on insects.

Rae 1989. Chironomids, Ohio, nutrient enrichment.

Walker 1993. Paleolimnological biomonitoring; chironomids.

Specific Effects: Dams and Hydropower

Barwick et al. 1985. Differences in prey and fish feeding patterns below dams.

Blinn et al. 1995. Effects of dams, desiccation/freezing fluctuations on recolonization.

Bournaud et al. 1996. Longitudinal pattern in Rhone River; PCA analysis.

Cereghino and LaVandier 1998a. French study; Plecoptera, hydropeaking.

Cereghino and LaVandier 1998b. French study; hydropeaking, mayflies.

Cereghino et al. 1997. French study; Trichoptera, hydropeaking.

Camargo and de Jalon 1995. Spanish study; taxonomic richness, scrapers, BA-design, impoundments.

Converse et al. 1998. Colorado River flow regulation, shoreline habitat loss. Although orientated to fish, results are significant to insects.
Englund et al. 1997. Flow regulation, caddisflies, Sweden, low versus high flow effects.
Fisher and LaVoy 1972. Hydroelectric dams; effects on macroinvertebrate community.
Gislason 1985. Flow fluctuations, aquatic insects, standing crop in Washington.
Hooper 1993. Wisconsin study; seasonality, habitat, impoundment effects, snag sampling.
Inverarity et al. 1983. Welch study; use of Jaccard Similarity coefficients; clustering.
Knight and White 1982. Unpublished review article on environmental effects of dams on fish and insects. (available from DNR library)
Malmqvist and Englund 1996. Hydropower flows, mayflies in Sweden, shallow communities affected.
Mason and Lehmkuhl 1983. Chironomids and dams.
Matter et al. 1983. Mississippi River inverts and peaking hydropower.
Mundahl and Kraft 1988. Growth rates of mayflies and stoneflies below dams.
Olmsted and Bolin 1996. Industrial perspective; electric utility; dams.
Pardo et al. 1998. Dams, mayflies in Australian streams.
Petts and Greenwood 1985. Substrate changes; characteristic fauna.
Richardson and MacKay 1991. Changes in filter feeders below lake outlets.
Smock et al. 1989. Debris dams, macroinvertebrates in Virginia.
Troelsrup and Hergenrader 1990. Power peaking, functional-feeding group analysis in the Missouri River.

Biological Community Analyses

Aartila 1996. Sheboygan River; community assessment.
Anderson and Day 1986. Invertebrate habitat association on Mississippi River.
Angradi 1996. Benthic community structure, functional feeding groups, log dams.
Angradi 1996. Community structure analysis using PCA ordination, functional classes.
Bargos et al. 1990. Use of correspondence analysis ordination; biotic index in Spain.
Barmuta 1989. Australian study; erosional versus depositional community structure using Twinspan analysis.
Barton 1996. Use of Percent Model Affinity index in Ontario streams.
Bechara 1996. Argentinan lakes, community structure, PCCA analysis.
Bournaud et al. 1996. France study; large river; family level ordination analysis, taxonomic resolution issues.
Bowman and Bailey 1997. Examines issue of taxonomic resolution on study of community structure.
Bruns and Minshall 1985. Functional feeding groups; ordination river continuum.
Bunn 1995. Australian study; overview, community persistence using ordination.
Cao et al. 1997b. Evaluation of nine similarity indices in England.
Chessman 1995. Family level BI; multihabitats in Australia.
Clements 1991. Discharge impacts taxa richness and CV, percent dominance, sampling methods.
Corkum 1989. Use of cluster analysis; invertebrates in northwest North America.Dudley 1988. Aquatic plants and drift invertebrate associations.
Corkum 1992. Ecoregions, biomes; multivariate and discriminate analysis.
Corkum and Ciborowski 1988. Community associations with environmental data using CANOCO.
Cortes 1992. Portuguese study; decorna, twinspan, and CCA ordination benthic communities.
Courtemanch et al. 1989. Biological standards in Maine.
Crunkilton and Duchrow 1991. Effects of stream order on biotic index.
Culp and Davies 1980. Saskatchewan study; substrate, invertebrate densities, reciprocal averaging, polar ordination.
Elstad 1986. General community composition; Mississippi River.
Evans and Norris 1997. Macroinvertebrate assemblages, habitat, microscale.
Ferraro and Cole 1992. Taxonomic level resolution required for detecting impacts in Washington.
Fleituch 1992. Uses Jaccard Similarity Index; biotic indices; environmental variables.
France 1992. Amphipods in lakes.
Gregg and Rose 1985. Influence of macrophytes on stream invertebrate community.

Growns et al. 1992. Australian wetlands; ordination MDS.
Growns et al. 1995. Australian rivers; ordination in rapid assessments.
Growns et al. 1995. Use of cluster analysis and ordination.
Gumbiner 1995. Wisconsin community analysis of Nicolet Forest streams.
Hawkins et al. 1997. Temperature and channel morphology, community structure.
Hornbach et al. 1989. Upper Mississippi River, CANOCO, habitat and invertebrates.
Jackson 1993. Use of multivariate analysis ordination invert communities; good review of methods.
Johnson and Gage 1997. Aquatic ecosystems, landscape scale, GIS.
Journal of the North American Benthological Society 1988 Vol 7(4). Issue is devoted to Community Structure and Stream Ecosystem Theory (many good articles).
Kerans et al. 1992. Tennessee study; pools versus riffles, sampling differences.
Koetsier et al. 1996. Alkalinity; community associations, landscape scale.
Lyons 1996. Wisconsin fish communities; ecoregions; methods.
Mackay 1992. Invertebrate colonization, good review article.
Malmquist and Maki 1994. Sweden; CCA and Twinspan community analysis, richness, predictions.
Marchant et al. 1997. Prediction of macroinvertebrate assemblages, richness in Australian streams.
Mason 1974. Early work using chironomids.
Mason and Lehmkuhl 1985. Chironomid communities in Canada.
Matthews et al. 1991. Non metric clustering techniques; comparisons communities.
Moss et al. 1987. Prediction of total community in Great Britain; Twinspan.
Muck and Newman 1992. Minnesota amphipods.
Nilsen and Larimore 1973. Early large woody debris paper.
Obi and Conner 1986. Composition of invertebrate drift in lower Mississippi River.
Paine 1966. Classic food web paper; species diversity, 'keystone' species.
Pearson 1986. Species co-occurrence emphasis.
Poff 1997. Landscape habitat filters, species traits, prediction of community structure.
Quinn and Hickey 1990b. New Zealand rivers; taxa richness, invert community index.
Rabeni and Gibbs 1980. Ordination and Bray-Curtis; PS similarities, Maine rivers.
Rae 1985. PCA chironomid community comparisons in Ohio; sediments.
Rae 1989. Ohio chironomid communities.
Richards and Host 1994. Land uses; communities; PCA analysis, richness, filterers.
Richoux 1994. Habitat templets; beetles; species traits, Upper Rhone River.
Rossaro and Pietrangelo 1993. Correspondence analysis; BIs; richness, etc.
Sheaffer and Nickum 1986. Invertebrates, habitats; backwaters of the Mississippi River.
Sheldon 1972. Classic paper on classification systems; ordination of aquatic communities.
Sheldon and Haick 1981. PCA and canonical correlation in Montana.
Smith and Kaster 1983. Taxa richness; percent composition southeastern Wisconsin.
Smock et al. 1989. LWD (large woody debris); percent shredders.
Soluk 1985. Chironomids in Canada shifting sand bottomed rivers.
Stelzer and Lamberti 1994. Unpublished report; Macroinvertebrate community structure in Chequamegon Nicolet Forest streams.
Tachet et al. 1994. Habitat templets; caddisflies; species traits, Upper Rhone River.
Tolkamp 1985. Netherlands evaluation of different indices.
Usseglio-Polatera 1994. Habitat templet, aquatic insect richness, species traits, Upper Rhone River.
Vannote et al. 1980. Classic river continuum concept paper.
Walley and Fontama 1998. Use of 'neural networks' to predict invertebrate index scores in Britain; ASPT, family level.
Waterhouse and Farrell 1985. Chironomid communities; similarity/dominance.
Whitehurst 1991. Gammarus:Asellus index.
Whiting and Sheard 1990. Ordination mayfly communities in Canada.

Wohl et al. 1995. Euclidean similarity/Cluster analysis, southern Appalachian streams.
Wright 1995. RIVPACS; biological assessment Britain.
Wright et al. 1993. British study; Biological Monitoring Working Party System and RIVPACS.
Wright et al. 1995. Influence of taxonomic resolution on ordination of communities in Australia.
Wright et al. 1995. NMDS ordination; Bray-Curtis, taxa richness; tax. resolution.
Zamora-Munoz and Alba-Tercedor 1996. Spanish study; BMWP score, twinspan and CCA ordination.

Paired Comparisons: Similarity/Dissimilarity

Barton and Metcalfe-Smith 1992. Comparison of eight indices or metrics in Canada.
Beisel et al. 1996. Comparison of four dominance indices.
Bloom 1981. Compares four similarity indices, Bray-Curtis best.
Boyle et al. 1990. Evaluation of nine diversity and seven similarity indices, perturbations; 'B' similarity best.
Brock 1977. Early comparisons of similarity measures.
Cao et al. 1997a. Effect of sample size on eleven similarity measures.
Cao et al. 1997b. Nine similarity indices evaluated.
Clarke and Ainsworth 1993. Complex paper; multivariate analysis/ MDS ordination.
Cuffney et al. 1993. Bray-Curtis and percentage similarity coefficients; QA/QC evaluations.
Faith et al. 1995. BACI and paired comparisons; Bray-Curtis, Kulczynski; family level.
Hendrickson 1978. Presence/absence data, Jaccard Index.
James and Rathbun 1981. On avian communities, but valuable discussion on rarefaction and diversity indices.
Lillie and Isenring 1996. Application of Pinkham-Pearson index to Baraboo Hills communities.
Novak and Bode 1992. Presentation of Percentage Similarity Index.
Pedersen and Perkins 1986. Comparisons of six similarity coefficients; Canberra Metric Index.
Perkins 1983. Diversity and similarity comparisons; Bray-Curtis review.
Pinkham and Pearson 1976. Presentation of 'B' Similarity Index and others.
Waterhouse and Farrell 1985. Similarity coefficients; chironomids.
Wolda 1981. Comparison of similarity indices; Morisita Index preferred.

Special Statistics

Berges 1997. Ratios and spurious correlations.
Beyers 1998. Inference, design considerations, randomization, replication issues.
Bunn 1995. Biological monitoring study design considerations.
Clarke and Ainsworth 1993. Multivariate analysis using community structure and environmental variables.
Eggert et al. 1992. Comparisons of BACI and RIA analysis of diatoms.
Gauch 1982. Chapter in book on ordination techniques.
Hawkins 1986. Precaution to pseudoreplication.
Heffner et al. 1996. More on the 'pseudoreplication' debate.
Humphrey et al. 1995. Study design considerations; BACI.
Hurlbert 1984. Psuedoreplication, study design.
Jackson et al. 1990. Discusses statistical shortcomings of using ratios.
McDonald and Erickson 1994. Bioequivalence; hypothesis testing.
Nemec and Brinkhurst 1988a and 1988b. Ordination techniques; multivariate analysis.
North American Benthological Society 1993 Technical Workshop on "Use of Biostatistics in Benthic Ecological Studies".
North American Benthological Society 1996 Technical Workshop on Study Design and Data Analysis. Assessment using reference site approach.
Stewart-Oaken et al. 1986. More on pseudoreplication.

Other Indices: Morphological Deformities

Camargo 1991. Caddisfly deformities as biological indicators.
Diggins and Stewart 1998. Chironomid deformities associated with trace elements.
Lemly 1982. Wax and bacteria buildup on filamentous gills.
Vermeulen 1995. Review of use of chironomids, toxic sediments.
Warwick and Tisdale 1988. Use of chironomids in Canadian contaminant studies.

Other Methods

Cairns et al. 1968. Sequential Sampling Index.
Castella and Amoros 1988. Graphic presentations and comparisons; faunal typology.
Chevenet et al. 1994. 'Fuzzy coding theory'; correspondence analysis using beetles.
McCoy et al. 1986. Identifying "biotic boundaries" using similarity matrices.
McDonald and Erickson 1994. Innovative test for measuring 'bioequivalence'; disturbed versus reference site comparisons.
Peters et al. 1989. Utilization and habitat preference metrics for macroinvertebrates and fish in Nebraska.
Resh and Price 1984. Test of Sequential Sampling Index.
Roberts 1986. 'Fuzzy set' theory and use in ordination.
Smith 1989. Water quality index; 'minimum operator' concept.
Southwood 1977. Classic work on habitat templet theory.
Townsend and Hildrew 1994. Habitat templet theory, species traits.

Composite Indices/ Multimetric

Barbour et al. 1995. Multimetrics basics. A good review article.
Barbour et al. 1996. Florida ecoregions; 32 candidate metrics tested.
Fore et al. 1996. Multimetric and multivariate approaches evaluated.
Gaunt and Thorpe 1993. Michigan biological condition scoring criteria; EPT, dominance, richness, CLI, etc.
Gerritsen and White 1997. Florida biological index, metrics, scoring.
Karr and Kerans 1992. Evaluation of 28 metrics; development community index.
LaLiberte 1996. Development of a multimetric community Index for western Wisconsin streams.
Ohio Environmental Protection Agency 1987 and 1990. Invertebrate community Index; based on 10 metrics.
Plafkin et al. 1989. USEPA Rapid bioassessment protocols. see updated second version on line at **http://www.epa.gov/OWOW/monitoring/rbp/**.
Resh et al. 1995. Rapid bioassessment metrics; richness, diversity, similarity, BIs, etc.
Reynoldson et al. 1997. Reference conditions; assessment of multimetric versus multivariate methods.
Shackleford 1988. Bioassessment in Arkansas; dominants, taxa richness, indicators, etc.
Thorne and Williams 1997. Evaluation of multimetric, family level macroinvertebrate indices.
Weisberg et al. 1997. Estuarine benthic IBI for Chesapeake Bay.

Applications

Armitage et al. 1983. Evaluation and comparison of the performance of biotic indices used in Great Britain.
Barton and Metcalfe-Smith 1992. Richness, diversity, BIs.
Camargo 1992. A new diversity index.
Camargo 1993. Evaluation biotic indices; recommends BMWQ score.
Cook 1976. Poor performance of Shannon-Wiener diversity and BI indices.
Crunkilton and Duchrow 1991. Missouri; density, diversity, richness, modified Margalef; variation with stream order.
Day et al. 1992. Horn's Similarity Index; cluster analysis, drift, Upper Mississippi River.
Dilley 1992. Test evaluation of Ohio's ICI.
Drake 1997. Rapid bioassessment of Fort McCoy streams in Wisconsin.
Gaunt and Thorpe 1993. Michigan evaluation of six metrics.
Heckman et al. 1990. German study; evaluation of several multivariate methods and indices.

Hooper 1993. Use of 25 metrics Wisconsin; riffle/snag and dam effects; HBI, FBI, richness, trophic classes, and Margalef's diversity.
Hughes et al. 1990. Incorporating ecoregions into bioassessments.
Illinois EPA 1988. Pecatonica River assessment using modified HBI and richness.
Johnson 1998. Detecting impacts in Swedish lakes; benthic quality index, taxa richness, ASPT.
Lewis and Smith 1992. HBI, Shannon-Weaver diversity, CLI, similarity indices including Jaccard's.
Lillie and Schlesser 1994. Southwest Wisconsin streams; Bray-Curtis, functional classes; BIs.
Maret 1988. Use of Shannon Diversity; modified HBI in Nebraska streams.
Maxted et al. 2000. Update of draft protocols using FBI, richness, dominance, metrics.
Modde and Drewes 1990. Artificial vs. natural substrates; HBIs, percent similarities; effect of flows on biotic index scores.
Murphy 1978. Temporal variability in BIs and diversity indices; Margalef's.
Oda et al. 1991. Japanese study; diversity and BIs; PCA analysis.
Olive and Smith 1975. Richness and Shannon-Wiener diversity.
Pedersen and Perkins 1986. Compares 6 similarity coefficients; Canberra-Metric best; discusses weakness of Bray-Curtis.
Pontasch and Brusven 1988. Bray-Curtis similarity indices and diversity.
Rabeni et al. 1985. Response of inverts to pollution abatement; BIs and PS similarity and ordination.
Reynoldson et al. 1997. Comparison of multivariate methods versus multimetrics.
Rheume et al. 1996. Invertebrate in eastern Wisconsin; PCA and other metrics, geography.
Rico et al. 1992. Comparison of biotic indices in Spain.
Rosillon 1989. Richness, diversity (Shannon-Weaver), density, annual changes.
Rossaro and Pietrangelo 1993. Test of CA ordination and biotic indices in Italy.
Tolkamp 1985. Comparisons among various biotic indices in the Netherlands.
Twidwell and Davis 1989. Texas study; least disturbed streams, richness, diversity, dominance, etc.
Wallace et al. 1996. Evaluation of NCBI and EPT index to detect impacts of insecticides.
Zamora-Munoz and Alba-Tercedor 1996. CANOCO; twinspan evaluation of BMWP biotic index in Spanish rivers.
Zamora-Munoz et al. 1995. Evaluation of seasonality of several biotic indices.

Biological Integrity

Angermeier and Karr 1994. Definitions and policy applications; Biotic integrity versus diversity.
Barbour et al. 1996. Florida Stream Condition Index tested using reference sites; metric sensitivity discussed.
Cairns 1995. Ecological integrity, definitions, concept, scales, philosophy.
Done and Reichelt 1998. Performance indices, management goals. Desired versus desired states.
Fore et al. 1994. Statistical properties of the IBI; important to benthic BIs also.
Harig and Bain 1998. Biological integrity of wilderness lakes in the Adirondacks using fish, phytoplankton, and zooplankton.
Hawkins et al. 2000. Use of multivariate RIVPACS predictive models for measuring biological integrity.
Jackson and Davis 1994. Definitions, goals, policy perspectives.
Jennings et al. 1999. IBI for inland lakes in Wisconsin using fish, but good for concepts and principles.
Karr and Kerans 1992. Definitions of biotic integrity and development of an index.
Kerans and Karr 1994. TVA rivers (B-IBI) index, 13 invert attributes; pools/riffles.
Kleidal 1995. Benthic Index of Biotic Integrity for Washington streams.
Lyons 1992. Development of fish, warmwater IBI in Wisconsin; Methods.
Lyons 1996. Stream classification, fish, coldwater streams.
Polls 1994. Use of reference sites, etc; industry perspective.
Reice and Wohlenberg 1993. Review article on aquatic ecosystem health.
Simon and Lyons 1995.General review.
Steedman 1994. Ecosystem health.
Steedman and Haider 1993. Ecological integrity.

Appendix B. A History of the Development of Macroinvertebrate Biological Indices in Wisconsin

The use of macroinvertebrate biotic indices for water quality assessment in Wisconsin began with the development of the Hilsenhoff Biotic Index (HBI) at the University of Wisconsin Madison in cooperation with the DNR (Hilsenhoff 1977). The original index was based on collections from a set of 53 Wisconsin streams (29 undisturbed and 24 polluted or disturbed). Tolerance values were assigned to arthropod taxa on the basis of each taxon's affiliation with an assigned (subjective) stream disturbance class from 0 (undisturbed) to 5 (severely polluted). The original index provided for five water quality classes. Hilsenhoff (1982) added improvements to the index and methodology after sampling over 1000 streams, changing tolerance values for some taxa, assigning tolerance values for additional taxa, and expanding the water quality rating scale to include six water quality classes. Hilsenhoff modified and refined the HBI in a series of publications. These modifications include: incorporating additional tolerance values, expanding the scale from 0-5 to 0-10, adding a seventh water quality class, deriving seasonal correction values, and modifying the index to minimize undue effects arising from dominant taxa (Hilsenhoff 1987, Hilsenhoff 1988b, Hilsenhoff 1998).

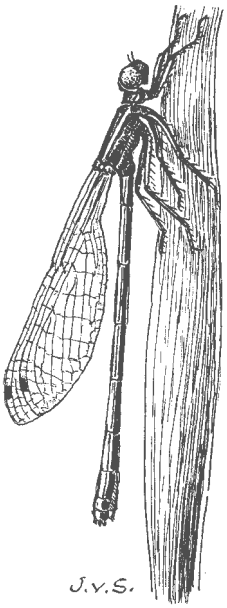
Hilsenhoff (1988a) developed a family level version of the HBI with seven water quality classes and Lillie and Schlesser (1994) proposed a complementary index measure based on a non weighted, mean tolerance value of all taxa represented in an HBI sample. In 1989, the standard HBI was incorporated into the national "Rapid Bioassessment Protocols for Use in Streams and Rivers" (Plafkin et al.1989). Lillie (2000) has developed a multimetric macroinvertebrate index for use in wetlands.

Early assessments of water quality in Wisconsin using the HBI were done by Dr. Hilsenhoff at UW-Madison. Beginning in 1983, a contractual agreement was established with the UWSP to process HBI macroinvertebrate samples. At that time there was a basic program that calculated the HBI from sample data. This program was used by Bob Young (DNR) and further developed at the North Central District headquarters by a computer programmer. The program was developed in the dBase® III environment and has since been modified to suit DNR research needs and interests. In 1987 the program was modified to include the various metrics and utilities that are currently used. In 1992 the program was moved to the dBase® IV environment to increase speed and allow functionality as a stand alone program outside of the dBase® IV environment. At this time additional utilities were added to make the program easier to use.

A 'Taxamaster' file, which includes unique organism ID numbers and their associated tolerance values, has been maintained since 1983. Because the taxonomic literature is continually monitored by UWSP staff, information derived from this effort is used to update the 'Taxamaster' file on a routine basis. The latest version of the program (BUG-PROGRAM 6.0) includes computation of the HBI (MAX *x*) (Hilsenhoff 1998) and replaces Margalef's Diversity Index with the Shannon Index.

When examining historical macroinvertebrate data, it is important to know why the samples were collected and where the collections were made. Some historical macroinvertebrate data may represent atypical samples (i.e., snags, pools, impounded areas) or anomalous conditions (i.e., fertilizer spill, during or after severe flooding or drought). In other cases, documentation and field records associated with collections may be incomplete or simply missing. Such data should be used with caution to avoid misapplication

A high degree of taxonomic training and experience is required to correctly identify aquatic insects. Under the direction of highly qualified taxonomists, macroinvertebrate work (including most HBI calculations) conducted in Wisconsin has been done at UW-Madison, UWSP, and the UW Superior Lake Superior Research Institute. The Lake Superior Research Institute has been contracted by the DNR to do taxonomic and field sampling for in-place pollutant studies, endangered resource inventories, and dam removal assessments. In addition, many independent research studies have been conducted outside of DNR by various universities and colleges and the resultant data are available in unpublished theses or published articles. Other studies have been conducted by the US Fish and Wildlife Service, US Geological Survey, Natural Resources Conservation Service, The Nature Conservancy, and several private consultants. Within the DNR, the Bureau of Integrated Science Services has conducted several research studies involving macroinvertebrate and HBI data. The Bureau of Endangered Resources has conducted survey work to maintain the Natural Heritage Inventory Database that tracks the occurrences and distributions of species listed as endangered, threatened, or of special concern. Some regional offices have also conducted their own family level biotic assessments and several department staff members maintain private collections. More recently, numerous studies (in some cases only collections) have been conducted by schools and non-profit organizations using simplified rapid bioassessment protocols.



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