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USE OF ARTHROPODS TO EVALUATE WATER QUALITY OF STREAMS

Technical Bulletin No. 100

DEPARTMENT OF NATURAL RESOURCES
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ABSTRACT

Arthropods were used to evaluate the water quality of Wisconsin streams. The biotic index based upon arthropod samples is a sensitive and effective method, for it yields information on present quality and past perturbations. Every species was assigned an index value on the basis of collections made previously and in this study, for the purpose of calculating the biotic index. Water quality determinations were then made for 53 Wisconsin streams based on these values.

A sampling procedure for evaluating all streams in an area is given.

USE OF ARTHROPODS TO EVALUATE WATER QUALITY OF STREAMS

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INTRODUCTION

Since the effect of stream pollution is an alteration of the aquatic ecosystem, evaluation of that ecosystem is the logical way to detect pollution. When a stream is stressed, the segment of its fauna that cannot tolerate the stress will immediately disappear. Stream community structure is a result of both long-term environmental factors and critical conditions of short duration, and an experienced stream biologist with knowledge of community structures of normal and stressed streams often can evaluate the water quality of a stream with considerable accuracy after ony a few minutes' examination of its fauna. Yet most evaluations of water quality rely on physical and chemical determinations, which evaluate specific characteristics of the water only at the time of sampling and do not measure past short-term pollutional stresses.

Macroinvertebrates, and especially arthropods, are an important component of the aquatic ecosystem and have long been used to evaluate the water quality of streams. Among members of the aquatic ecosystem, they are probably best suited because they are numerous in almost every stream, are readily collected and identified, are not very mobile, and generally have life cycles of a year or more. These last two factors are important in assessing past perturbations of short duration, because once an arthropod is eliminated from the ecosystem it will not reappear until the next generation. Since Kolkowitz and Marsson (1909) first used arthropods to evaluate water quality, much has been written concerning their potential and the methods for collecting and evaluating them. The third seminar on Biological Problems in Water Pollution (Tarzwell 1965), a book by Hynes (1960), and another by Cairns and Dickson (1973) are among the more significant publications on the subject, but there have been many others.

In using arthropods to evaluate the water quality of streams, one of three methods has usually been employed to evaluate the data. The indicator species concept is the oldest, having been used by Kolkowitz and Marsson (1909), Richardson (1928) and several others more recently. Tolerance of species to pollution is usually designated by terms such as "tolerant", "facultative", or "intolerant", but there has been much disagreement in the placement of many species, as can be seen in the summary by Weber (1973). A stream is judged to be polluted or unpolluted by the presence or absence of species in each classification, but "intolerant" species may occasionally be found in polluted areas, especially when the water is cold, and "tolerant" species frequently appear in samples collected from the cleanest streams. Since the indicator species concept does not take into account numbers of each species and community structure, it lacks sensitivity and may give an erroneous picture of water quality.

It is a well-known fact that pollution of a stream reduces the number of species to be found in that stream, while frequently creating an environment that is favorable to a few. Thus, in a polluted stream there are usually large numbers of a few species, while in a clean stream there are moderate numbers of many species. On the basis of these facts, indexes that measure community diversity have frequently been used to evaluate the water quality of streams. The most widely used index is based on information theory as proposed by Margalef (1957) and later modified by Wilhm and Dorris (1968), and others. Wilhm (1970) considered index values above 3 to indicate unpolluted water and values less than 1 to indicate pollution. Every year new indexes are proposed and used to evaluate water quality, but all have serious drawbacks. Most important is that many small, cold streams have a naturally low diversity that is entirely unrelated to pollution. Small streams typically have lower diversity than larger streams in similar habitats and with similar substrates, which may lead to erroneous conclusions about their water quality if evaluated with diversity indexes. Another problem is that for greatest sensitivity, everything should be identified to species, and this is rarely possible. Consequently, diversity is usually calculated on the basis of generic identifications or "taxa".

A biotic index as proposed by Chutter (1972) appears to have great potential for quantitatively evaluating the arthropod fauna of streams in relation to water quality, but has not been used in North America. It evaluates community structure and makes use of the indicator species concept without placing undue emphasis on species that do not appear in significant numbers. To calculate a biotic index, each species is assigned a number based on collections in streams of known water quality. Chutter (1972) assigned a value of 0 to species found only in the cleanest streams, and a value of 10 to species that could inhabit extremely polluted waters, with appropriate intermediate values assigned to species found in streams between these two extremes. The index is calculated by multiplying the assigned value for each species by the number of individuals of that species that were found, summing the products, and dividing by the total number of individuals collected. Streams with values of 0-2 were classed as "clean unpolluted waters", 2-4 "slightly enriched waters", 4-7 "enriched waters" and 7-10 "polluted waters".

ARTHROPOD COMMUNITY STRUCTURE AS RELATED TO WATER QUALITY

Because previous experience suggested that stream arthropod communities could be readily recognized in the field by their dominant genera, a study was initiated in June 1972 to develop a rapid, objective method for evaluating water quality through arthropod community structure as recognized by dominant genera. Objectives were to classify arthropod communities, determine the relationship of these communities to the water quality of the streams in which they occurred, provide for easy recognition of communities in the field through their dominant genera, and develop a scheme by which the water quality of all streams in Wisconsin can be evaluated.

MATERIALS AND METHODS

Selection of Streams. Twenty-nine streams that were presumed to be undisturbed by human activities were selected for study during the first year. These streams were considered to be representative of Wisconsin streams with respect to size, geographical distribution, current, mineral content of the water, substrate, and water temperature. Twenty-four additional streams were selected for study in the second year because they were suspected of being polluted or disturbed to various degrees by human activities. Sampling site locations are given in Fig. 1 and Table 1.

Arthropod Samples. The arthropod fauna of each stream was sampled four times, once in early September, and again in November, early May, and late June. Two sampling procedures were used. A D-frame aquatic net was used to sample a riffle area by disturbing the riffle with one's feet and allowing dislodged insects to drift into the net held downstream. Samples collected in this manner were placed in a shallow white pan with a little water, and all arthropods that could be found in 20 minutes were removed with a curved forceps and preserved in 70% ethanol for identification and enumeration. Larvae and nymphs less than 3 mm long were not collected. Abundant species were not collected exhaustively; only enough individuals of any recognizable genus to assure an arbitrary limit of 25 were collected. If the sampling site had no riffle, a rock or gravel run was sampled. In the absence of rocks or gravel, snags and debris were sampled.

In addition to net samples, 2 artificial substrate samplers (Hilsenhoff 1969) were placed in each stream at a point where the current was rapid and the water deep enough to cover the samplers. Samplers were allowed at least six weeks to become colonized, and insects and debris from the samplers were removed as previously described (Hilsenhoff 1969) and preserved in 70% ethanol for sorting and enumeration in the laboratory.

Physical and Chemical Measurements. Stream temperatures were recorded at every visit and maximum summer water temperatures were measured with a maximum-minimum thermometer. Maximum-minimum thermometers were also placed in streams that did not freeze in winter to determine minimum winter temperatures. Water samples were collected for physical and chemical analysis during low flow periods in late summer and again in January. Collection of water samples after summer rains or winter thaws was avoided. Water samples were refrigerated and returned for analysis of 5-day biochemical oxygen demand (B.O.D.), total suspended solids, total nitrogen, total phosphorus, total alkalinity,

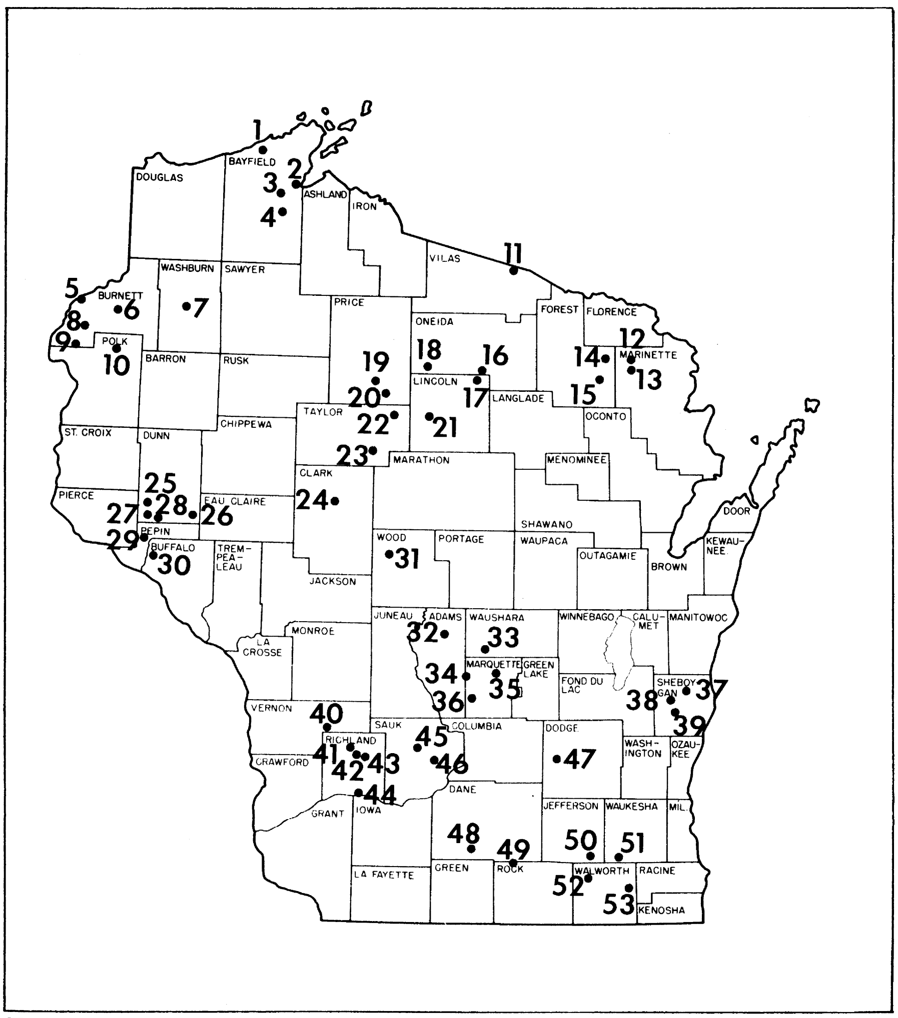

FIGURE 1. *Locations of the 53 sampling sites listed in Table 1*

TABLE 1. *Location of sampling sites in Figure 1.*

Site No.	Stream	County	Town	Range	Section*	Sampling Site**
1.	E. F. Cranberry R.	Bayfield	50-N	7-W	29N	Above wooden bridge
2.	Whittlesey Cr.	Bayfield	48-N	5-W	34E	Above School House Road
3.	Pine Cr.	Bayfield	47-N	6-W	13	Above N. Br. Fish Cr.
4.	White R.	Bayfield	46-N	6-W	25E	Above Highway 63
5.	St. Croix R.	Burnett	40-N	18-W	30	Norwegian Point
6.	Clam R.	Burnett	39-N	16-W	21W	Above Highway 35
7.	Namekagon R.	Washburn	40-N	11-W	31	Above Highway 63
8.	Wood R.	Burnett	38-N	18-W	21N	Above and below Highway 70
9.	Trade R.	Burnett	37-N	19-W	36	Below town road
10.	McKenzie Cr.	Polk	36-N	16-W	1N	Above Highway W
11.	Wisconsin R. #1	Vilas	42-N	10-E	11	Below Highways 32 and 45
12.	Sidney Cr.	Marinette	37-N	17-E	24	Below town road
13.	Chemical Cr.	Marinette	36-N	17-E	1	Above Goodman Park Road
14.	Armstrong Cr.	Forest	37-N	16-E	27	Above Highway 8
15.	Peshtigo R.	Forest	36-N	16-E	33	Below Swede Road
16.	Wisconsin R. #2	Oneida	36-N	8-E	34N	Below Hat Rapids bridge
17.	Wisconsin R. #3	Lincoln	35-N	8-E	4	Above Camp 10 Ski Area
18.	Little Somo R.	Oneida	36-N	4-E	24	Above town road
19.	Little Jump R.	Price	35-N	1-E	24W	Above Highway 13
20.	N. Br. Levitt Cr.	Price	34-N	2-E	16	Above town road
21.	Newood R.	Lincoln	33-N	5-E	30	Off town road
22.	Copper Cr.	Taylor	33-N	3-E	30E	Above town road
23.	Little Black R.	Taylor	30-N	1-E	1	Above Highway 13
24.	Poplar R.	Clark	27-N	2-W	10	Above Highway 73
25.	Eau Galle R. #1	Dunn	26-N	14-W	11	Below town road off Hwy. C
26.	Rock Cr.	Dunn	26-N	11-W	15	Above town road
27.	Missouri Cr.	Dunn	26-N	14-W	26E	Above town road
28.	Eau Galle R. #2	Dunn	26-N	13-W	31	Below Eau Galle Dam
29.	Arkansas Cr.	Pepin	25-N	14-W	24	Above town road
30.	Spring Cr.	Buffalo	24-N	13-W	18	Below Highway 25
31.	Yellow R.	Wood	24-N	3-E	7E	Above Highway 13
32.	Big Roche a Cri	Adams	19-N	6-E	12N	Above and below town road
33.	Mecan R. #1	Waushara	18-N	9-E	16	Above Highway 21
34.	Lawrence Cr.	Marquette	17-N	8-E	31W	Below county line road
35.	Mecan R. #2	Marquette	17-N	10-E	28	Above Highway 22
36.	Neenah Cr.	Marquette	15-N	8-E	20	At town road
37.	Sheboygan R.	Sheboygan	16-N	22-E	31N	Above Highway JJ
38.	Mullet R.	Sheboygan	15-N	21-E	8	Below Highway J
39.	Onion R.	Sheboygan	14-N	21-E	4S	Above Highway U
40.	Kickapoo R.	Vernon	13-N	2-W	32	Just below Bear Cr.
41.	Pine R. #1	Richland	12-N	1-E	17	Below Highway 80
42.	Pine R. #2	Richland	12-N	1-E	27	Along Highway 80
43.	Milancthon Cr.	Richland	12-N	1-E	27	Above Highway C
44.	Wisconsin R. #4	Richland	8-N	1-E	5	At public boat landing
45.	Narrows Cr.	Sauk	12-N	5-E	31	Along Highway 154
46.	Otter Cr.	Sauk	11-N	6-E	33W	At Stone's Pocket Road
47.	Beaver Dam R.	Dodge	11-N	14-E	30N	At Highway S
48.	Sugar R.	Dane	5-N	8-E	3	Above road in Paoli
49.	Badfish Cr.	Rock	4-N	11-E	5	Below Highway 59
50.	Steel Brook	Jefferson	5-N	15-E	25	At Highway 59
51.	Jericho Cr.	Waukesha	5-N	17-E	25	Above Highway 99
52.	Bluff Cr.	Walworth	4-N	15-E	23	Below Highway P
53.	Sugar Creek	Walworth	3-N	18-E	14	Below town road

*N means at north section line, E at east section line, W at west section line, and S at south section line.

**Above indicates upstream from and below indicates downstream.

hardness, chlorides, and pH. Maximum current, stream width, and stream substrates were recorded for all streams during a period of low flow in August 1973. Dissolved oxygen (D.O.) was recorded at each summer visit with a YSI portable D.O. meter, and on a very warm night in the summer of 1975 the D.O. of each stream was recorded sometime between sunrise and two hours before sunrise to give an estimate of minimum summer D.O. levels for each stream.

Sample Size. In June 1973 the samples from 23 streams were divided into two parts to determine how sample size affected results and if the 20-minute picking time was unnecessarily long. Arthropods removed from the sample in the first 5 minutes were kept separate from those removed in the remaining 15 minutes of the 20-minute picking time.

RESULTS AND DISCUSSION

On several occasions, one or both of the artificial substrate samplers were tipped over or missing due to interference by the curious public. Because of the large amount of missing data in the sampler samples, only data collected in net samples were analyzed. Numbers and species of arthropods collected by samplers usually were similar to those collected with a net from the same stream. All arthropods collected in the net samples were identified to species if possible, but species could not be identified in many genera*.

When efforts were made to classify the community structure of the study streams it was found that among this diverse group of streams almost none had similar community structures. It would have been impossible to devise a simple classification of arthropod communities that could be readily recognized by their dominant species, so the initial objective of this study to develop a rapid, objective method to evaluate water quality through field recognition of arthropod community structure was abandoned. However, the vast amount of data that had been collected provided an excellent opportunity to evaluate diversity index ($\bar{d}$) and biotic index (B.I.) values as a means for evaluating water quality. In calculating biotic index or diversity index values no more than 25 individuals in each genus were used from any collection since under the initial objectives of this study no effort had been made to collect more than that number of individuals in any genus, except when the genus could not be recognized in the field. If all individuals had been used, unequal weight would have been given to species and genera that could not be recognized in the field and were therefore collected in greater numbers.

Because it was not possible to identify many species, the diversity index was calculated for each stream on the basis of numbers of individuals in each genus using the formula:

$$\bar{d} = \frac{N \log_2 N - \Sigma n_i \log_2 n_i}{N}$$

where N is the total number of arthropods in the sample and n_i is the number of individuals in each genus. The arbitrary limit of 25 individuals in each genus caused calculated diversity values to be high.

A biotic index similar to one proposed by Chutter (1972) was also calculated for each stream, but a scale of 0 to 5 was used instead of 0 to 10. Every species was assigned an index value (Append. I) on the basis of collections made previously and in this study, 0 values being assigned to species collected only in unaltered streams of very high water quality and values of 5 assigned to species known to occur in severely polluted or disturbed streams. Intermediate values were assigned to species known to occur in streams with various degrees of disturbance or pollution. When species could not be identified, genera were assigned values instead. The biotic index was calculated from the formula:

$$B.I. = \frac{\Sigma n_i a_i}{N}$$

where n_i is the number of individuals in each species or genus, a_i is the index value for that species or genus (Append. I), and N is the total number of individuals in the sample. Since no effort had been made to collect more than 25 individuals in each genus, when two or more species from the same genus totaled more than 25, the percentage of the total in that genus was multiplied by 25 times the index value to obtain the number used to calculate the biotic index.

In Table 2 the streams are arranged according to biotic index values, which are compared with physical and chemical parameters, and in Table 3 these parameters are compared by rank correlation analysis using the formula:

$$r' = 1 - \frac{6(\Sigma d^2)}{n(n^2-1)}$$

where d is the difference in rank and n is the total number of ranked observations. B.O.D., lowest D.O., suspended solids, total nitrogen, total chlorides, and lowest maximum temperature all had a highly significant correlation with biotic index values. It is also evident in Table 2 that all streams that had no known perturbations had very low biotic index values and were considered clean streams.

Biotic index values for each season are compared in Table 4 with the value calculated from totals of the four seasonal samples. Values for June and September, when water temperatures were highest, average higher than May and November values, when water temperatures were colder. Biotic index values are also compared with ranked diversity index values in Table 4, and although there is a highly significant rank correlation of these two values, many of the cleanest streams such as Sidney Creek, Whittlesey Creek, Spring Creek, East Fork Cranberry River, and Pine Creek have relatively low diversity index values and would have been judged of relatively low water quality if the diversity index had been used as the criterion. These are all small, cold streams that typically have a restricted fauna. When diversity index values were compared with physical and chemical parameters by rank correlation (Table 3), there was a highly significant correlation only with alkalinity, which is not normally considered as contributing to deterioration of water quality, but is known to enhance production in streams.

The results show that the diversity index does not accurately assess the water quality of streams, ranking some of the cleanest undisturbed wilderness streams with moderately enriched or polluted streams. The

*A list of species collected from each stream is available from the author.

TABLE 2. *Physical and chemical characteristics of Wisconsin streams arranged by Biotic Index (B.I.) values.*

Site No.	Stream	B.I.	Temp.°C	Width Meters	Stream Substrate*	Current M/sec	Sampling Site Substrate**	Perturbations***	Low D.O. ppm	5-day BOD ppm	pH	Tot. Alk. ppm	Tot. N ppm	Tot. P ppm	Tot. Solids ppm	Tot. Cl ppm
1	E. F. Cranberry R.	0.95	0.0-15.0	27	R	0.47	RRi	none	10.1	1.0	6.8-6.9	51	0.17	0.03	82	0.0
2	Whittlesey Cr.	1.00	1.0-16.5	5	Sa, R	0.40	R, D	none	10.2	1.0	6.5-7.1	71	0.18	0.02	89	0.5
3	Pine Cr.	1.04	0.0-16.5	11	R, Sa	0.43	RRi	none	9.8	0.7	7.2	64	0.14	0.03	86	0.5
46	Otter Cr.	1.07	0.5-20.5	5	R	0.30	RRi	none	8.2	1.7	6.3-7.3	16	0.30	0.03	62	0.0
18	Little Somo R.	1.19	0.0-24.0	5	G, Sa	0.55	GRi	none	6.1	1.6	6.8-7.1	25	0.89	0.04	94	0.0
30	Spring Cr.	1.21	0.0-21.5	3	Sa, R	0.54	R	PCU	8.4	1.5	7.6-7.7	137	2.84	0.11	241	4.0
10	McKenzie Cr.	1.22	0.0-26.0	8	Sa, Si, G	0.17	G	none	8.1	2.0	6.9-7.1	117	0.43	0.05	150	0.0
12	Sidney Cr.	1.29	0.0-18.0	5	Sa, R	0.73	RRi	none	9.2	1.0	7.5-7.6	128	0.63	0.01	167	0.5
19	Little Jump R.	1.37	0.0-25.5	11	G, R, Sa	0.35	GRi	PCU	7.0	1.6	6.9-7.1	51	0.99	0.07	119	0.0
34	Lawrence Cr.	1.39	4.5-18.0	2	Sa, G	0.30	G	none	10.9	1.5	7.9-8.0	158	1.78	0.03	189	0.0
7	Namekagon R.	1.44	0.0-28.0	21	R	0.66	RRi	none	7.5	2.0	6.7-7.2	67	0.49	0.03	103	5.0
33	Mecan R. #1	1.49	0.5-25.0	8	Sa, G, R	0.35	GRi	none	8.6	1.3	8.1-8.2	173	1.86	0.03	202	0.0
14	Armstrong Cr.	1.55	0.0-23.0	8	R	0.42	RRi	none	6.9	1.5	6.9-7.2	107	0.71	0.02	167	0.0
15	Peshtigo R.	1.56	0.0-23.5	18	R	0.64	RRi	none	7.8	1.0	6.9-7.5	96	0.68	0.02	143	0.0
20	N. Br. Levitt Cr.	1.62	0.0-20.0	2	Sa, G	0.30	GRi	none	–	1.0	6.7-6.8	38	0.81	0.09	102	0.0
5	St. Croix R.	1.65	0.0-28.5	(91)[1]	G, R	0.35	G	none	6.7	2.0	6.7-7.7	73	0.81	0.03	121	0.0
32	Big Roche a Cri	1.69	0.0-26.0	10	Sa, R	0.51	R	none	8.3	1.6	6.9-7.7	108	1.71	0.03	167	0.5
21	Newood R.	1.70	0.0-28.0	15	R	0.42	RRi	none	4.8	1.5	6.6-6.8	37	1.04	0.08	115	0.0
13	Chemical Cr.	1.73	0.5-16.0	5	Sa, Si, R	0.34	R	UE	10.0	2.0	7.8-7.9	126	0.77	0.08	186	3.0
4	White R.	1.82	0.0-23.0	15	Sa, G	0.45	G	none	7.9	1.2	7.1-7.3	78	0.33	0.03	120	0.0
26	Rock Cr.	1.86	0.5-23.5	11	G, R	0.39	GRi	none	7.5	1.8	6.7-7.0	64	1.70	0.23	135	2.0
51	Jericho Cr.	1.98	0.0-27.5	5	G, R	0.32	GRi	PCU	7.9	1.9	8.2-8.3	286	4.32	0.02	423	20.5
53	Sugar Cr.	1.98	0.5-27.5	9	G, R	0.73	GRi	PCU	7.0	2.0	7.9-8.0	292	3.26	0.11	498	16.0
31	Yellow R.	2.03	0.0-28.5	24	R, G	0.36	GRi	PCU	5.9	0.7	7.3-7.9	96	1.18	0.06	153	7.0
24	Poplar R.	2.05	0.0-29.0	20	R, G	0.36	GRi	PCU	6.8	1.2	8.0-8.3	126	1.47	0.33	193	13.0
38	Mullet R.	2.09	1.0-26.5	8	R, G	0.48	RRi	PCU	8.2	2.3	8.0-8.2	282	1.53	0.17	391	9.5
43	Milancthon Cr.	2.13	1.0-22.0	5	G	0.79	GRi	PCU	8.2	1.9	7.4-8.3	221	1.28	0.06	245	1.5
25	Eau Galle R. #1	2.13	0.0-26.5	19	Sa	0.27	Sn, G	none	8.5	1.8	7.4-7.8	233	1.25	0.13	287	4.0
42	Pine R. #2	2.26	1.5-24.0	9	G, Si, Sa	0.38	GRi	PC	6.8	2.3	8.0-8.1	195	1.31	0.04	249	3.0
35	Mecan R. #2	2.28	0.0-25.0	21	Sa, Si, R	0.20	R, D	PCU	7.9	1.3	7.6-8.2	151	1.64	0.03	191	0.0
22	Copper Cr.	2.35	0.0-27.0	7	G, Si	0.46	GRi	PCU	4.2	1.3	6.6-7.4	32	1.43	0.10	105	0.0
23	Little Black R.	2.43	0.0-28.5	12	R, G	0.15	GRi	PCU	4.5	1.0	7.2-7.6	70	1.24	0.04	140	1.0
39	Onion R.	2.45	0.0-23.5	5	Sa, G, R	0.49	GRi	PCU	8.4	1.2	8.2	271	2.12	0.05	360	9.0
27	Missouri Cr.	2.45	0.0-25.0	5	Si, G, Sa	0.40	GRi	PC	7.5	1.2	8.2	272	1.32	0.10	326	2.0
36	Neenah Cr.	2.48	0.0-25.0	10	Sa, R	0.32	RRi	PCU	7.5	0.9	7.9-8.1	169	1.11	0.03	205	0.0
40	Kickapoo R.	2.49	0.0-24.5	25	G, Si	0.46	GRi	PCU	7.1	2.6	7.8-8.2	228	1.01	0.07	298	4.0
6	Clam R.	2.49	0.0-28.0	27	Sa, G	0.30	G, D	PCU	5.7	1.9	6.9-7.9	104	0.56	0.06	139	0.0
44	Wisconsin R. #4	2.53	0.5-29.0	320	R, Sa, Si	0.61	R	I, PCU	6.5	4.6	7.4-8.1	84	1.29	0.10	177	6.0
29	Arkansas Cr.	2.62	0.0-23.5	8	Sa, G	0.20	GRi	UE, PCU	8.6	0.9	7.9-8.0	267	1.48	0.08	331	2.0
41	Pine R. #1	2.67	0.0-28.0	7	G, Sa, Si	0.50	GRi	CF, PCU	6.2	2.4	8.0-8.3	200	1.15	0.04	247	3.0
45	Narrows Cr.	2.72	0.0-25.0	14	R, G, Si	0.52	RRi	PCU	5.9	2.1	7.8-8.0	211	2.01	0.14	285	9.0
48	Sugar R.	2.79	0.5-25.5	11	R, G, Si	0.48	GRi	D, PCU	8.4	2.9	8.1-8.2	258	4.53	0.23	385	15.0
37	Sheboygan R.	2.90	0.0-30.5	24	G, R	0.37	GRi	PCU	5.0	3.1	8.0-8.5	309	2.28	0.06	463	22.0
50	Steel Brook	2.91	0.0-26.0	5	Si, G	0.16	G, D	PC	8.4	2.1	8.1-8.2	277	1.53	0.06	409	7.0
52	Bluff Cr.	2.96	6.5-21.0	4	Si, G, R	0.48	RRi	PCU	6.2	1.8	7.4-8.0	282	1.05	0.02	348	4.0
9	Trade R.	3.03	0.0-28.0	11	R, Sa	0.45	RRi	D	2.2	1.8	7.6-7.7	116	1.30	0.05	163	3.0
11	Wisconsin R. #1	3.14	0.0-25.0	14	Sa, Si, G	0.28	G	EL	7.8	3.9	6.9	37	0.63	0.04	70	2.0
28	Eau Galle R. #2	3.25	0.5-26.5	20	R, G	0.55	GRi	D	8.2	1.9	8.0	222	1.37	0.06	265	3.0
8	Wood R.	3.36	0.0-29.5	9	Si	0.14	D	PCU	5.5	1.2	7.4-7.8	111	0.71	0.02	140	2.0
17	Wisconsin R. #3	4.07	0.0-27.0	(76)	R, G	0.63	RRi	PM, D	2.6	5.2	6.8-7.3	26	1.29	0.07	95	2.0
47	Beaver Dam R.	4.25	0.5-29.0	14	G, R, Si	0.29	GRi	D, UE, PC	2.4	6.5	8.0-8.1	243	3.17	1.55	443	29.0
16	Wisconsin R. #2	4.28	0.0-26.5	73	R, G, Sa	0.40	R, G	PM, D	3.8	3.8	7.0-7.2	26	1.26	0.07	94	1.0
49	Badfish Cr.	4.29	3.0-26.0	15	G, R, Sa	0.36	GRi	UE	3.2	21.5	7.7-7.9	330	5.77	2.40	717	152.0

*Stream Substrate: R=rocks, Sa= sand, G=gravel, Si=silt.

**Sampling Site Substrate: R=rocks, Ri=riffle, D=debris, G=gravel, Sn=snags.

***Perturbations: PCU=pasturing cattle upstream, UE=urban effluent, PC=pasturing cattle, I=industry, CF=cheese factory, D=dam, EL=eutrophic lake, PM=paper mill.

[1] Widths in parentheses are estimates.

biotic index, however, did an excellent job of ranking streams according to water quality and should prove to be a reliable tool. The reliability of the biotic index is dependent upon several factors, and assignment of index values to each species is the most important. To an extent, assignment of values has been subjective, being based upon previous experience and knowledge. Errors in judgment are most likely to be made with rarer species with which there has been little previous experience, but because these species are rare, their impact upon calculations of a biotic index is always minimal. It is common species that have the greatest impact upon calculated index values.

Species in several genera can not be identified and index values must be assigned to genera. This affects the sensitivity of the index, because the value assigned to a genus is equal to the value of its most pollution-tolerant species. The inability to identify species in some common genera such as *Hydropsyche* causes abnormally high biotic index values for cleaner streams. These clean streams probably would have mostly intolerant species of *Hydropsyche* with low index values had it been possible to assign values to species. When biotic index values were computed using only generic identifications and assigning a value to each genus equal to that of its most tolerant species, biotic index values averaged 42% higher for the ten cleanest streams, 9% higher for the middle eleven streams and only 1% more for the ten most polluted or disturbed streams. This demonstrates that species identification enhances the sensitivity of the biotic index, and is essential mostly in the detection of minor degrees of pollution or disturbance. It is apparently unnecessary for the detection of significant disturbance or pollution.

The greatest drawback to the use of the biotic index is the time involved in sorting and identification. This is closely tied to sample size; smaller samples take less time to sort and identify and perhaps give sufficient information. Arthropods collected in this study could normally be sorted and identified to genus in less than one hour, but species identification required about another hour. The time factor is difficult to evaluate because it depends upon the skill and knowledge of the person doing the sorting and identification.

Results of the sample size study are reported in Table 5. Generally about half of the arthropods collected were found in the first 5 minutes of picking. Comparison of biotic index values for 5-minute and 20-minute samples showed substantial differences in some streams but in most the difference was not great. Larger, more conspicuous organisms such as large stoneflies, active mayflies, dragonflies, and large amphipods were mostly collected in the first 5-minute period, while smaller, less conspicuous insects such as riffle beetles, chironomids, small secretive mayflies, and some caddisflies were more likely to be found in the last 15 minutes of the 20-minute picking period.

It appears that samples picked for 20 minutes are larger than necessary, and that while a 5-minute picking time is perhaps inadequate, 10 minutes would be sufficient. About 100 arthropods appear to constitute an adequate sample (Table 5), so another method would be to collect until the first 100 arthropods have been removed from the sample, exercising care so as not to remove only the largest arthropods and bias the sample. To avoid bias, samples that obviously contain more than 100 arthropods can be divided into subsamples and arthropods removed from each subsample until 100 have been collected. Sample sizes less than 100 (Table 5), even after 20 minutes of picking, were usually the result of the arbitrary limit of 25 placed on the number of each genus that was collected. Only in extremely polluted waters might there be difficulty in securing 100 specimens, and a time limit could be used in these extreme situations.

TABLE 3. *Rank correlation of the biotic index and generic diversity (d) with various physical and chemical parameters in 53 Wisconsin streams.*

Physical or Chemical Parameter	Biotic Index	Diversity
Biochemical Oxygen Demand (5-day)	.515**	.213
Dissolved Oxygen (lowest recorded)	.548**	-.012
Suspended Solids	.464**	.399*
Total Nitrogen	.432**	.208
Total Phosphorus	.363*	.111
Total Alkalinity	.388*	.436**
Total Chlorides	.531**	.377*
Maximum Current	-.112	.195
Lowest Maximum Temperature	.516**	.300*

** significant correlation at the 1% level
* significant correlation at the 5% level

TABLE 4. *Seasonal and yearly biotic index values for Wisconsin streams ranked by yearly values in order of decreasing pollution or disturbance and compared with diversity index values and ranking.*

Stream	County	Site No. (Fig. 1)	Biotic Index					Diversity	
			May	June	Sept.	Nov.	Year	index	rank
E. F. Cranberry R.	Bayfield	1	0.74	1.16	1.05	0.94	0.95	3.40	32
Whittlesey Cr.	Bayfield	2	1.12	1.46	0.90	0.28	1.00	2.87	45
Pine Cr.	Bayfield	3	1.24	1.29	1.19	0.25	1.04	3.51	30
Otter Cr.	Sauk	46	0.44	1.51	1.14	1.14	1.07	4.34	5
Little Somo R.	Oneida	18	0.50	2.00	1.72	0.82	1.19	4.02	12
Spring Cr.	Buffalo	30	0.79	1.28	1.66	1.33	1.21	3.34	36
McKenzie Cr.	Polk	10	0.84	1.40	1.70	1.22	1.22	4.46	3
Sidney Cr.	Marinette	12	1.09	1.49	1.59	1.31	1.29	2.62	51
Little Jump R.	Price	19	0.89	1.58	1.79	1.06	1.37	4.06	11
Lawrence Cr.	Marquette	34	1.08	1.42	1.73	1.35	1.39	3.81	17
Namekagon R.	Washburn	7	1.30	1.40	1.63	1.36	1.44	4.36	4
Mecan R. #1	Waushara	33	1.53	1.51	1.64	1.27	1.49	3.91	15
Armstrong Cr.	Forest	14	1.22	1.65	2.02	1.29	1.55	3.69	23
Peshtigo R.	Forest	15	1.43	1.46	2.22	–	1.56	3.76	20
N. Br. Levitt Cr.	Price	20	1.41	1.43	1.76	2.03	1.62	3.98	14
St. Croix R.	Burnett	5	1.24	1.90	1.90	2.00	1.65	4.50	1
Big Roche a Cri	Adams	32	1.86	1.75	2.20	1.16	1.69	3.61	25
Newood R.	Lincoln	21	1.58	1.85	1.95	1.43	1.70	4.18	7
Chemical Cr.	Marinette	13	1.48	2.06	1.58	1.68	1.73	3.26	39
White R.	Bayfield	4	2.00	1.61	2.10	1.68	1.82	4.07	10
Rock Cr.	Dunn	26	1.56	2.39	2.05	1.42	1.86	3.72	22
Jericho Cr.	Waukesha	51	1.87	2.66	1.92	1.42	1.98	3.85	16
Sugar Cr.	Walworth	53	1.33	2.59	2.61	1.31	1.98	3.76	20
Yellow R.	Wood	31	2.13	2.06	2.16	1.79	2.03	4.11	8
Poplar R.	Clark	24	1.95	2.20	1.98	2.04	2.05	4.32	6
Mullet R.	Sheboygan	38	1.90	2.28	2.44	1.81	2.09	3.81	17
Milancthon Cr.	Richland	43	1.81	2.43	2.57	1.65	2.13	3.54	27
Eau Galle R. #1	Dunn	25	2.28	2.33	2.51	1.50	2.13	3.24	40
Pine R. #2	Richland	42	2.01	2.38	2.26	2.71	2.26	3.54	27
Mecan R. #2	Marquette	35	2.03	2.25	2.70	2.21	2.28	3.77	19
Copper Cr.	Taylor	22	2.18	2.21	2.82	2.16	2.35	4.11	8
Little Black R.	Taylor	23	2.36	2.25	2.59	2.36	2.43	4.00	13
Missouri Cr.	Dunn	27	2.64	2.51	2.44	2.31	2.45	2.68	49
Onion R.	Sheboygan	39	2.51	2.72	2.16	2.53	2.45	3.45	30
Neenah Cr.	Marquette	36	2.39	2.43	2.53	2.62	2.48	3.28	38
Clam R.	Burnett	6	2.27	2.84	2.22	2.54	2.49	4.47	2
Kickapoo R.	Vernon	40	2.07	2.72	2.47	2.52	2.49	3.34	36
Wisconsin R. #4	Richland	44	2.30	2.23	2.98	2.97	2.53	3.55	26
Arkansas Cr.	Pepin	29	2.27	2.78	2.85	2.56	2.62	2.83	46
Pine R. #1	Richland	41	2.59	2.73	2.87	2.43	2.67	3.20	41
Narrows Cr.	Sauk	45	2.63	2.96	2.38	2.63	2.72	3.17	42
Sugar R.	Dane	48	2.62	3.19	2.67	2.72	2.79	3.39	34
Sheboygan R.	Sheboygan	37	2.92	2.80	2.92	2.98	2.90	2.88	44
Steel Brook	Jefferson	50	3.04	2.99	2.89	2.76	2.91	3.42	31
Bluff Cr.	Walworth	52	2.77	3.03	3.03	3.00	2.96	2.93	43
Trade R.	Burnett	9	3.00	2.97	3.07	3.05	3.03	3.50	29
Wisconsin R. #1	Vilas	11	3.25	3.62	2.60	2.86	3.14	3.63	24
Eau Galle R. #2	Dunn	28	3.50	3.05	3.18	3.34	3.25	2.79	47
Wood R.	Burnett	8	3.03	3.24	3.82	3.55	3.36	3.39	34
Wisconsin R. #3	Lincoln	17	3.81	4.50	4.10	4.06	4.07	2.78	48
Beaver Dam R.	Dodge	47	4.51	4.74	4.00	4.18	4.25	2.66	50
Wisconsin R. #2	Oneida	16	4.52	4.80	3.73	4.64	4.28	2.06	53
Badfish Cr.	Rock	49	4.58	5.00	4.09	4.30	4.29	2.40	52
Average			**2.08**	**2.40**	**2.41**	**2.09**	**2.22**		

TABLE 5. *Comparison of numbers of arthropods and biotic index from 5-minute and 20-minute samples in June 1973.*

Stream	Numbers 5 Minutes	Numbers 20 Minutes	Percent in 5 Minutes	Biotic Index Value 5 Minutes	Biotic Index Value 20 Minutes	Biotic Index Value Difference
E. F. Cranberry R.	65	111	59	0.91	1.16	-0.25
Spring Cr.	81	107	76	1.26	1.28	-0.02
Pine Cr.	63	118	53	1.27	1.29	-0.02
Namekagon R.	133	201	66	1.41	1.40	+0.01
McKenzie Cr.	90	156	58	1.04	1.40	-0.36
Lawrence Cr.	94	126	75	1.18	1.42	-0.24
Peshtigo R.	74	143	52	1.64	1.46	+0.18
Whittlesey Cr.	37	84	44	1.46	1.46	0.00
Sidney Cr.	29	39	74	1.83	1.49	+0.34
Mecan R. #1	65	164	40	1.28	1.51	-0.23
Little Jump R.	87	181	48	1.49	1.58	-0.09
White R.	74	145	51	1.78	1.61	+0.17
Armstrong Cr.	57	116	49	1.75	1.65	+0.10
Big Roche a Cri	50	80	63	1.70	1.75	-0.05
St. Croix R.	44	106	42	1.95	1.90	+0.05
Wisconsin R. #4	21	66	32	2.10	2.23	-0.13
Mecan R. #2	67	110	61	2.46	2.25	+0.12
Mullet R.	56	137	41	2.54	2.28	+0.26
Eau Galle R. #1	64	95	67	2.31	2.33	-0.02
Rock Cr.	62	132	47	2.48	2.39	+0.09
Milancthon Cr.	75	144	52	2.16	2.43	-0.27
Clam R.	52	119	44	2.37	2.84	-0.47
Wisconsin R. #1	25	74	34	2.96	3.62	-0.66
Average	**64**	**120**	**53**	**1.80**	**1.86**	**-0.06**

MATERIALS AND METHODS

To determine how long it would take to evaluate the water quality of all streams in Wisconsin by sampling their arthropod fauna, a sampling scheme was devised whereby every stream would be sampled at the road nearest the point at which it flowed out of every township. Three transects were run along rows of townships, the starting points being selected at random. Transects were determined to run east or west at random, and any transect reaching the border of the state was reversed in a clockwise direction. Transects sampled were: Dane-Jefferson County, T6N, R11E east to T6N, R15E; Wood-Clark County T23N, R3E west to T23N, R3W; and Barron-Polk County T35N, R14W west to T35N, R19W and T36N, R20W east to R36N, R16W.

Each transect was sampled for an 8-hour period. Arthropods were collected from riffles with a D-frame aquatic net, picked from a white pan for 10 minutes, and preserved in 70% ethanol. In the absence of riffles, a gravel or rocky run, or debris was sampled. All samples were returned to the laboratory for identification and enumeration. Biotic index values were calculated for each stream to evaluate water quality, according to Table 6, which was developed as a result of this study of 53 Wisconsin streams.

RESULTS AND DISCUSSION

Water quality in each stream is evaluated in Tables 7-9 from biotic index values according to Table 6. The number of townships that can be covered in one day is mostly dependent on the number of streams present. In the Barron-Polk county transect 14 streams were sampled in 10 townships, while in the Dane-Jefferson County transect 14 streams were sampled in only 5 townships. From the three transects that were sampled, it appears that an average of 14 streams and 7 townships can be sampled in an 8-hour day so that it would take one person about 222 days to sample all streams in the state using this procedure. Sorting, enumeration, and identification of specimens would take at least an equal amount of time.

Finding a suitable sampling site was often a problem. Many streams had no riffles or other suitable substrate at the site of sampling, and inadequate samples resulted. A sampling procedure whereby 100 arthropods were collected at each site would have been superior. Very large and very small streams presented the greatest problems, and streams less than 1 meter wide should probably not be sampled. Some streams had no perceptible flow and were not sampled.

TABLE 6. *Water quality determination from biotic index values.*

Biotic Index*	Water Quality	State of the Stream
<1.75	Excellent	Clean undisturbed
1.75-2.25	Good	Some enrichment or disturbance
2.25-3.00	Fair	Moderate enrichment or disturbance
3.00-3.75	Poor	Significant enrichment or disturbance
> 3.75	Very Poor	Gross enrichment or disturbance

* Biotic index values are based on combined samples collected in late spring, early summer, late summer, and late autumn. For summer samples only, 0.18 would be subtracted from calculated biotic indexes for evaluation, and for spring and autumn samples 0.13 would be added to calculated values.

CONCLUSIONS AND RECOMMENDATIONS

An experienced aquatic biologist can make a fairly reliable judgment of the water quality of a stream by viewing a sample of its arthropod fauna in the field, but there is no simple procedure for quantifying this judgment in the field. Samples must be returned to the laborabory. The biotic index is a very sensitive and effective way to evaluate the water quality of streams on the basis of these arthropod samples, and its use should be promoted. It is more sensitive than physical and chemical procedures, and since several chemical and physical parameters usually have to be tested in each stream, it takes less time and is more economical. Finally, it will detect past perturbations, while physical and chemical analyses detect only present pollution and disturbance. Diversity indexes calculated from arthropod community structure should be used to evaluate water quality only under special circumstances, because small, cold, undisturbed streams have a naturally low diversity and would be judged as disturbed or polluted.

Many sampling procedures can be effectively used to collect arthropods from streams for evaluation of water quality by the biotic index. I recommend the following. Use a D-frame aquatic net to sample riffles by disturbing the substrate above the net and allowing dislodged arthropods to be washed into the net by the current. If riffles are absent, rock or gravel runs or debris may be similarly sampled. Place a sample containing about 100 arthropods in a shallow white pan containing a little water. When collecting the sample it is important to not collect significantly more than 100 arthropods because in large samples, larger and more easily captured arthropods will be most readily removed from the pan, creating a biased sample. Using a

TABLE 7. *Evaluation of water quality by biotic index values of the arthropod fauna in the Dane-Jefferson County transect, June 9, 1975.*

Stream	Number of Genera	Number of Individuals	Biotic Index	Water Quality
Door Creek	7	90	3.63	Poor
Yahara River	6	15	3.13	Fair
Mud Creek - Hwy. W	6	32	3.28	Poor
Mud Creek - Hwy. 73	9	69	3.20	Poor
Koshkonong Creek - Hwy. 18	7	67	2.99	Fair
Koshkonong Creek - Rockdale	10	78	2.99	Fair
Creek Hwy. C - East	6	85	3.12	Fair
Creek Hwy. C - West	5	65	2.62	Fair
Creek Hwy. J	8	104	3.16	Fair
Creek off Hwy. G	7	25	2.60	Fair
Deer Creek - West	3	25	2.84	Fair
Deer Creek - East	4	25	3.64	Poor
Bark River	20	92	2.72	Fair
Duck Creek	11	60	3.50	Poor

TABLE 8. *Evaluation of water quality by biotic index values of the arthropod fauna in the Wood-Clark County transect, June 11, 1975.*

Stream	Number of Genera	Number of Individuals	Biotic Index	Water Quality
Little Hemlock Creek	9	58	2.21	Good
Yellow River	16	80	2.41	Good
Tributary Rocky Run Creek	3	27	1.93	Excellent
Rocky Run Creek	9	69	2.41	Good
E. F. Black River - Wood Co. South	14	64	2.16	Good
Hay Creek	8	32	3.22	Poor
E. F. Black River - Clark Co.	14	70	2.24	Good
E. F. Black River - Wood Co. North	11	69	2.36	Good
Cunningham Creek	8	107	1.94	Good
Tributary Cunningham Creek	6	44	2.32	Good
Rock Creek	13	89	2.13	Good
Black River	24	172	1.72	Excellent
Arnold Creek	14	87	1.39	Excellent

TABLE 9. *Evaluation of water quality by biotic index values of the arthropod fauna in the Barron-Polk County transect, June 24, 1975.*

Stream	Number of Genera	Number of Individuals	Biotic Index	Water Quality
Staples Creek	5	36	3.97	Very Poor
Rice Beds Creek	10	70	2.39	Good
Fox Creek	16	81	2.57	Fair
Parker Creek	4	4	2.75	Fair
Harder Creek	12	92	2.26	Good
Wolf Creek - South	9	58	2.41	Good
Wolf Creek - North	14	143	2.29	Good
Trade River #1	12	23	1.35	Excellent
Cowan Creek	11	65	0.38	Excellent
Trade River #2	11	68	2.84	Fair
Butternut Creek	15	101	2.56	Fair
Trade River #3	15	119	2.24	Good
McKenzie Creek	20	156	1.22	Excellent
Straight River	7	61	2.89	Fair

curved forceps, remove and preserve in 70% ethanol arthropods still clinging to the net and those in the pan until 100 have been obtained. Do not collect arthropods less than 3 mm long, except adult Elmidae, because they are difficult to sample and identify. If 100 arthropods cannot be found in 30 minutes, those collected within that time period would constitute a sample, but this situation is unlikely to occur except in extremely polluted streams. Return samples to the laboratory for sorting, identification, enumeration, and calculation of the biotic index from the formula:

$$B.I = \frac{\sum n_i a_i}{N}$$

where n_i is the number of each species, a_i is the value for that species (Append. I), and N is the total number of arthropods in the sample (usually 100). Evaluate water quality according to Table 6.

Water quality of streams can be most thoroughly evaluated by pooling samples from different seasons, but this is probably not necessary. The best time to sample streams in Wisconsin is in the spring after streams have returned to normal flow, and from mid-September through November. From December through March most streams are frozen, and in April water levels are often very high from snow melt. Summer is less desirable because of a reduced summer fauna, and in late July and August the flow in some streams may almost cease. Sampling streams during periods of high water should be avoided because many insects burrow deep into the substrate to escape the turbulent flow.

An effective way to randomly sample all streams in an area is to construct a grid on a map of the area and sample at access points on roads closest to where each stream crosses a grid line. Range and township lines form a convenient grid for sampling most areas. If every third township and range line were used to form an 18 mile by 18 mile grid over the state of Wisconsin, and all streams were sampled according to procedures outlined above, one person should be able to make collections, sort samples, and evaluate the water quality of all these streams in one year. Such a survey would indicate where pollution is a problem and would provide valuable baseline data upon which to judge future improvement or deterioration of water quality. It would be especially valuable for delineating areas of non-point source pollution.

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APPENDIX I.

Values assigned to species and genera for the purpose of calculating a Biotic Index.

PLECOPTERA

Capniidae: *Allocapnia nivicola* 0, *Paracapnia angulata* 0

Chloroperlidae: *Hastaperla brevis* 0

Leuctridae: *Leuctra ferruginea* 0, *L. tenella* 0, *L. tenuis* 0, *L. truncata* 0

Nemouridae: *Amphinemura delosa* 0, *A. linda* 0, *Nemoura trispinosa* 0, *Shipsa rotunda* 0

Perlidae: *Acroneuria abnormis* 0, *A. internata* 0, *A. lycorias* 0, *Paragnetina media* 1, *Perlesta placida* 2, *Phasganophora capitata* 0

Perlodidae: *Isogenoides frontalis* 0, *Isoperla bilineata* 0, *I. clio* 0, *I. cotta* 0, *I. dicala* 0, *I. frisoni* 0, *I. lata* 0, *I. richardsoni* 0, *I. signata* 0, *I. slossonae* 0, *I. transmarina* 0

Pteronarcidae: *Pteronarcys* spp. 1

Taeniopterygidae: *Oemopteryx glacialis* 0, *Strophopteryx fasciata* 1, *Taeniopteryx* spp. 1

EPHEMEROPTERA

Baetidae: *Baetis brunneicolor* 3, *B. frondalis* 2, *B. intercalaris* 3, *B. levitans* 3, *B. macdunnoughi* 2, *B. phoebus* 3, *B. propinouus* 3, *B. pygmaeus* 3, *B. spinosus* 3, *B. vagans* 2, *B.* sp. A 2, *Callibaetis* spp. 3, Centroptilum spp. 1, *Cloeon alamance* 1, *Cloeon* spp. 2, *Heterocloeon curiosum* 1, *Pseudocloeon carolina* 2, *P. cingulatum* 2, *P. dubium* 2, *P. myrsum* 2, *P. parvulum* 2, *P. punctiventris* 2

Baetiscidae: *Baetisca bajkovi* 2, *B. obsea* 2, *B.* sp. A 2

Caenidae: *Brachycercus* spp. 2, *Caenis* spp. 4

Ephemerellidae: *Ephemerella attenuata* 0, *E. aurivillii* 0, *E. bicolor* 0, *E. deficiens* 0, *E. dorothea* 0, *E. excrucians* 0, *E. funeralis* 0, *E. invaria* 0, *E. lita* 1, *E. needhami* 1, *E. simplex* 1, *E. sordida* 0, *E. subvaria* 0, *E. temporalis* 4, *E.* sp. A 0, *E.* sp. B 0

Ephemeridae: *Ephemera simulans* 1, *Hexagenia limbata* 2

Heptageniidae: *Epeorus vitrea* 0, *Heptagenia diabasia* 3, *H. flavescens* 2, *H. hebe* 0, *H. lucidipennis* 1, *H. pulla* 0, *Rhithrogena impersonata* 0, *R. pellucida* 0, *Stenacron interpunctatum* 3, *Stenonema bipunctatum* 1, *S. exiguum* 3, *S. fuscum* 1, *S. integrum* 1, *S. medipunctatum* 2, *S. pulchellum* 1, *S. rubrum* 0, *S. terminatum* 2, *S. tripunctatum* 1

Leptophlebiidae: *Leptophlebia* spp. 3, *Paraleptophlebia* spp. 1

Polymitarcidae: *Ephoron leukon* 1

Potamanthidae: *Potamanthus myops* 2, *P. rufous* 2, *P. verticus* 2

Siphlonuridae: *Isonychia* spp. 2, *Siphlonurus* spp. 2

Tricorythidae: *Tricorythodes* spp. 2

ODONATA

Aeshnidae: *Aeshna* spp. 2, *Basiaeschna janata* 0, *Boyeria vinosa* 0

Calopterygidae: *Calopteryx* spp. 1, *Hetaerina americana* 1

Coenagrionidae: *Amphiagrion hastatum* 3, *Argia moesta* 2, *Chromagrion conditum* 3, *Enallagma* spp. 4, *Ischnura verticalis* 4

Corduligastridae: *Cordulegaster maculatum* 0

Corduliidae: *Tetragoneuria* spp. 2

Gomphidae: *Gomphurus* spp. 1, *Gomphus* spp. 1, *Hagenius brevistylus* 1, *Hylogomphus brevis* 0, *Ophiogomphus* spp. 0

Lestidae: *Lestes* spp. 3

Macromiidae: *Macromia* spp. 1

TRICHOPTERA

Brachycentridae: *Brachycentrus americanus* 0, *B. numerosus* 1, *B. occidentalis* 1, *Micrasema rusticum* 0, *M.* sp. A 0, *M.* sp. B 0, *M.* sp. C 0

Glossosomatidae: *Glossosoma* spp. 1, *Protoptila* spp. 0

Goeridae: *Goera stylata* 0

Helicopsychidae: *Helicopsyche borealis* 1

Hydropsychidae: *Cheumatopsyche* spp. 4, *Diplectrona modesta* 0, *Hydropsyche* spp. 3, *Macronema zebrata* 1, *Parapsyche apicalis* 0, *Potamyia flava* 1

Hydroptilidae: *Hydroptila* spp. 3, *Leucotrichia* spp. 3, *Neotrichia* spp. 3, *Ochrotrichia* spp. 3

Lepidostomatidae: *Lepidostoma* spp. 2

Leptoceridae: *Ceraclea* spp. 2, *Mystacides sepulchralis* 2, *Nectopsyche candida* 2, *N. diarina* 2, *N. pavida* 2, *N.* sp. A 2, *Oecetis avara* 2, *O.* sp. C 2, *Triaenodes* sp. B 1

Limnephilidae: *Hesperophylax designatus* 1, *Hydatophylax argus* 1, *Limnephilus* spp. 1, *Neophylax* spp. 1, *Onocosmoecus quadrinotatus* 0, *Platycentropus* spp. 2, *Pycnopsyche* spp. 1

Molannidae: *Molanna tryphena* 1

Philopotamidae: *Chimarra atterima* 0, *C. ferria* 0, *C. obscura* 2, *C. socia* 0, *Dolophilodes distinctus* 0, *Wormaldia moestus* 0

Phryganeidae: *Oligostomis ocelligera* 0, *Phryganea* spp. 2, *Ptilostomis* spp. 2

Polycentropodidae: *Neureclipsis* spp. 4, *Nyctiophylax* sp. A 0, *Phylocentropus placidus* 0, *Polycentropus cinereus* 0, *P. flavus* 0, *P. interruptus* 1, *P. remotus* 0

Psychomyiidae: *Psychomyia flavida* 2

Rhyacophilidae: *Rhyacophila fuscula* 0, *R. ignota* 0, *R. vibox* 0

Sericostomatidae: *Agarodes distinctum* 0

MEGALOPTERA

Corydalidae: *Chauliodes rasticornis* 2, *Corydalis cornutus* 2, *Nigronia serricornis* 1

Sialidae: *Sialis* spp. 2

LEPIDOPTERA

Pyralidae: *Neocatalysta* spp. 1, *Nymphula* spp. 1

COLEOPTERA

Dryopidae: *Helichus striatus* 1

Dytiscidae: *Agabus* larvae 1

Elmidae: *Ancyronyx variegata* 1, *Dubiraphia bivittata* 1, *D. minima* 3, *D. quadrinotata* 3, *D. vittata* 3, *Dubiraphia larvae* 3, *Macronychus glabratus* 1, *Microcylloepus pusillus* 1, *Optioservus fastiditus* 2, *O. trivittatus* 0, *Optioservus larvae* 2, *Stenelmis bicarinata* 2, *S. crenata* 3, *S. musgravei* 2, *S. sandersoni* 2, *S. vittipennis* 2, *Stenelmis larvae* 3

Gyrinidae: *Dineutus larvae* 1, *Gyrinus larvae* 2

Psephenidae: *Ectopria nervosa* 2, *Psephenus herricki* 2

DIPTERA

Blepharoceridae: *Blepharocera* spp. 0

Ceratopogonidae: *Atrichopogon* spp. 1, *Bezzia* spp. 3, *Palpomyia* spp. 3

Chironomidae: *Brillia* spp. 3, *Cardiocladius spp. 4, Chironomus* spp. 5, *Coelotanypus* spp. 2, *Conchapelopia* spp. 4, *Cricotopus* spp. 4, *Cryptochironomus* spp. 3, *Demicryptochironomus* spp. 3, *Diamesa* spp. 2, *Dicrotendipes* spp. 3, *Einfeldia* spp. 5, *Endochironomus* spp. 2, *Eukiefferiella* spp. 2, *Glyptotendipes* spp. 5, *Micropsectra* spp. 0, *Microtendipes* spp. 2, *Orthocladius* spp. 4, *Parachironomus* spp. 2, *Parametriocnemus* spp. 1, *Phaenopsectra* spp. 1, *Plecopteracoluthus* spp. 1, *Polypedilum* spp. 3, *Psectrocladius* spp. 2, *Psectrotanypus* spp. 2, *Rhecricotopus* spp. 1, *Rheotanytarsus* spp. 0, *Stictochironomus* spp. 0, *Sympotthastia* spp. 0, *Tanytarsus* spp. 0, *Zavrelimyia* spp. 4

Dixidae: *Dixella* spp. 2

Empididae: 4

Ephydridae: 4

Muscidae: *Limnophora* spp. 0

Rhagionidae: *Atherix variegata* 2

Simuliidae: *Eusimulium aurium* 1, *E. croxtoni* 0, *E. latipes* 0, *Prosimulium magnum* 0, *P. mixtum* 0, *Simulium corbis* 0, *S. jenningsi* 1, *S. tuberosum* 2, S. venustum 3, *S. vittatum* 4

Tabanidae: *Chrysops* spp. 2, *Tabanus* spp. 2,

Tipulidae: *Antocha* spp. 2, *Dicranota* spp. 0, *Hexatoma* spp. 3, *Limonia* spp. 0, *Pseudolimnophila* spp. 0, *Tipula* spp. 2

AMPHIPODA

Gammaridae: *Crangonyx gracilus* 4, *Gammarus pseudolimneus* 2

Talitridae: *Hyallela azteca* 4

ISOPODA

Asellidae: *Asellus intermedius* 5

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