ENVIRONMENTAL AUDITING

Assessing Biotic Integrity of Streams: Effects of Scale in Measuring the Influence of Land Use/Cover and Habitat Structure on Fish and Macroinvertebrates

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ABSTRACT / Fish and macroinvertebrate assemblage composition, instream habitat features and surrounding land use were assessed in an agriculturally developed watershed to relate overall biotic condition to patterns of land use and channel structure. Six 100-m reaches were sampled on each of three first-order warm-water tributaries of the River Raisin in southeastern Michigan. Comparisons among sites and tributaries showed considerable variability in fish assemblages measured with the index of biotic integrity, macroinvertebrate assemblages characterized with several diversity indexes, and both quantitative and qualitative measurements of instream habitat structure. Land use immediate to the tributaries predicted biotic condition better than regional land use, but was less important than local habitat variables in explaining the variability observed in fish and macroinvertebrate assemblages. Fish and macroinvertebrates appeared to respond differently to landscape configuration and habitat variables as well. Fish showed a stronger relationship to flow variability and immediate land use, while macroinvertebrates correlated most strongly with dominant substrate. Although significant, the relationships between instream habitat variables and immediate land use explained only a modest amount of the variability observed. A prior study of this watershed ascribed greater predictive power to land use. In comparison to our study design, this study covered a larger area, providing greater contrast among subcatchments. Differences in outcomes suggests that the scale of investigation influences the strength of predictive variables. Thus, we concluded that the importance of local habitat conditions is best revealed by comparisons at the within-subcatchment scale.

Physical habitat is a primary factor influencing the structure and composition of stream faunal communities (Gorman and Karr 1978, Schlosser 1982, 1987, Frissell and others 1986, Angermeier 1987, Cummins 1988, Osborne and Wiley 1992, Richards and others 1993, Richards and Host 1994, Poff and Allan 1995). Recent work in ecology (Wiens 1989) and stream ecology (Taylor and others 1993) has raised the question of the effect of scale in habitat investigations. The paradigm that has emerged holds that environmental variability affecting stream organisms occurs at multiple spatial and temporal scales (Frissell and others 1986, Addicott and others 1987, Downes and others 1993, Taylor and others 1993, Townsend and Hildrew 1994).

Explicit recognition of scale is also a central concern of studies relating landscape structure to stream ecosystem processes (Hunsaker and Levine 1995). According to the model of natural river systems by Frissell and others (1986), stream systems are spatially nested hierarchies of segments, reaches, pool/riffle units, and microhabitats. The larger scale features constrain the development of smaller units, and the resulting physical patterns, across both spatial and temporal scales, strongly influence the biology of the stream (Frissell and others 1986, Hawkins and others 1993, Rosgen 1994). However, this model, which links the physical structure of streams and their surrounding landscape to the distribution of stream organisms, remains a largely untested hypothesis (Schlosser 1991).

The study described here explores the hierarchical model of stream ecosystems by relating stream biotic integrity to patterns of land use and instream habitat in three agriculturally impacted streams. We sampled fish and macroinvertebrates and measured local habitat structure and adjacent land use following several studies that have sought to relate patterns of stream community composition to specific instream habitat and landscape features (Schlosser 1982, 1985, 1987, Steedman 1988, Wiley and others 1990, Osborne and Wiley 1992, Richards and others 1993, Malmqvist and Mäki 1994,

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Richards and Host 1994, Roth and others 1996). We focused on agricultural impacts because agriculture has caused extensive landscape changes (Allan 1995). Diffuse nutrient and sediment pollution from agriculture has been identified as the leading cause of water quality degradation in the United States (Osborne and Kovacic 1993).

Biotic indexes were used to measure biotic integrity for this study. Declines in stream ecosystems despite improvements in water quality associated with implementation of the Clean Water Act (33 U.S.C. 1251–1387) have spurred development of biological measures of water quality based on attributes of fish and macroinvertebrate communities (Karr and others 1986, Plafkin and others 1989, Quinn and Hickey 1990, Osborne and others 1991, Meador and others 1993, Rosenberg and Resh 1993, Kerans and Karr 1994). The faunal composition of streams is thought to reflect ambient conditions and integrate the influences of water quality and habitat degradation (Meador and others 1993).

Fish and macroinvertebrate indexes measure faunal diversity, functional diversity, and pollution tolerance and are used to rate sites against reference conditions for same sized streams within an ecoregion. However, these two taxonomic groups offer different advantages as water quality indicators. The multimetric index of biotic integrity (IBI) comprises fish species richness, dominance, abundance, trophic structure, tolerance to degraded conditions, and individual health (Karr and others 1986). Two principal advantages of using the IBI are its widespread use and the availability of reference data on fish. Macroinvertebrates are ubiquitous to streams and often exhibit greater taxonomic and trophic variety than fish. Plafkin and others (1989) suggest that macroinvertebrates are more indicative of local habitat conditions while fish reflect conditions over broader spatial areas because of their relative mobility and longevity.

Little agreement exists, however, about how to describe macroinvertebrate assemblages for biological assessment (Rosenberg and Resh 1993). An index comparable to the IBI has been developed for macroinvertebrates (Kerans and Karr 1994), but this index, the benthic index of biotic integrity (B-IBI), has yet to be applied widely. Other indexes that have been used widely include the invertebrate community index (ICI) (Ohio EPA 1988), which is similar to the B-IBI, and the biotic index (BI), which is based on pollution tolerances (Hilsenhoff 1987).

Our first objective was to test the hypothesis that differences in land use among catchments account for differences in biotic integrity among the streams of those catchments. Our second objective was to analyze the effect of local habitat variation on the relationship between land use and stream biota. The third objective was to compare the information generated by fish and macroinvertebrate assemblage measures, both in terms of their ability to distinguish or rank site quality and to determine if fish and macroinvertebrates responded similarly to instream habitat and landscape features.

Methods

Study Reach

The study area lies within the River Raisin watershed, a 2776 km^2 drainage basin located in southeastern Michigan, USA. Surficial geology is primarily fine-textured end moraine with coarse-grained end moraine and outwash deposits interspersed in the upper basin and fine-textured glacial lake deposits in the lower basin. Our study sites were within morainal regions (Figure 1), which minimized the differences in geology among sites, although some local variation was evident. Land use within the River Raisin watershed is predominantly agriculture, but varies among subcatchments. We selected three tributaries, Iron, Evans, and Hazen creeks, which represent subcatchments with differing amounts of agriculture. Six sites on each tributary were systematically spaced to cover the entire length of the tributary in a balanced one-way analysis of variance (ANOVA) design.

Based on visual inspection and our subjective impressions, we expected Iron Creek to contain more forested land and to have sites of higher quality than either Evans or Hazen creeks. Evans Creek is known to have been channelized, and Hazen Creek appeared strongly impacted by surrounding agricultural activity. We used the land use/cover classification system developed by the Michigan Department of Natural Resources to describe the three subcatchments in terms of seven major land use/cover categories: urban/extractive/open miscellaneous, agricultural, rangeland, forested, water, wetland, and barren. The agriculture category includes cropland, orchards, confined feed operations, and permanent pasture. Rangeland is either herbaceous vegetation or shrubland. The catchment areas of Iron Creek, Evans Creek, and Hazen Creek are 51.7 km^2, 49.8 km^2, and 75.5 km^2, respectively. The predominant land use/cover types in all three catchments are agricultural land, forested land, and rangeland. Together these three categories account for 81% of Iron Creek's catchment, 88% of Evans Creek's, and 96% of Hazen Creek's. Urban land use is low throughout the basins of all three tributaries, accounting for only 1% of the Hazen Creek drainage area, 7% of Iron Creek's, and 9% of Evans Creek's.

Fish

Following Karr and others (1986) and Roth and others (1996), fish populations were sampled at each site over a 100-m reach using an ABP-3 backpack-mounted electroshocking unit. Each 100-m pass was fished with an average of 225 V, 10% duty cycle and 60 pulses per second, by the same operator. Blocking seines were placed at the ends of the reach, and also at the 50-m point whenever high densities of fish were anticipated. Fishing effort varied per site depending on the complexity of the habitat present and the density of the fish, but was standardized as much as was practical. Fish were identified and counted on site, and the approximate standard length was noted. Individual fish that could not be identified on site were preserved in 10% formalin and subsequently identified in the laboratory.

Macroinvertebrates

Macroinvertebrates were sampled using a rapid bioassessment technique developed for streams of the Mid-Atlantic states (Mid-Atlantic Coastal Streams Workgroup 1993), many of which have low gradients, low velocities, and few riffle habitats, characteristics shared by Raisin tributaries. Approximately 300 organisms were collected from each site using a 425-μm mesh D-frame net, sampling microhabitats over the same reach sampled for fish and surveyed for instream habitat. Microhabitat types included snags and submerged woody debris, submerged and overhanging macrophytes, banks, riffles, and depositional areas, and they were sampled in approximately ten locations per reach in proportion to their occurrence. All macroinvertebrates were preserved in the field in 70% ethyl alcohol. Macroinvertebrates were identified to the lowest feasible taxonomic level in the laboratory. Heptageniid mayflies and hydropsychid caddisflies were identified to species, the majority of the remaining taxa to genus, and some dipterans, including Chironomidae, to family.

Instream Habitat

Instream habitat was assessed using measures adapted from the US Geological Survey (USGS) protocol for the National Water-Quality Assessment Program (Meador and others 1993), and the habitat quality evaluation index (HI), a qualitative method used by the Michigan Department of Natural Resources (MDNR 1989).

Quantitative measurements of channel features were made along six transects at 30-m intervals, covering a 150-m reach. Geomorphic feature (pools, riffles, and runs) and stream type were characterized once per reach. At each transect measures of channel structure included bank angle, bank width and height, channel width, and dominant substrate. Assessment of bank stability was based on the amount of vegetative and rock coverage. Riparian vegetation was characterized with an estimate of canopy cover and width of the riparian corridor. Flow velocity and depth were measured at three points across the channel for each transect. To characterize bankfull flow events, an index of flow stability was computed as the ratio between depth under low flow conditions and the estimated bankfull depth. The ratio of the estimated bankfull width to bankfull depth also was calculated as a measure of the channel's cross sectional shape (Gordon and others 1992, Rosgen 1994).

The HI provides a qualitative assessment of habitat condition at each site. This MDNR protocol assigns numbers corresponding to poor, fair, good, and excellent to nine metrics representing substrate and instream cover, channel morphology, and riparian and bank structure.

Land Use and Land Cover Assessment

We used the Michigan Resource Information System (MIRIS) to assess land use / cover at three landscape scales—for the entire subcatchment upstream of a given site, for a 250-m buffer area (extending 125 m on each side of the stream), and for a 100-m buffer area (extending 50 m on each side of the stream) (Figure 2). This land use / cover database was developed from aerial photographs for the years 1979–1985 and allows classification of areas at a resolution of approximately 1 ha (Fay 1995). Roth and others (1996) determined that 100-m resolution was reasonable for MIRIS. Our visual inspection of MIRIS land use maps superimposed on georeferenced, scanned Agricultural Stabilization and Conservation Service (USDA) aerial slides provided additional confirmation of MIRIS classification accuracy.

Our intent in assessing land use / cover was to see if the effects of local and subcatchment land use / cover could be distinguished. At the catchment scale, we were interested in the effect of the whole drainage area upstream of a site. Land use / cover at the subcatchment scale was calculated in two ways: for the entire drainage area upstream of a site (thus, the land area associated with a site increased as we proceeded downstream) and for the drainage area just up to the next site. These measurements were highly correlated: subsequent analyses used the entire drainage area percentages for each site. For the local and mesoscale measures, we wanted to isolate the effects of the more immediate landscape and configuration. Thus, the longitudinal extent of the

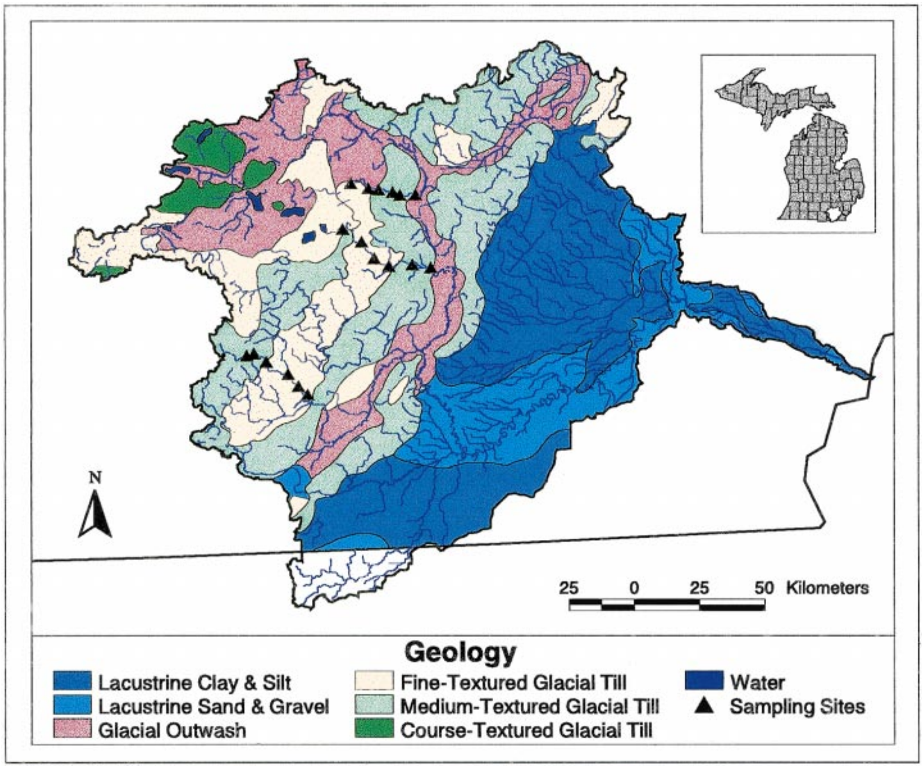

Figure 1. The quaternary geology of sampling locations on three tributaries of the River Raisin watershed (from north to south, the tributaries are Iron, Evans and Hazen creeks). The inset shows the location of the River Raisin watershed in Michigan.

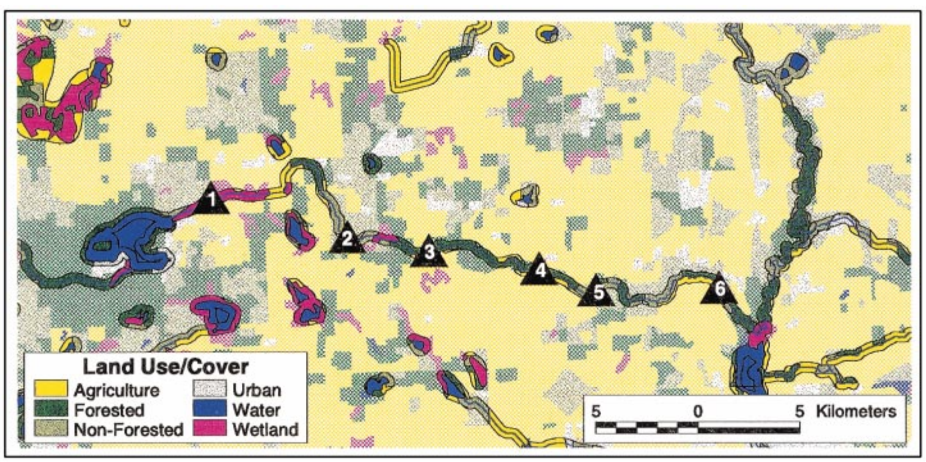

Figure 2. Land use/cover within the Iron Creek subcatchment, showing the outline of the 250-m buffer.

250-m and 100-m buffers was limited to the distance upstream to the next study site, approximately 1 km.

Subcatchment areas were digitized for each site from USGS 7.5′ quadrangle maps using the mapping program C-MAP (Enslin 1991). These drainage polygons were then merged with MIRIS land use/cover data in the GIS program ARC/INFO (Environmental Systems Research Institute 1994) to yield the land use/cover data for each catchment area. All land use/cover data were then converted to a percentage of the specific land area considered. ARC/INFO also allowed for the construction of buffer zones from which land use/cover data were extracted. The buffer data thus represent a subset of the catchment area information.

Sinuosity was calculated as the ratio of stream length to the linear distance between a study location and a point upstream, either the next site upstream, or a location approximately 2.5 km upstream in linear distance (Rosgen 1994). All distances were determined using C-MAP.

Measuring Biological Condition

Community composition data for fish were summarized using the IBI (Karr and others 1986), adjusted for

Table 1. Metrics and scores for the benthic index of biotic integrity (B-IBI) metrics[a]

Metric	Score 1	3	5
Total taxa (Intolerant snail and mussel richness)	14–22	23–30	30–39
Mayfly taxa	0–3	4–6	7–9
Caddisfly taxa	2–3	4–5	6–7
Dipteran taxa (Stonefly taxa)	3–4	5–6	7–8
(Relative abundance of *Corbicula* sp.)			
(Relative abundance of oligochaetes)			
Relative abundance of omnivores	0.70–0.81	0.57–0.69	0.43–0.56
Relative abundance of filterers	0.51–0.69	0.50–0.31	0.09–0.30
Relative abundance of grazers	0.06–0.19	0.20–0.32	>0.33
Relative abundance of predators	0.24–0.33	0.14–0.23	0.02–0.13
Dominance (Total abundance)	0.68–0.88	0.47–0.67	0.24–46

[a]Modified from Kerans and Karr (1994). The description of each metric appears in the left column and the scoring criteria to the right. Metrics omitted from this study are enclosed in parentheses.

the River Raisin following Roth and others (1996) and MDNR (1989). Multiple indexes were available to characterize macroinvertebrate communities. Resh and Jackson (1993) argue against relying on a single measure; thus, we used two multimetric indexes, the B-IBI and the ICI, four single-value metrics (taxa richness, taxa evenness, sensitive taxa richness, and number of mayfly taxa), and the BI, which is based on organic pollution tolerance. Calculation of the B-IBI, originally developed for streams in the Tennessee Valley drainage (Kerans and Karr 1994), required modification for use in southeastern Michigan to account for taxonomic differences. Four of the 13 metrics were eliminated and the scoring criteria were adjusted by trisecting the range of values obtained in this study and assigning scores of 1 (poor), 3, and 5 (best observed condition) (Table 1). The ICI uses a scoring system scaled to the drainage area for all of the metrics except percent mayfly composition and the percent tribe Tanytarsini midge composition, and assigns a score of 0 (worst condition), 2, 4, or 6 (best condition) (Ohio EPA 1988). Due to differences in taxonomic identification level, metrics for presence of tolerant species and for the presence of Tanytarsini midges were omitted. Taxa evenness was calculated using Shannon's index (Shannon 1948). The remaining single-value indexes are self-evident, except for sensitive taxa richness, which refers to the number of Ephemeroptera, Plecoptera, and Trichoptera (EPT) taxa.

Biotic index scores were determined according to the procedures described by Hilsenhoff (1987) and Lenat (1993). Tolerance values were assigned to the taxa based on the values given in Lenat (1993) or in Hilsenhoff (1987, 1988) where Lenat (1993) offered no value. The number of individuals in each taxonomic group was multiplied by its tolerance value, and a weighted average tolerance score for that sample was calculated (Hilsenhoff 1987). A higher score indicates a more degraded site in terms of organic pollution but is not necessarily an indicator of habitat degradation.

Results

Fish

Over 3000 fish were collected from the 18 sites. Six fish species accounted for 75% of the individuals captured, with the creek chub (*Semotilus atromaculatus*) being the most abundant and ubiquitous fish, found at 17 of 18 sites. Other commonly occurring species included blacknose dace (*Rhinichthys atratulus*), stoneroller (*Campostoma anomalum*), mottled sculpin (*Cottus bairdi*), white sucker (*Catostomus commersoni*), and johnny darter (*Etheostoma nigrum*). Rare species, those that comprised less than 1% of the total catch, accounted for 14 of the 28 species collected (see Lammert 1995 for a full list). Fish abundance varied considerably among sites, ranging from a low of six individuals to a high of 769, with a mean of 174 fish per site. The density of fish varied from 0.04 to 1.42 fish/m^2 (Figure 3A). Sites on Evans Creek generally had the lowest abundance, species richness, and densities. Although Iron Creek had lower average fish abundance than Hazen Creek, the two streams had similar average species richness.

IBI scores varied among sites from a high of 42 of a possible 50 points at two upper Iron Creek sites to a low of 14 at one mid-stream site on Evans Creek (overall mean $\pm$ SD; 31.7 $\pm$ 8.7; Figure 3B). Scores from Evans Creek sites were significantly lower than those of Iron and Hazen creeks ($F = 11.6$, $df = 2$, $P = 0.001$), but Iron and Hazen creeks were not significantly different (Tukey $P = 0.425$).

We were interested in which metrics contributed most significantly to the IBI scores. Using multiple linear regression, we determined that a two-metric model explained over 85% of the score value. Species richness was most highly correlated to the total IBI score ($R = 0.822$). Of the remaining metrics, only percent insectivorous fish and the number of intoler-

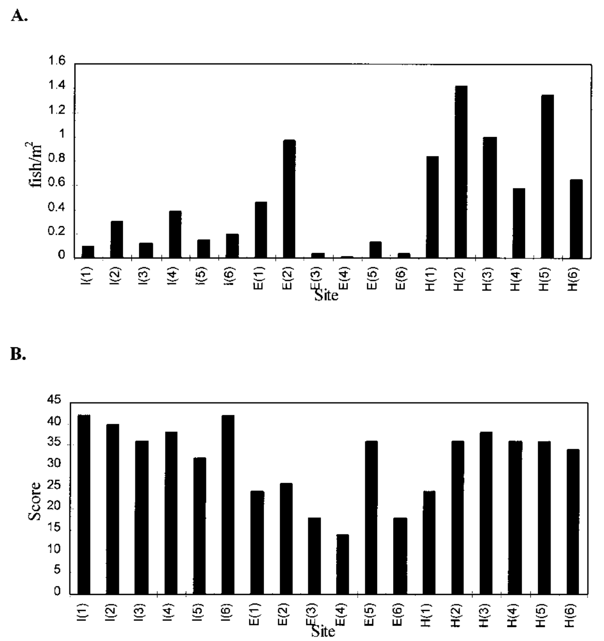

Figure 3. Fish collections from six sites on Iron (I), Evans (E) and Hazen (H) creeks in the River Raisin watershed. **(A)** Fish density, **(B)** IBI scores. The sites on each tributary are numbered from upstream to downstream.

ants (individuals of species considered intolerant to siltation and pollution) were significantly correlated to the residuals of the regression between species richness and total score. The addition of percent insectivorous fish to the regression relating IBI score to taxa richness increased the R^2 value from 0.68 to 0.85. However, the addition of number of intolerants as a third metric only increased the R^2 value slightly to 0.91. The number of intolerants was also moderately correlated to species richness ($r = 0.56$). Thus, the best model, with a transformation to correct for slight departures from the assumption of normality was (IBI score)$^2 = 11.58 + 1.61$ (species richness) $+ 19.19$ (percent insectivorous fish) ($R^2 = 0.85$, $P = 0.000$).

Macroinvertebrates

Almost 6000 macroinvertebrates in 84 distinct taxonomic groups were collected from the 18 sites. The 14 most common taxa comprised 73% of total individuals collected. Chironomidae, Calopterygidae (*Calopteryx* spp.), and Hydropsychidae (*Cheumatopsyche* spp.) were found at every site and, with Baetidae (*Baetis* spp.), were the four most abundant taxa (see Lammert 1995 for full list).

The single value measures of diversity—taxa richness, number of mayfly taxa, evenness, and EPT taxa—reflected the pattern of the fish samples, where Iron Creek sites again had the highest average values and Evans Creek the lowest. The three tributaries did not differ significantly in taxa richness ($F = 2.46$, $df = 2$, $P = 0.12$). However, the number of mayfly taxa was significantly different among the three tributaries ($F = 9.82$, $df = 2$, $P = 0.002$). The average number of mayfly taxa at the Evans and Hazen sites was significantly lower than at the Iron sites (Iron–Evans Tukey $P = 0.002$, Iron–Hazen Tukey $P = 0.02$), but the Hazen and Evans sites were not distinct (Tukey $P = 0.40$). However, the EPT taxa (in actuality Ephemeroptera and Trichoptera, since Plecoptera were found only at one site) only differed between sites on Iron and Evans creeks ($F = 7.22$, $df = 2$, $P = 0.006$, Iron–Evans Tukey $P = 0.005$). Shannon diversity (H′) was significantly different among the three tributaries ($F = 3.83$, $df = 2$, $P = 0.05$). Pairwise comparisons showed that the average of the Evans Creek sites was significantly lower than that of the Iron Creek sites (Tukey $P = 0.05$). However, due to the departure of this ANOVA model from assumptions of normality, these results should be interpreted with caution.

B-IBI scores ranged from 16 to 38 of a possible 40 points (mean ± SD: 25.9 ± 6.2). Again, sites at Evans scored significantly lower that the other two tributaries ($F = 5.58$, $df = 2$, $P = 0.02$). ICI scores had a broader range, from 6 to 34, of a possible 35 points (mean ± SD: 22.5 ± 7.6). However, the mean ICI scores of the three tributaries were not significantly different ($F = 3.12$, $df = 2$, $P = 0.07$). The coefficient of variation for the ICI scores was 34%, compared to 22% for the B-IBI. The two multimetric indexes were moderately correlated ($R^2 = 0.51$, $P = 0.001$).

We repeated the analysis used for fish integrity (IBI) to find out which metrics were driving B-IBI scores for these data. The best index to predict the total B-IBI scores included the number of mayfly taxa and the percentage of individuals in the grazer functional feeding group. Mayfly taxa had the highest correlation to total B-IBI score ($R^2 = 0.63$, $P = 0.000$). With the exception of percent grazers, the remaining metrics were not significantly correlated to the residuals of this regression and had little influence on the total B-IBI score. The addition of percent grazers increased the R^2 value to 0.73 ($P = 0.000$). Most of the remaining metrics were also moderately to highly correlated to the number of mayfly taxa and percent grazers. The best regression model was B-IBI score $= 15.00 + 1.47$ (number of mayfly taxa) $+ 18.62$ (% grazers).

Correlation and an analysis of regression residuals showed that the number of EPT taxa and percent caddisflies explained most of the variation in ICI scores. The EPT count by itself accounted for 84% of the variance, and the addition of percent caddisflies raised

Table 2. Pearson correlation matrix of macroinvertebrate assemblage measures[a]

	TAXA	TMAY	H′	EPT	B-IBI	ICI
TMAY	0.670					
H′	0.799	0.769				
EPT	0.767	0.919	0.781			
B-IBI	0.618	0.791	0.681	0.822		
ICI	0.782	0.828	0.820	0.914	0.713	
BI	−0.515	−0.648	−0.476	−0.712	−0.755	−0.669

[a]Measures include two multimetric indexes: the benthic index of biotic integrity (B-IBI) and the invertebrate community index (ICI); four single-value measures: number of taxa (TAXA), number of mayfly taxa (TMAY), Shannon diversity (H′), and number of Ephemeroptera, Plecoptera, and Trichoptera taxa (EPT); and the pollution tolerance biotic index (BI). All correlations are significant at $P \leq 0.05$.

the R^2 value to 0.92. EPT count and percent caddisflies were moderately correlated (Pearson $r = 0.54$). The next most correlated metric was taxa richness, but its addition to the model raised the R^2 only by 0.007. Taxa richness was also moderately correlated to the EPT count (Pearson $r = 0.77$). Thus, the best two-metric model was ICI score = 4.53 + 1.31 (EPT count) + 67.94 (% caddisflies) ($R^2 = 0.92$, $P = 0.000$).

Biotic Index scores ranged from 4.38 at a mid-stream site on Iron Creek to 6.28 at a mid-stream site on Evans Creek (mean ± SD: 5.7 ± 0.5). The coefficient of variation for all of the sites was 9%, the lowest for any of the indexes and diversity measures. The mean for the Iron Creek sites was 5.41, slightly lower than that found in the other two tributaries, but differences among the tributaries were not significant ($F = 1.76$, $df = 2$, $P = 0.252$). The BI had a high negative correlation to the EPT count and to the percent grazers (Pearson $r = -0.712$ and -0.743).

All macroinvertebrate diversity measures were significantly correlated to each other (Table 2). The highest correlations occurred between the ICI and EPT taxa, and EPT and number of mayfly taxa. The interdependence of these values indicates that the measures of macroinvertebrate biotic integrity calculated here depend primarily on taxa richness.

Instream Habitat Measures

Although all study sites except the three downstream sites on Hazen Creek are located on first-order reaches of the three tributaries, measures of channel morphology varied considerably among tributaries. Iron Creek had the widest channels, as well as lowest bank heights and bank angles. The flow stability index and ratio of bankfull width to bankfull depth reflect this morphology and suggest that Iron Creek is less flashy than the other two tributaries. Iron Creek also had the most stable bank vegetation and highest shading (Table 3), and the highest mean HI scores (mean ± SD: 70.33 ± 17.53) followed by Hazen (56.67 ± 19.19, then Evans 47.33 ± 19.95). However, mean HI scores were not significantly different among tributaries ($F = 2.24$, $df = 2$, $P = 0.140$). HI scores were most highly correlated to substrate size and embeddedness ($r = 0.944$ and 0.912). However, embeddedness explained little of the remaining variation in HI scores as it was highly correlated to substrate size ($r = 0.866$). Bank vegetation condition was the second most important factor for habitat condition, and with substrate explained 95% of the variation in HI scores. The best model for the HI was HI score = 14.71 + 3.38 (substrate) + 2.87 (bank vegetative stability) ($R^2 = 0.95$, $P = 0.000$).

Landscape Assessment

At each of the three scales of measurement, comparison of the tributaries revealed that Iron Creek has the least agricultural land and the greatest amount of forested land of the three subcatchments (Table 4). Hazen Creek has more agricultural land than Evans, except within the 100 m buffer, where values are similar. Within its entire catchment, Hazen Creek has the lowest percentage of forest cover of the three tributaries. However, within the 250-m and 100-m buffers, Hazen Creek has more forested land than Evans Creek, and almost as much as Iron Creek (Table 4).

Land use / cover percentages within each scale of measurement—across the entire catchment, and within the 250-m and the 100-m buffers—were highly correlated. For agriculture, forest, wetland, and urban land use / cover, all Pearson correlations ranged from 0.53 to 0.99. These correlations were expected given that each land use / cover displaces another. However, land use patterns within each scale category were not consistently correlated. Urban land use differed little whether measured within a buffer or across the entire subcatchment, showing that urban land use was consistent in its distribution relative to the three streams. In contrast, forest cover percentages differed greatly by scale of measurement (Pearson r values ranged from -0.29 to 0.33), showing that forest cover is not evenly distributed throughout the three subcatchments.

Relationships between instream and landscape variables. A number of instream physical variables were significantly correlated to land use / cover categories at each scale of measurement (Table 5). At the catchment scale, five habitat variables—bank angle coefficient of variation, bank width, bank width standard deviation, channel width, and the riparian index—were correlated to the extent of forest and agriculture. At the 250-m buffer scale, correlations among habitat features and land use were more variable. Canopy openness and discharge

Table 3. Comparisons of channel characteristics among the three tributaries

	Iron			Evans			Hazen		
Variable	Mean	SD	CV[a]	Mean	SD	CV	Mean	SD	CV
Channel width (m)	4.30	0.93	0.22	3.70	1.60	0.43	3.90	1.90	0.49
Bank angle (degrees)	34.00	8.50	0.25	41.00	15.00	0.37	39.00	5.70	0.14
Bank height (m)	0.60	0.13	0.24	1.60	1.60	1.10	0.90	0.41	0.44
Flow stability index	0.40	0.08	0.31	0.16	0.08	0.45	0.25	0.16	0.03
Bank stability index	2.10	0.66	0.31	1.70	0.77	0.45	1.60	0.42	0.27
Shading index	3.20	1.10	0.36	1.60	0.70	0.43	1.90	0.97	0.50
Habitat index	70.30	17.50	0.25	47.30	20.00	0.42	56.70	19.20	0.34

[a]Coefficient of variation.

Table 4. Extent of agricultural and forested land expressed as a proportion of total area in three tributary subcatchments at each of three scales of measurement

	Catchment			250-m buffer			100-m buffer		
Tributary	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
Agriculture									
Iron	0.42	0.38	0.49	0.28	0.12	0.50	0.09	0.07	0.33
Evans	0.53	0.21	0.69	0.44	0.08	0.61	0.30	0.00	0.50
Hazen	0.66	0.53	0.73	0.52	0.42	0.67	0.29	0.12	0.39
Forest									
Iron	0.23	0.21	0.24	0.37	0.22	0.68	0.69	0.30	0.99
Evans	0.19	0.14	0.29	0.28	0.08	0.36	0.40	0.11	0.56
Hazen	0.17	0.14	0.23	0.35	0.21	0.49	0.54	0.35	0.64

Table 5. Pearson correlations coefficients relating land use/cover at 100-m, 250-m, and catchment scales to instream habitat variables[a]

	Scale					
	Agriculture			Forest		
Variables	100 m	250 m	Catchment	100 m	250 m	Catchment
Bank angle CV[b]			−0.468		0.629	0.481
Bank width	0.620	0.605	0.594		−0.527	−0.539
Bank width CV	−0.473					
Bank width SD	0.643	0.669	0.658		−0.541	−0.0602
Bank stability	−0.529	−0.547				
Canopy	−0.497				0.648	
Channel width			0.507			−0.622
Discharge	−0.500				0.486	
Pools	0.487	0.540		0.689		
Riparian width			−0.522			0.529
Runs				−0.692		
Velocity	−0.672	−0.614			0.480	

[a]Only significant ($P \leq 0.05$) correlations are shown.

[b]Coefficient of variation.

were significantly correlated to forest cover, but not to agriculture, which was correlated to bank vegetative stability and percent of the channel occurring as pools. Finally, within the 100-m buffer, agriculture was correlated to many more measures of channel structure and flow than was forest cover. Bank width, bank stability, canopy cover, discharge, percent of the channel occurring as pools and velocity were significantly correlated to agriculture, while only percent pools and percent runs were related to forest cover within the 100-m buffer.

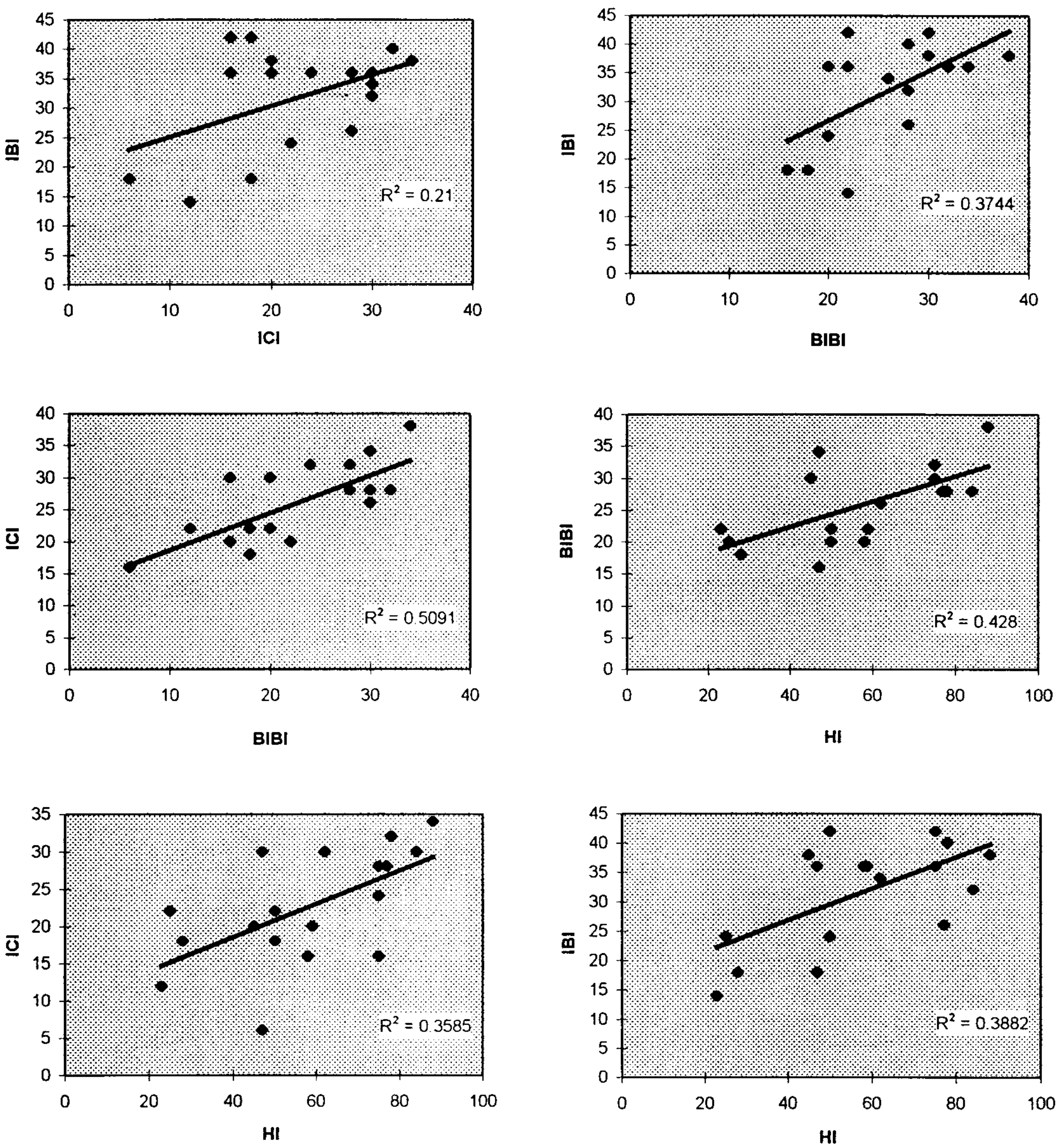

Figure 4. Comparisons among three biomonitoring indexes, the fish index of biotic integrity, (IBI), two macroinvertebrate indexes, the index of community integrity (ICI) and the benthic index of biotic integrity (B-IBI); and a habitat index (HI), using simple linear regression. Individual points are the six sampling sites located on three tributaries. All three biotic measures show a similar relationship to habitat quality. Higher correlations indicate that the metrics being compared provide similar rankings of sites. The strongest relationship is between the two macroinvertebrate indexes.

Relationships Between Biotic and Physical Variables

Relationships among major indexes. We compared macroinvertebrate and habitat measures to determine if they ranked sites similarly in terms of overall quality. Spearman's rank correlation allowed us to make pairwise comparisons of the B-IBI, ICI, BI, and HI at each site. This test showed that with one exception, ranking of the sites by IBI (fish) scores had the least concordance with rankings by other indexes. The IBI ranking of sites was most consistent with the B-IBI (Spearman's $r = 0.59$), but the remaining correlations were below $r = 0.50$ (Lammert 1995). This result lends credence to the view that indexes based on macroinvertebrates and fishes give somewhat different indications of stream condition.

Plots of index scores against each other provided a second means of comparing site rankings (Figure 4). The strongest linear correlation occurred between the two multimetric macroinvertebrate indexes, B-IBI and

ICI. The B-IBI was moderately correlated to IBI scores, but the apparent weak linear relationship between ICI scores and the IBI was not significant. However, when corrected for substrate size, the correlation between the IBI and B-IBI scores was strong ($R^2 = 0.52$). The biotic indexes all were significantly related to the HI. The B-IBI showed the strongest association with the HI, followed by the IBI and the ICI. Substrate size was a key metric for the HI, and as will be discussed below, substrate also showed a strong relationship to the B-IBI.

To determine whether site rankings varied with the index used, we compared index performance using multivariate analysis of variance (MANOVA). The IBI and B-IBI differentiated one stream from the other two when tested with univariate ANOVA. MANOVA allowed us to treat all of the indexes as dependent variables at the same time. When taken all together, the indexes showed that the difference between the tributaries was highly significant (Wilks' lambda $P = 0.013$; Hotelling-Lawley trace $P = 0.009$). This analysis confirmed our initial expectation that the three tributaries would show distinct levels of biologic integrity given land use and land cover.

Associations between biotic measures and instream habitat variables. Metrics describing the fish assemblages showed strong and significant associations with a number of channel characteristics (Table 6). IBI scores correlated most strongly with the coefficient of variation of channel width and flow stability. The number of fish taxa also had a positive association with channel width standard deviation. Fish abundance was strongly correlated to measures of channel morphology, including percent pool ($r = 0.50$) and percent run ($r = -0.65$).

Macroinvertebrate indexes were strongly correlated to dominate substrate size (Table 6). The strongest pairwise association between any of the biotic measures and instream habitat was between B-IBI and substrate size. Other relatively high correlations included the BI and substrate size, and the BI and average velocity.

Relationships between biotic measures and landscape variables. The clearest pattern of associations was between fish and macroinvertebrate integrity measures and between forest and agriculture, particularly at the 100-m buffer scale. Urban land, rangeland, wetlands, and surface waters showed no significant relationships (all r values were less than 0.45). With the exception of rangeland, these categories averaged less than 10% of the buffers and subcatchment, values too low perhaps to allow detection of significant relationships. Results will be reported in detail only for forest cover and agriculture.

Forested cover within the 100-m buffer was moderately correlated to most fish and macroinvertebrate measures. Agricultural land use within the 100-m buffer generally had significant negative correlations to all biotic measures. Simple linear regression analysis was used to explore further these trends in the relationship between biotic measures and extent of agricultural and forested lands at the three landscape scales—the subcatchment, 250-m buffer, and 100-m buffer. In all cases, the linear relationships were strongest at the 100-m buffer scale (Table 7). The percent forested area within the 100-m buffer was a better predictor of IBI scores, B-IBI scores, HI scores, and number of EPT taxa than was percent agricultural land. In contrast, ICI and BI scores had stronger relationships to percent agriculture in the 100-m buffer.

Multiple factor models for biotic integrity. Land use alone explained only 35% or less of the variation in biotic integrity scores (Table 7). We thus explored with multiple linear regression whether the addition of instream habitat variables would explain the remaining variation (Table 8). Analysis of both landscape and habitat variables showed that multiple factor models were better predictors of biotic integrity. However, the best models (in terms of parsimony and regression values) for fish and macroinvertebrates contained only local habitat variables. Flow stability and percent forested

Table 6. Instream habitat variables showing correlation to biotic measures where $r > 0.50$

Biotic measure	Instream variable	Pearson correlation
IBI	Channel width CV[a]	0.523
	Flow stability	0.551
Number of fish taxa	Channel width CV	0.698
Fish abundance	Percent pool	0.580
	Percent run	−0.651
B-IBI	Substrate size	0.803
	Mean velocity	−0.671
BI	Mean velocity	−0.622

[a]Coefficient of variation.

Table 7. Simple linear regression values for correlations (R^2) between biotic measures and land use/cover at each of three scales of measurement[a]

	Agriculture			Forest		
Index	100 m	250 m	Catchment	100 m	250 m	Catchment
IBI	**0.22**	0.09	0.00	**0.28**	**0.23**	0.01
B-IBI	**0.32**	0.07	0.01	**0.35**	0.18	0.00
ICI	**0.25**	0.02	0.04	0.13	0.17	0.03
HI	**0.22**	0.02	0.04	**0.29**	0.13	0.00
EPT	**0.31**	**0.31**	0.07	**0.28**	0.03	0.07
BI	**0.36**	0.11	0.09	**0.25**	**0.24**	0.03

[a]Values significant at $P \leq 0.05$ are shown in bold type.

Table 8. Multiple linear regression models to predict biotic measures[a]

Dependent variable	Independent variable	R^2	ΔR^2
IBI	1 Percent forested, 100 m buffer	0.276	
With land use included	2 Flow stability	0.435	+0.159
	3 Channel width SD	0.525	+0.090
Without land use included	1 Flow stability	0.303	
	2 Channel width SD	0.468	+0.165
B-IBI	1 Substrate size	0.645	
	2 Flow stability CV[b]	0.814	+0.169
	3 Percent forested, 100 m buffer	0.872	+0.058
	4 Percent run	0.889	+0.017
	5 Bank height	0.895	+0.007
ICI	1 Substrate size	0.297	
	2 Flow stability	0.423	+0.126
	3 Sinuosity	0.480	+0.057
BI	1 Substrate size	0.450	
	2 Velocity	0.777	+0.327
	3 Percent agriculture, 100 m buffer	0.795	+0.018

[a]The single best predictor variable is listed first as the independent variable. The next best predictors are evaluated for their reduction of residual variation (R^2 and ΔR^2).

[b]Coefficient of variation.

land in the 100-m buffer together explained 44% of the variation in IBI scores. A model with flow stability and the standard deviation of channel width proved a slightly better predictor of IBI scores ($R^2 = 0.47$), indicating that at-site channel features predicted the IBI at least as well as land use measures.

Substrate size explained the greatest proportion of the variance in all of the multimetric macroinvertebrate indexes. For the B-IBI, flow stability was a significant additional factor, increasing the variance explained by 20% ($R^2 = 0.814$). Flow stability was also the next strongest variable after substrate size in explaining variation in the ICI, increasing the R^2 by 10% ($R^2 = 0.42$). Finally, substrate size explained 45% of the variance in BI scores alone. The addition of average velocity to the model increased the variance explained by 32% ($R^2 = 0.78$).

Discussion

We demonstrated that habitat and immediate land use predicted biotic integrity in three warmwater streams in the midwestern United States. Flow stability for fish and dominant substrate for macroinvertebrates were good predictors of biotic condition. Our results also demonstrate that land use within 100-m of the stream was significantly related to biotic integrity. Catchment-wide (regional) land use showed no relationship. These results do not, however, provide a conclusive answer to the general question raised by landscape ecologists Hunsaker and Levine (1995) of whether local or catchment-wide factors have more of an impact on the biologic integrity of streams. Based on this and a previous study previous of the River Raisin watershed (Roth and others 1996), we recommend that this question not be cast as "either—or." The physical factors we measured have effects and are the result of processes that occur at multiple scales. We believe that our study demonstrates that the ability to tease apart sources of variation turns greatly on sampling design.

The finding that local land use and habitat were better predictors of biotic integrity than regional land use contrasts with findings of a previous study of the River Raisin watershed that found regional land a stronger predictor of fish assemblages than local habitat. Roth and others (1996) sampled broad areas of the Raisin River basin, and their data represent regional conditions for at least seven subcatchments. In the present design, sites were located close together and regional land use did not differ greatly within each subcatchment. Our 18 sites, in fact, represented only three regional conditions. As Poff and Allan (1995) note, the ability to detect a significant relationship depends on the range of conditions within the variable group of interest. Therefore, the scale of measurement in the present study was most appropriate for determining the effects of local variation, which was our intent.

Local habitat variables were shown to be superior to land use in predicting biotic integrity, particularly for macroinvertebrates. Predictive models between land use and biotic integrity were greatly enhanced by the addition of instream variables (see Table 8). Channel morphology and substrate conditions (for macroinvertebrate only) actually explained more of the variation in fish and macroinvertebrate biotic integrity than did land use/cover alone. In several cases, land use explained no additional variance beyond that explained by substrate and flow stability. These results suggest that riparian land use and instream habitat are not independent variables in this stream system, supporting the hypothesis of Frissell and others (1986) that the regional configuration of a watershed constrains the local structure. Richards and others (1996) found that local stream habitat could be traced to catchment-wide impacts as well as local land use and inherent characteristics of the land immediately adjacent to the stream. However, they also concluded that we are limited in our ability to measure these effects by data resolution and confounding factors such as geology and land use practices.

We expected the biological condition of the three tributaries to be more distinct based on differences in agricultural land use in each subcatchment, initial reconnaissance, and the conclusions of Roth and others (1996) regarding the importance of regional land use. However, along each stream, sampling sites varied greatly in channel structure and forest cover, and we thus found that variation in the faunal assemblages within tributaries was greater than that between tributaries. Multiple lines of evidence showed that sites on Iron Creek had the greatest ecological integrity. However, the average biotic and habitat condition of Iron Creek and Hazen Creek were not distinguishable, although Hazen Creek had the highest agricultural land use for all three scales considered. The two creeks were similar in forest cover within the 100-m buffer (Table 4), and channelization was evident in Evans Creek, demonstrating the importance of local habitat alteration. These findings recommend further investigation of the effect of the longitudinal forest configuration on streams.

Difference in habitat, possibly unrelated to land use, also might contribute to differences among tributaries in their biotic indexes (Schlosser 1987). Higher abundances of fish at the Hazen Creek sites than at the Iron Creek sites suggest that greater pool volume in Hazen Creek compensated for apparently superior habitat conditions in Iron Creek. While Iron Creek had greater flow stability and exhibited habitat features that suggested a less degraded stream (Table 2), Hazen Creek's channel was 22% pool, compared to only 9% of Iron's channel, which provided habitat to support greater concentrations of fish. Iron Creek did have higher IBI scores due to higher species diversity and more individuals of intolerant species, but high abundances of fish at Hazen Creek made their biotic condition hard to distinguish with the IBI (Figure 3).

The sensitivity, validity, and calibration of the biotic measures also influenced the ability to discern differences among sites and tributaries. Use of the IBI is widespread across the Midwest and has been employed in at least two studies of the River Raisin watershed (Fausch and others 1984; Roth and others 1996). This use assured availability of reference data at least at the ecoregional scale. No such reference data were available for the B-IBI. Thus, we used that index to rank sites relative to each to other. Further validation studies of the type described in Kerans and Karr (1994), and Karr and Chu (1997) will be required to establish the B-IBI as a means to detect water quality impacts in this region.

We chose to use a multiplicity of indexes to search for consistent relationships between biologic condition, land use, and instream habitat. High correlations between single-factor and multiple-factor indexes suggest that the indexes employed in this study were equally useful for determining the relative condition of study locations. For the B-IBI and ICI, taxonomic diversity metrics explained most of the variance in scores. This finding supports the view of Resh (1994) that simple indexes can be as effective as indexes that require additional analysis to assign taxa to functional groups.

IBI scores were more strongly correlated to land use / cover than were macroinvertebrate indexes, which were very strongly correlated to local habitat measures. These findings support the conclusion of Plafkin and others (1989) that macroinvertebrate assemblages are more indicative of local habitat conditions. While habitat heterogeneity creates refugia for fish and macroinvertebrates (Scarnecchia 1988, Sedell and others 1990), fish assemblages did not show as strong relationship to measures of habitat structure in comparison to macroinvertebrates. Substrate size, which is positively associated with substrate heterogeneity (Minshall 1984), was strongly correlated to most macroinvertebrate assemblage measures.

The nonconcordance of fish and macroinvertebrate biotic integrity may indicate that habitat measurements did not reflect variability in stream structure at a scale meaningful to fish. The mobility of fish and their possible linkage into larger metapopulations may reduce their sensitivity to the patchiness of stream habitat (Schlosser and Angermeier 1995). Angermeier (1987) found that regionally abundant fish species are less sensitive to habitat differences across space and time. The fish assemblages in this study were dominated by a few common taxa, which may explain why the fish did not have as well-defined associations with habitat as macroinvertebrates.

The successive studies of the River Raisin watershed (Roth and other 1996, Lammert 1995) offer significant insight into the effect of spatial scale on detecting land use influence on stream biotic integrity. The unexplained variation in biotic condition does, however, warrant further study. Richards and other (1996, 1997) and Wiley and others (1997) suggest that mapping sources of variation in aquatic assemblages requires a spatially hierarchical sampling design. Wiley and others (1997) also demonstrate the need to account for temporal variation. We recognize that nested designs replicated in space and time are ideal to develop the comprehensive picture of local and regional stream ecosystem mechanisms described by Wiley and others (1997), but we believe that our study shows that single event sampling is an effective and economic means to uncover patterns and hone hypotheses for future investigations.

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