

The Great Lakes Entomologist

Volume 20
Number 1 - Spring 1987 *Number 1 - Spring 1987*

Article 7

April 1987

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Recommended Citation

Hilsenhoff, William L. 1987. "An Improved Biotic Index of Organic Stream Pollution," *The Great Lakes Entomologist*, vol 20 (1)

DOI: <https://doi.org/10.22543/0090-0222.1591>

Available at: <https://scholar.valpo.edu/tgle/vol20/iss1/7>

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AN IMPROVED BIOTIC INDEX OF ORGANIC STREAM POLLUTION¹

William L. Hilsenhoff²

ABSTRACT

Major improvements were made in using a biotic index of the arthropod fauna to evaluate organic stream pollution. All tolerance values were reevaluated, many were changed, and the scale for tolerance values was expanded to 0–10 to provide greater precision. Keys to larvae of *Ceratopsyche* have been developed and tolerance values for species in this important genus are provided. Sorting of samples in the laboratory instead of in the field is recommended, and directions for processing and evaluating samples are included.

A “saprobic index” (Pantel and Buck 1955) and a “biotic index” (Chutter 1972) were proposed for evaluating the water quality of streams through a study of their fauna. I introduced a similar biotic index (Hilsenhoff 1977) that used only arthropods for evaluation, thus simplifying collecting, sorting, and identification. It was based on a sample of 100 or more arthropods collected from a riffle area. This index is a measure of organic and nutrient pollution, which causes lower dissolved oxygen levels, especially at night during the summer and after heavy rain. Lowered levels of dissolved oxygen in turn affect the ability of each species of arthropod to survive in a particular stream. For the purpose of calculating the biotic index, every species or genus was assigned a tolerance value of 0–5, with 0 assigned to species most intolerant of organic pollution and 5 assigned to the most tolerant species. Intermediate values were assigned to species intermediate in their tolerance of organic pollution. The biotic index is an average of tolerance values for all individuals collected from a site.

Initially the index was used mostly in Wisconsin. In 1979 and 1980 the Wisconsin Department of Natural Resources (DNR), in cooperation with the University of Wisconsin, used the index to evaluate more than 1000 stream sites in spring and autumn. Personnel in my laboratory identified all the arthropods and calculated biotic index values for all sites. Experience from this cooperative study and several other studies enabled me to publish new recommendations for using the biotic index, revised tolerance values, and regional keys to species in important genera (Hilsenhoff 1982).

Recently, additional improvements have been made in the biotic index. Most important are an expansion of the scale of tolerance values to 0–10 to provide greater precision, a reevaluation of all tolerance values, and inclusion of tolerance values for many additional species. Procedures for sampling and sorting were updated, and a discovery that *Simulium vittatum* is really two genetically distinct species (Rothfels and Featherston 1981) with differing ecological requirements (Adler and Kim 1984) altered my recommendation for dealing with these sibling species. Adequate correction factors for current, temperature,

¹Research supported by the College of Agricultural and Life Sciences, University of Wisconsin—Madison, and by Hatch Research Project 2785.

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and seasonal differences are needed, and studies to correct these deficiencies will be completed soon.

REASSIGNMENT OF TOLERANCE VALUES

Initial tolerance values for each species were based mostly on a study of 53 Wisconsin streams in which physical and chemical parameters were evaluated to determine the degree of organic and nutrient pollution in each stream (Hilsenhoff 1977). Tolerance values for species not found or occurring rarely in these streams were based on their occurrence in other streams and their association with species to which tolerance values had been assigned.

Data from more than 2000 samples collected in the 1979–80 cooperative study with the DNR were used to reevaluate tolerance values by comparing the tolerance value of each species with the average biotic index value of streams in which that species most commonly occurred. A description of the procedure that was used can be obtained from the author. It became apparent that intermediate values would increase precision, so the 0–5 scale of tolerance values was expanded to 0–10 to accommodate intermediate values while retaining whole numbers for ease in making calculations. New tolerance values assigned to 359 species or genera found in the DNR samples are listed in Appendix 1.

Forty-nine additional species that were not collected from the 1979–80 study streams, mostly because their life cycles precluded their being present in spring or fall samples, were assigned tolerance values based on our knowledge of streams in which they occurred. Previous experience was also used to adjust tolerance values of nine species that were found in less than five samples from the study streams, and subsequent experience resulted in adjusting values of *Asellus*, *Crangonyx* and *Hyallela*.

IDENTIFICATION

Accurate identification of arthropods to species is often necessary, especially when a species is numerous in a stream and thus greatly influences calculation of the biotic index. Tolerance values marked with an asterisk (Appendix 1) are least reliable, but the species involved are uncommon in stream riffles and rarely exert much influence on the biotic index. Generic identifications are used only when species identifications are not possible or very difficult, or when in a region all known species in a genus have the same tolerance value. Merritt and Cummins (1984) included keys to North American genera and also referenced regional generic keys, which are easier to use. Hilsenhoff (1981) provided a regional key to genera and references to species keys, which are widely scattered in the literature. Brigham, Brigham and Gnilka (1982) contained regional species as well as generic keys, and Merritt and Cummins (1984) listed species keys, regional keys, and other taxonomic references published through 1982. Recent species keys and identification problems as they relate to the biotic index are discussed below for each order.

PLECOPTERA. Mature nymphs of *Acroneuria*, *Allocapnia*, *Pteronarcys*, and *Taeniopteryx* can be identified, but there is a degree of uncertainty in many identifications, especially when nymphs are immature. Because in these genera all species in the western Great Lakes region appear to have similar tolerance values, only a generic tolerance value is used. *Perlesta placida* may be a species complex, with one or more of the species being much less tolerant of organic pollution.

EPHEMEROPTERA. Identification of mayfly nymphs in many genera is not possible, and in other genera species identification must be regarded as somewhat tentative. McCafferty (1975) keyed species of Ephemeridae Polymitarcyidae, and Potamanthidae, but separation of nymphs in these families is not always possible. Since they are relatively uncommon in riffle samples, and because species within each genus appear to have similar tolerance values, only generic tolerance values have been assigned. A recent key by Kondratieff and Voshell (1984) identified nymphs of *Isonychia*, but separation of

species requires mature nymphs and is difficult. Only a generic tolerance value was provided for this genus, and it may be too low for one or more of the species. *Ephemerella* species A (Hilsenhoff 1982), which occurs in western Wisconsin, is probably *E. inermis* or a sibling species of that species. Reared adults most closely fit the description of *E. inermis*, as do dark-colored nymphs. *Pseudocloeon* species A (Hilsenhoff 1982) may be a new species, or a species for which nymphs have not been described. Arwin Provonsha at Purdue University has revised the genus *Caenis* and developed keys to the nymphs, which he will publish soon. Identification of *Caenis* nymphs is difficult, and because the three riffle species all have similar tolerance values, only a generic value is included.

ODONATA: Several Odonata that were assigned tolerance values are essentially lentic and occur only occasionally in streams and rarely in riffles. Only nine species or genera were found in 10 or more of the 1979–80 study streams. They rarely made up more than 10% of a sample, so this order is usually not important to biotic index values. Generic tolerance values are used for uncommon genera when species identification is difficult. Only mature nymphs of *Ophiogomphus* can be identified to species (Walker 1958, Needham and Westfall 1955), so a generic tolerance value is also used for this important genus.

TRICHOPTERA: Species identification of larvae in this important order is often difficult or impossible. Recent studies of larvae of Brachycentridae (Chapin 1978, Flint 1984) and Hydropsychidae (Schuster and Etnier 1978, Scheffer and Wiggins 1986) and regional keys to most species in these families (Hilsenhoff 1985, Schmude and Hilsenhoff 1986) permitted assignment of tolerance values for many species of these important families. Unfortunately larvae of *Cheumatopsyche* cannot be identified, and apparently relatively intolerant as well as tolerant species exist.

COLEOPTERA: Elmidae adults were observed crawling out of an artificial stream when dissolved oxygen levels become too low, so their value as indicator organisms may be questioned, but larvae probably do not react in the same manner. The genus *Stenelmis* is being revised, and several undescribed species exist. However, almost all adults, and probably larvae also, that were collected from riffles in Wisconsin streams were *S. crenata*. The tolerance value for this species is 5, and that value has been assigned to the entire genus because there is no indication that other species have significantly different tolerance values. Larvae of *Optioservus* and *Dubiraphia* cannot be identified to species so generic tolerance values are used.

DIPTERA: Identification of larvae to species is usually not possible, and sometimes even genera cannot be separated. Larvae of Simuliidae are often common, and well illustrated keys to Canadian species have been published (Wood et al. 1963, Peterson 1970), but their identification is difficult. Pigmentation of the head is variable within the same population, and pupal respiratory filaments must be dissected from mature larvae to identify some species. Furthermore, several species are complexes of genetically distinct species that cannot be separated by morphological characters as was discovered for *Simulium vittatum* (Rothfels and Featherston 1981). Fortunately many species occur on macrophytes or in deeper water and are unlikely to be collected from riffles. Efforts were also made to distinguish species groups within several genera of Chironomidae, but much more work is needed on this important family and tolerance values for Chironomidae have been assigned only to genera. Recent keys separate *Tribelos* from *Endochironomus* and *Phaenopsectra* (Simpson and Bode 1980), *Tvetenia* from *Eukiefferiella* (Bode 1983), and *Xylotopus par* from *Brillia* (Oliver 1982).

COLLECTION AND EVALUATION OF SAMPLES

Organic stream pollution is evaluated by calculating biotic index values for arthropod communities that inhabit rock or gravel riffles. Samples from pools or under the banks of streams should not be used. Arthropods that inhabit riffles are found on rocks and pebbles or in sand and gravel associated with the riffle, and especially in organic debris that

accumulates between rocks and pebbles. In deeper streams that have no riffles, samples from rock or gravel runs may be substituted, and in sand-bottomed streams samples of debris that collects on sticks or other objects wedged into the sand in swift current may be used. Slow flowing, silt-bottomed streams presently cannot be evaluated with the biotic index. The season of the year affects the biotic index, with much higher biotic index values often being recorded during the summer months. Results from the first year of a study to develop seasonal correction factors suggest that evaluation may not be possible in the summer months, and that streams should be evaluated from samples collected in the spring before 1 June or after 1 September and before 15 October in the autumn. Samples collected from organically enriched or polluted streams tend to have distinctly higher biotic index values after mid-October and very much higher values in the summer.

In 1982 I recommended a procedure for collecting and sorting samples that involved picking 100+ live arthropods from a sample in the field and included an alternative procedure for picking samples in the laboratory. Experience since that time suggests that many more streams can be sampled in a given period of time if valuable field time is not used to pick the samples. The greatest difficulty in collecting samples for processing in the laboratory was in knowing when an adequate sample had been collected. Our experience with several hundred samples from six diverse streams suggests that when there is enough debris in the sample to fill an 8-ounce (237-ml) jar there will be enough arthropods, and such a sample takes less than 5 minutes to collect. Revised procedures for collecting, sorting, and evaluating samples with the biotic index follow.

1. Use an aquatic net to sample a site where the current is greater than 0.30 m/sec (1.0 ft/sec) and the substrate is composed of gravel, pebbles, and (or) small rocks. This should preferably be a riffle area where the substrate causes a disturbance of the water's surface. All riffle areas with a depth of at least 10 cm will have a current of at least 0.30 m/sec. In sand-bottomed streams, sample debris collected on snags in fast current. Collection of arthropods is best accomplished by placing the net against the stream bottom and disturbing the substrate immediately upstream from the net with your feet. In very fast currents avoid having the net so close to your feet that gravel and pebbles are washed into it. Avoid collecting from rooted macrophytes and filamentous algae.

2. Collect until there is enough debris in the net to fill an 8-ounce (237 ml) jar, or until it is obvious that more than 100 arthropods have been captured. Remove sticks and large undecomposed leaves from the rest of the debris, washing arthropods from them by rinsing in the net in a pool area.

3. Place the arthropods along with all debris in a labeled jar and add enough 95% ethanol to produce a concentration of about 70% when mixed with the water in the debris. Include all arthropods clinging to the net that are 3 mm long and all adult Elmidae.

4. After returning to the laboratory, or if in the field for more than a day, sample jars completely filled with debris should be drained of ethanol by pouring through a screen or net with a 1.0 mm or smaller mesh. The ethanol should then be replaced with 70% ethanol. An alternative is to refrigerate the samples to allow ample time for diffusion of the ethanol throughout the debris and into the arthropods.

5. About 15 minutes before picking and sorting a sample in the laboratory, strain the ethanol from the sample and replace it with water. No arthropods should remain floating on the surface of the water.

6. Place the contents of the jar in a large, flat pan marked with a grid, add two or three additional jars full of water, and spread the contents evenly over the bottom of the pan. If the jar is completely filled with fine debris, especially filamentous algae, only half of the sample should be initially placed in the pan for sorting, with care being taken to assure that each half of the sample contains the same amount and kinds of debris. A 30 by 45-cm pan with a 5-cm grid is satisfactory. Larger pans allow the debris to be spread more thinly, making it easier to see the arthropods, but pans that are too large are unwieldy. Number the squares in the grid and select a starting square for each sample by picking a number from a box of corresponding numbers or from a table of random numbers. Remove all arthropods from the starting square and then remove arthropods from each successively

higher numbered square. An arthropod on a line is considered to be in the square that contains its head, or in the square closest to its head. After the highest numbered square has been sampled, return to square 1. Remove and preserve at least 100 arthropods. Remove all arthropods from the last square to be picked. Do not collect Hemiptera, or Coleoptera other than Dryopoidea. Except for adult Elmidae and fifth instar Hydroptilidae larvae, which have expanded abdomens and are usually in cases, do not collect arthropods less than 3 mm long because most cannot be identified. An illuminated 5X magnifier on a long, movable arm (Luxo®) will facilitate finding and removing arthropods from the pan.

7. Preserve all arthropods in 70% ethanol for identification to genus or species in the laboratory. Isopropyl alcohol may also be used.

8. Sort and identify all arthropods to genus, except Chironomidae, which should be placed in a separate vial. When all samples have been identified to genus, species identification should be made whenever necessary and possible. This is best accomplished by working on one genus at a time and identifying species in that genus from all samples before identifying species in another genus.

9. Chironomidae are sorted to genus by placing those that look alike together. Head color, head size and shape, markings on the head, antennal length and structure, number and location of eye spots, general shape and pigmentation of the mentum, length and color of preanal papillae and setae, length of prolegs and color of their claws, and general coloration are among the characters that can be used to separate genera. Mount the two most dissimilar larvae from each group in Hoyer's medium under separate cover slips on the same slide. If both are found to be the same genus, the remainder may be assumed to be also the same and need not be mounted. If they are different, further sorting and slide mounting is needed or all must be mounted on slides. An alternative is to clear all larvae in 10% KOH and make temporary mounts in glycerine for identification.

10. Record the number of each species on a data sheet and multiply the number by the tolerance value for that species. Sum the products and divide by the total number of arthropods in the sample to obtain the biotic index for the stream. Table 1 is a general guide to the water quality of streams. Replicate samples, or both spring and fall samples, will add to the confidence of the evaluation.

Table 1. Evaluation of water quality using biotic index values of samples collected in March, April, May, September, and early October.

Biotic Index	Water Quality	Degree of Organic Pollution
0.00-3.50	Excellent	No apparent organic pollution
3.51-4.50	Very Good	Possible slight organic pollution
4.51-5.50	Good	Some organic pollution
5.51-6.50	Fair	Fairly significant organic pollution
6.51-7.50	Fairly Poor	Significant organic pollution
7.51-8.50	Poor	Very significant organic pollution
8.51-10.00	Very Poor	Severe organic pollution

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Appendix 1. Tolerance values for stream arthropods.^a

PLECOPTERA

Acroneuria spp. 0, *Agnatina capitata* 2, *Allocapnia* spp. 3, *Alloperla* spp. 0*, *Amphinemura delosa* 3, *A. linda* 0*, *Attaneuria ruralis* 1*, *Clioerla clio* 1, *Haploperla brevis* 1, *Hastaperla* = *Haploperla*, *Isogenoides frontalis* 0*, *I. olivaceus* 0*, *Isoperla bilineata* 4*, *I. clio* = *Clioerla clio*, *I. cotta* 1, *I. dicala* 2, *I. frisoni* 0, *I. lata* 0, *I. marlynia* 4, *I. nana* 5, *I. richardsoni* 2, *I. signata* 2, *I. stlossonae* 2, *I. transmarina* 0, *Leuctra ferruginea* 0*, *L. sibleyi* 0*, *L. tenella* 0*, *L. tenuis* 0*, *Nemoura trispinosa* 1, *Neoperla stewarti* 1, *Oemopteryx glacialis* 1, *Paracapnia angulata* 1*, *Paragnetina media* 1, *Perlستا placida* 5, *Perlinella drymo* 1*, *P. ephyre* 1*, *Phasganophora* = *Agnatina*, *Prostoia completa* 1*, *P. similis* 2*, *Pteronarcys* spp. 0, *Shipsa rotunda* 2*, *Soyedina vallicularia* 0*, *Strophopteryx fasciata* 3*, *Taeniopteryx* spp. 2.

EPHEMEROPTERA

Ameletus spp. 0*, *Arthroplea bipunctata* 5*, *Attenella attenuata* 3*, *Baetis brunneicolor* 4, *B. flavistriga* 4, *B. frondalis* 5*, *B. hageni* 5*, *B. intercalaris* 6, *B. longipalpus* 5*, *B. macdunnoughi* 5, *B. propinquus* 6*, *B. pygmaeus* 4, *B. vagans* 2, *Baetisca lacustris* 5*, *B. laurentina* 3*, *B. obesa* 5*, *Brachycercus* spp. 3*, *Caenis* spp. 7, *Callibaetis* spp. 9, *Centropilum* spp. 2*, *Choroterpes basalis* 2*, *Cloeon alanceae* 2*, *Cloeon* spp. 4*, *Dannella lita* 4*, *D. simplex* 2*, *Drunella cornutella* 0, *Epeorus vitreus* 0, *Ephemera simulans* 1, *Ephemerella aurivillii* 0*, *E. catawba* 1*, *E. dorothea* 1*, *E. excrucians* 2, *E. inermis* 1, *E. invaria* 1, *E. needhami* 2, *E. subvaria* 1, *Ephoron* spp. 2*, *Eurylophella aestiva* 5*, *E. bicolor* 1, *E. funeralis* 0*, *E. lutulenta* 6*, *E. temporalis* 5, *Habrophlebiodes americana* 6*, *Heptagenia diabasia* 3, *H. flavescens* 4*, *H. hebe* = *Leucrocuta*, *H. lucidipennis* = *Nixe*, *H. pulla* 0*, *Heterocloeon curiosum* 2*, *Hexagenia* spp. 6, *Isonychia* spp. 2, *Leptophlebia* spp. 4, *Leucrocuta hebe* 1, *Lito-brancha recurvata* 6*, *Nixe lucidipennis* 2*, *Paraleptophlebia* spp. 1, *Pentagenia vittigera* 6*, *Potamanthus* spp. 4, *Pseudiron centralis* 5*, *Pseudocloeon carolina* 2, *P. dubium* 4, *P. myrsum* 3*, *P. parvulum* 4, *P. punctiventris* 5, *P. species A* 4*, *Rhithrogena* spp. 0, *Serratella deficiens* 2, *S. sordida* 2, *Siphonurus* spp. 7, *Siphlo-plecton* spp. 2*, *Spinadis* sp. 5*, *Stenacron interpunctatum* 7, *Stenonema exiguum* 5, *S. femoratum* 5, *S. integrum* = *S. mexicanum*, *S. mediopunctatum* 3, *S. mexicanum* 4*, *S. modestum* 1, *S. pulchellum* 3, *S. terminatum* 4, *S. vicarium* 2, *Tricorythodes* spp. 4.

ODONATA

Aeshna spp. 5, *Amphiagrion saucium* 9, *Anax junius* 8*, *Argia apicalis* 8*, *A. moesta* 6, *A. tibialis* 6*, *Basiaeschna janata* 6, *Boyeria vinosa* 2, *Calopteryx aequabilis* 5, *C. maculata* 5, *Chromagrion conditum* 4*, *Coenagrion resolutum* 8*, *Cordulegaster maculatus* 3, *Didymops transversa* 4*, *Enallagma antennatum* 8, *E. civile* 9*, *E. ebrium* 9, *E. hageni* 9*, *Epitheca* spp. 7, *Gomphurus* spp. 6*, *Gomphus* spp. 5*, *Hagenius brevistylus* 1*, *Hetaerina americana* 6, *Hylogomphus brevis* 3, *Ischnura verticalis* 9, *Lestes* spp. 9, *Leucorrhinia intacta* 9*, *Libellula pulchella* 9*, *Macromia illinoensis* 2, *Neurocordulia molesta* 5*, *N. obsoleta* 0*, *N. yamaskanensis* 0*, *Ophiogomphus* spp. 1, *Plathemis lydia* 8*, *Somatochlora* spp. 1*, *Stylogomphus albistylus* 0*, *Stylurus* spp. 4*, *Sympetrum* spp. 10*.

TRICHOPTERA

Agapetus spp. 0, *Agarodes distinctum* 3, *Agraylea* spp. 8*, *Agrypnia* spp. 7*, *Anobolia* spp. 5, *Asynarchus montanus* 6, *Brachycentrus americanus* 1, *B. incanus* 0*, *B. lateralis* 1*, *B. numerosus* 1, *B. occidentalis* 1, *Ceraclea* spp. 3, *Ceratopsyche alhedra* 3, *C. alternans* 3, *C. bronja* 5, *C. morosa bifida*^b 6, *C. morosa morosa*^b 2, *C. recurvata* = *alternans*, *C. riola* = *alhedra*, *C. slossonae* 4, *C. sparna* 1, *C. walkeri* 1, *C. vexa* 3, *Cheumatopsyche* spp. 5, *Chimarra aterrima* 4, *C. feria* 1, *C. obscura* 4, *C. socia* 0, *Cyrnellus fraternus* 8*, *Diplectrona modesta* 0*, *Dolophilodes distinctus* 0, *Frenesia missa* 0*, *Glossosoma* spp. 0, *Glyphopsyche irrorata* 1*, *Goera stylata* 0*, *Helicopsyche borealis* 3, *Hesperophylax designatus* 3, *Hydatophylax argus* 2, *Hydropsyche arinale* 5, *H. betteni* 6, *H. bidens* 3, *H. cuanis* 6, *H. dicantha* 2, *H. leonardi* 0, *H. orris* 5*, *H. phalerata* 1*, *H. placoda* 3, *H. scalaris* 2, *H. simulans* 7, *Hydroptila* spp. 6, *Ironoquia* spp. 3, *Lepidostoma*, spp. 1, *Leucotrichia pictipes* 2, *Limnephilus* spp. 3, *Lype diversa* 2, *Macronema* = *Macrostemum*, *Macrostemum zebratum* 3, *Micrasema kluane* 1, *M. rusticum* 2, *M. wataga* 2, *Molanna* spp. 6*, *Mystacides sepulchralis* 4, *Nectopsyche* spp. 3, *Nemotaulius hostilis* 3, *Neophylax* spp. 3, *Neotrichia* spp. 2*, *Neureclipsis* spp. 7, *Nyctiophylax* spp. 5, *Ochrotrichia* spp. 4*, *Oecetis* spp. 8, *Oligotomis ocelligera* 2, *Onocosmoecus quadrinotatus* 2*, *Oxyethira* spp. 3*, *Parapsyche apicalis* 0*, *Phryganea* spp. 8*, *Phycotropus placidus* 5*, *Platycentropus* spp. 4, *Polycentropus* spp. 6, *Potamyia flava* 5*, *Protophila* spp. 1, *Pseudostenophylax uniformis* 0*, *Psilotreta indecisa* 0*, *Psychoglypha subborealis* 0*, *Psychomyia flavida* 2, *Ptilostomis* spp. 5, *Pycnopsyche* spp. 4, *Rhyacophila brunnea* 0*, *R. fuscula* 0*, *R. lobifera* 4*, *R. vibox* 2*, *Setodes* spp. 2, *Stactobiella* spp. 2*, *Symphitopsyche* = *Ceratopsyche*, *Triaenodes* spp. 6, *Wormaldia moestus* 0*.

MEGALOPTERA

Chauliodes pectinicornis 4*, *C. rastricornis* 4*, *Corydalus cornutus* 6, *Nigronia serricornis* 0, *Stalis* spp. 4.

LEPIDOPTERA

Nymphula spp. 7*, *Petrophila* (= *Paragyraetis*) spp. 5, *Parapoynx* spp. 5.

COLEOPTERA

Ancyronyx variegata 6, *Dubiraphia bivittata* 8*, *D. minima* 5, *D. quadrinotata* 5, *D. vittata* 6, *Dubiraphia* larvae 6, *Ectopria* spp. 5, *Helichus lithophilus* 5*, *H. striatus* 5, *Macronychus glabratus* 4, *Microcylloepus pusillus* 3*, *Optioservus fastiditus* 4, *O. trivittatus* 2, *Optioservus* larvae 4, *Psephenus herricki* 4, *Stenelmis* spp. 5.

DIPTERA—except Chironomidae

Antocha spp. 3, *Atherix variegata* 2, *Bezzia* spp. 6, *Blepharicera tenuipes* 0*, *Chaoborus* spp. 8*, *Chrysogaster* spp. 10*, *Chrysops* spp. 6, *Cnephia dacotensis* 0*, *Culicoides* spp. 10, *Dicranota* spp. 3, *Dolichopodidae* 4, *Ectemnia* spp. 1*, *Empididae* 6, *Ephydriidae* 6*, *Erioptera* spp. 7, *Eristalis* spp. 10*, *Eusimulium* = *Simulium*, *Helius* spp. 4*, *Hesperoconopa* spp. 1, *Hexatoma* spp. 2, *Limnophila* spp. 3, *Limonia* spp. 6*, *Monohalea* spp. 6*, *Muscidae* 6, *Nilobezzia* spp. 6*, *Palpomyia* spp. 6, *Pedicia* spp. 6*, *Pericoma* spp. 4*, *Pilaria* spp. 7, *Probezzia* spp. 6, *Prosimulium fuscum* 6*, *P. gibsoni* 6*, *P. magnum* 1*, *P. mixtum* 3, *P. mysticum* 2*, *Pseudolimnophila* spp. 2, *Psychoda*

spp. 10, *Simulium aurium* 7*, *S. corbis* 2*, *S. croxtoni* 1*, *E. euryadmiculatum* 2*, *S. fibrinflatum* 6*, *S. jenningsi* 4, *S. johannseni* 0*, *S. latipes* 2*, *S. luggeri* 5*, *S. pictipes* 4*, *S. rugglesi* 5*, *S. tuberosum* 4, *S. venustum* 5, *S. verecundum* 6, *S. vittatum*^c, *Sphaeromais longipennis* 6*, *Stegopterna mutata* 5, *Tabanus* spp. 5*, *Tipula* spp. 4.

DIPTERA—Chironomidae

Ablabesmyia spp. 8, *Acricotopus* spp. 10, *Brillia* spp. 5, *Cardiocladius* spp. 5, *Chaetocladius* spp. 6, *Chironomus* spp. 10, *Cladopelma* spp. 9*, *Cladotanytarsus* spp. 7, *Clinotanytus* spp. 8, *Conchapelopia* spp. 6, *Cordites* spp. 4*, *Corynoneura* spp. 7, *Cricotopus* spp. 7, *Cryptochironomus* spp. 8, *Cryptotendipes* spp. 6, *Demicryptochironomus* spp. 8*, *Diamesa* spp. 5, *Dicotendipes* spp. 8, *Diplocladius* spp. 8*, *Djalmabatista* spp. 3*, *Einfeldia* spp. 9*, *Endochironomus* spp. 10, *Epoicocladius* spp. 4, *Eukiefferiella* spp. 8, *Glyptotendipes* spp. 10, *Guttipelopia* spp. 5*, *Harnischia* spp. 8, *Heterotrissocladius* spp. 0*, *Hydrobaenus* spp. 8, *Kiefferulus* spp. 10*, *Labrundinia* spp. 7*, *Larsia* spp. 6, *Limnophyes* spp. 8, *Microcricotopus* spp. 5*, *Micropsectra* spp. 7, *Microtendipes* spp. 6, *Monodiamesa* spp. 7*, *Nanocladius* spp. 3, *Natarsia* spp. 8, *Nilotanytus* spp. 6, *Nilothauma* spp. 2, *Odontomesa* spp. 4, *Orthocladius* spp. 6, *Pagastia* spp. 1, *Parachironomus*, spp. 10, *Paracladopelma* spp. 7*, *Paralauterborniella* spp. 8, *Parametriochemus* spp. 5, *Paraphaenocladius* spp. 4*, *Paratanytarsus* spp. 6, *Paratendipes* spp. 8, *Pentaneura* spp. 6, *Phaenopsectra* spp. 7, *Polypedilum* spp. 6, *Potthastia* spp. 2, *Procladius* spp. 9, *Prodiamesa* spp. 3, *Psectrocladius* spp. 8*, *Psectrotanytus* spp. 10, *Pseudochironomus* spp. 5, *Pseudorthocladius* spp. 0*, *Pseudosmittia* spp. 6*, *Rheocricotopus* spp. 6, *Rheotanytarsus* spp. 6, *Saetheria* spp. 4, *Smitia* spp. 6*, *Stempellina* spp. 2, *Stempellinella* spp. 4, *Stenochironomus* spp. 5, *Stictochironomus* spp. 9, *Sympotthastia* spp. 2, *Synorthocladius* spp. 2*, *Tanytus* spp. 10, *Tanytarsus* spp. 6, *Thienemanniella* spp. 6, *Tribelos* spp. 5, *Tvetenia* spp. 5, *Xenochironomus* spp. 0*, *Xylotopus par* 2*, *Zalutschia* spp. 7*, *Zavrelimyia* spp. 8.

AMPHIPODA AND ISOPODA

Asellus intermedius 8, *A. racovitzai* 8, *Crangonyx gracilis* 8, *C. pseudogracilis* 8*, *Gammarus pseudolimnaeus* 4, *Hyalolella azteca* 8.

^aAn asterisk denotes decreased reliability because collections were made in less than five of the 1979–80 study streams or because fewer than 10 individuals were collected.

^bAlthough Schefter and Unzicker (1984) synonymized *Ceratopsyche bifida* with *C. morosa*, we found that larvae of both forms can be readily identified (Schmude and Hilsenhoff 1986). Since *C. morosa morosa* form occurs only in northern Wisconsin in clean streams while *C. morosa bifida* form occurs statewide and often in organically enriched streams, I recommend that both forms be identified.

^c*Simulium vittatum* has a tolerance value of 7 unless three or more species with tolerance values of 2 or less make up at least 10% of the sample. Then the tolerance value is 4.