Temporal Variation in Ohio River Macroinvertebrates: A Historical Comparison of Rock Basket Sampling (1965-1971 and 2002)

Matthew S. Wooten[a,b], Brent R. Johnson[c], and Erich B. Emery[a]

ABSTRACT

The United States Environmental Protection Agency (USEPA) used rock basket artificial substrates to sample benthic macroinvertebrates of the Ohio River from 1965 to1971. The objective of this study was to evaluate changes in the benthic assemblage and to assess health of the community since passage of the 1972 Clean Water Act (CWA). Rock baskets of the same configuration were placed in the vicinity of the historic locations and were allowed to colonize for the same six-week period in summer 2002. Macroinvertebrates collected from 2002 baskets were compared to those of historic samples by using non-metric multidimensional scaling, by employing proportional indices of community similarity, and by comparing commonly used macroinvertebrate metrics. Analyses were generally performed at the genus (insects) or order (non-insect) level to minimize taxonomic discrepancies between time periods. A total of 62 taxa groups was identified across all years, with midges and oligochaetes generally dominating. Only 10-16 taxa groups accounted for >93% of all individuals in each year, but taxa contributions varied greatly among years. The 2002 benthic assemblage was distinct from earlier years, with increases in total taxa richness and Ephemeroptera, Plecoptera, and Trichoptera taxa, and with a decline in the number of oligochaetes. Differences in 2002 data are partially attributed to a replacement of the amphipod *Crangonyx* sp. by *Gammarus* sp. and the invasion of the zebra mussel (*Dreissena polymorpha*) during the late 1980s. Our findings indicate the Ohio River benthic community today is markedly different from that of the 1960s and has shown general improvement since passage of the 1972 CWA.

INTRODUCTION

Over the past 200 years human activities have dramatically altered large rivers of the United States. Examples of impacts that have contributed to the widespread degradation of large river systems include channelization, impoundment, surface runoff, and point source discharges. As a result of these activities, large rivers are disproportionately degraded when compared with smaller wadeable systems (Karr et al. 1985). Dam construction alone has resulted in fewer than 43 free-flowing rivers >200 km in length in the United States (Benke 1990). Impoundment of large rivers interrupts the natural flow regime (Poff et al. 1997) and dramatically alters ecosystem integrity. Large river biota may be susceptible to cumulative effects of multiple stressors because of the wide variety of disturbance types that exist within their large drainage areas. Many rivers have also suffered a general homogenization of habitats and biological communities as a result of flow regulation and anthropogenic inputs (Thorp 1992, Ward and Stanford 1995).

Bioassessment of large river ecosystems is often difficult because of inappropriate reference conditions and the frequent absence of a well-defined disturbance gradient. Non-wadeable rivers also present researchers with several other challenges dealing with

[a] Ohio River Valley Water Sanitation Commission, Cincinnati, Ohio 45228 USA.

[b] Current address: Northern Kentucky Sanitation District No. 1, 1045 Eaton Drive, Fort Wright, Kentucky 41017 USA; E-mail: mwooten@sd1.org

[c] US E.P.A., Ecological Exposure Research Division, Cincinnati, Ohio 45268 USA.

sampling logistics, spatial scale issues, and lack of consensus on sampling methodologies. Large rivers are often overlooked by biomonitoring agencies as a result of these difficulties, and large river bioassessment in the United States currently lags far behind that of wadeable streams. Most large river monitoring that is conducted tends to rely on fish assemblages because fish generally become more abundant and diverse in the downstream direction (Emery et al. 2003). Monitoring agencies rarely conduct comprehensive assessments of large river macroinvertebrate communities due to the aforementioned challenges. However, the difficulty in assessing large rivers may require use of multiple indicator assemblages.

The Ohio River is exceptional for a large river in that some degree of biological monitoring has taken place on the river continuously since the 1950s. Lockchamber surveys of fish assemblages began in the 1950s, and electrofishing along the river began in the early 1990s. Benthic macroinvertebrate sampling began on the Ohio River in the early 1960s. From 1965 to1971, the United States Environmental Protection Agency (USEPA) sampled macroinvertebrate assemblages at various points along the length of the river using rock-filled baskets as artificial substrates (Mason et al. 1971). Since 1972, Hester-Dendy artificial substrates have been used by the USEPA and the Ohio River Valley Water Sanitation Commission (ORSANCO). The objective of this study was to document any chnges in the macroinvertebrate assemblage of the Ohio River since passage of the 1972 Clean Water Act (CWA). To do so comparatively, we used the same rock basket samplers in 2002 that had been employed in per-1972 surveys.

METHODS

The Ohio River drains 528,000 km^2 and begins at the confluence of the Allegheny River and Monongahela River at Pittsburgh, Pennsylvania. The river flows southwesterly for 1578 km before entering the Mississippi River at Cairo, Illinois and over its course, the river is regulated by 20 navigation dams. Along the length of the river, there are 194 municipal wastewater treatment facilities, 383 industrial discharges, 114 contaminated process discharges, and 49 power generating facilities. The U.S. Army Corps of Engineers also routinely dredges the channel for navigation.

Rock basket samplers used for macroinvertebrate collection in this study were similar to those used for historic sampling described by Mason et al. (1971). Wire baskets (17.8cm in dia. x 27.9cm length, 2.5cm mesh) were filled with ca. 25-30 stones (limestone aggregrate, 5-8cm). Rock baskets were deployed in Sept. 2002 in the same manner as historical samples and were retrieved following a six-week colonization period. Baskets were suspended from steel cables from the upstream side of lockchamber walls of the first 12 dams in the system, beginning near Pittsburgh, Pennsylvania with Emsworth Lock and Dam at river kilometer 9.9, and ending with Meldahl Lock and Dam at river kilometer 701.9, just upstream of Cincinnati, Ohio. At the end of the incubation period, an 18.9 liter bucket was carefully placed underneath each sampler to minimize loss of individuals as the baskets were retrieved. Baskets were opened in the bucket, and rocks were individually scrubbed to remove all organisms. Bucket contents were then sieved (600 μm mesh) and preserved with 10% formalin. All samples were identified to the lowest practical taxonomic level, usually species. However, in order to be conservative in our comparisons with historic data, we used the same level of taxonomic identification, typically genus level for insects and order or family level for non-insect taxa.

Between 1965 and 1971, the USEPA collected a total of 125 rock basket samples from the Ohio River. However, because these samples covered a wide temporal range, only those consistent with the 2002 sampling window were used for analysis. Only baskets that were historically deployed in late summer were considered, leaving 41 total baskets for the period 1965-1971. Historical sampling locations and number of baskets deployed varied among years. Therefore, in 2002, rock baskets were deployed at or near each location where historic samples were collected (n = 11 locations). Efforts were made to deploy baskets at the exact original locations, but some lock and dam sites have since been removed or relocated. In these cases, baskets were deployed on the lock and dam structure nearest the original location. Rock baskets from 2002 were paired with the nearest historical location for purposes of statistical comparison. The distances from the original locations were typically <8 river km, but ranged up to 42 river km. Baskets collected from such distances were still combined to represent one river location because Applegate (2002) found that river km generally has little effect on the Ohio River benthic community.

Macroinvertebrate assemblages collected in 2002 were compared to those from historic rock basket samples of USEPA (1965-1971) by three methods. Data were first ordinated using nonmetric multi-dimensional scaling (NMDS, Kruskal and Wish 1978) to visually evaluate differences among years based on separation or overlap in ordination space. Taxa abundance values were square-root transformed prior to analyses. Taxa that appeared in fewer than three rock baskets (approximately 4% of total used in analyses) were also excluded to reduce the effects of rare taxa. Ordinations were based on a Bray-Curtis dissimilarity matrix of square-root transformed abundance data. NMDS analyses were run using the medium setting on autopilot mode in PC-ORD (PC-ORD, v.4.25, MjM Software Design, Gleneded Beach, Oregon). Proportional indices of community similarity (Whittaker 1975) were also calculated for each year combination to provide simple quantification of any differences in benthic community composition.

ORSANCO has developed a preliminary macroinvertebrate index based on Hester-Dendy sampling that was designed to monitor point sources on the Ohio River (Applegate 2002). This preliminary index consists of 12 metrics that responded to low quality effluents in a pilot study (ORSANCO, unpublished data). The metrics are: 1) total number of taxa; 2) number of taxa within the orders Ephemeroptera, Plecoptera, and Trichoptera (EPT); 3) number of Diptera taxa; 4) number of individuals, 5) % Tanypodinae; 6) % Hydroptilidae; 7) number of Ephemeroptera and Trichoptera individuals; 8) number of chironomid individuals; 9) ratio of Ephemeroptera and Trichoptera to chironomid individuals, 10) % Amphipoda; 11) % EPT individuals; and 12) % Oligochaeta individuals. We applied these metrics to historic (1965-1971) and 2002 rock basket samples. Although this index was developed using data collected from a different type of artificial substrate sampler (Hester-Dendy multiplates [Hester and Dendy 1962]), the two methods provide results that compare favorably (Fullner 1971, ORSANCO unpublished data). Additionally, we calculated four other common metrics that have been used for large river benthic studies in the Midwest (Blocksom and Flotemersch 2005). These metrics were: 1) number of taxa within orders Ephemeroptera, Plecoptera, Odonata, and Trichoptera (EPOT); 2) % tolerant taxa; 3) % Chironomidae individuals, and 4) % dominant taxon. Metric values were compared among years with a one-way repeated measures analysis of variance (RM-ANOVA) followed by the Holm-Sidak test, with 2002 as the control year. Metric values were transformed prior to analysis to meet assumptions of normality and homogeneity of variance. If transformed data failed to meet those assumptions, we used a Friedman test, which is equivalent to a RM-ANOVA on ranks. Only two basket samples were collected in 1971, and these were excluded from metric comparisons.

RESULTS

A total of 62 taxa groups was collected for all years combined, though this number is conservative given the higher level of taxonomic identifications. The most taxa were collected in 2002 (n = 40), whereas the number of taxa collected each year from 1965 to1970 ranged from 16 to 28. After omission of rare taxa (<1% of individuals within a year), only 9-16 taxa groups accounted for >93% of all individuals in each year (Table 1). Dominance of taxa groups varied somewhat among years, but the years 1966-1971 were generally dominated by midges (Chironomidae) and oligochaetes. These two groups combined accounted for between 69% and 95% of all individuals collected during this time period, whereas in 2002 they only accounted for ca. 43% of individuals. Only five baskets were collected from 1965, and these were generally dominated by the net-spinning caddisfly *Cyrnellus* sp. (57.9% of individuals). The 2002 baskets were generally dominated by the invasive zebra mussel, *Dreissena polymorpha* (46.3% of total individuals), and by the chironomid *Dicroptendipes* sp. which averaged >25% of individuals per basket. Zebra mussels were absent from historic samples and their abundance in 2002 varied greatly among the 11 baskets (mean = 396.7 ± 353.2 SE). Other major taxa (>1% of individuals) found only in 2002 include *Hydra* sp., *Gammarus* sp., *Hydroptila* sp., and *Pseudochironomus* sp., whereas minor taxa (<1% of individuals) unique to 2002 included *Enallagma* sp., *Ceraclea* sp., and *Oecetis* sp.

Table 1. Major Ohio River macroinvertebrate taxa from rock baskets for the years 1965-1971 and 2002. Values are mean percentage of total individuals per basket; n = total number of individuals from all baskets. Only taxa that averaged > 1% of total within a year are included.

Taxon	1965 n=331	1966 n=1394	1967 n=15663	1968 n=6353	1969 n=2978	1970 n=5650	1971 n=73	2002 n=9416
Hydra sp.								1.7
Turbellaria	2.4	18.4	5.6		8.9	17.4		
Nemertea					5.1			
Nematoda		1.4						
Oligochaeta	12.5	29.8	32.3	16.8	26.2	17.0	5.4	2.8
Hirudinea	1.1	1.8						
Cladocera						1.1	2.6	15.4
Gammarus sp.								2.2
Orconectes sp.	6.5							
Stenacron sp.								7.2
Stenonema sp.					3.1	1.5		
Tricorythodes sp.			2.7					
Argia sp.	10.2		1.2	16.0	3.7	1.8		
Neurocordulia sp.						1.9		
Cyrnellus sp.	37.3	7.1	23.3	26.4	23.3	22.6	3.6	6.9
Polycentropus sp.			7.2					
Hydroptila sp.								1.0
Chironomidae				3.7		2.0		
Ablabesmyia sp.	8.4	4.7						4.1
Cricotopus sp.			4.5			2.1	26.7	1.1
Nanocladius sp.								2.4
Psectrocladius sp.	10.1	10.7	6.5	5.6	10.9	12.7	37.4	
Chironomus sp.			3.4					
Cryptochironomus sp.				2.2		1.2	3.7	
Dicrotendipes sp.	6.0	15.8	7.0	18.1	1.4	4.5	10.5	27.1
Glyptotendipes sp.		1.6			1.9	3.4	4.5	
Polypedilum sp.			1.9		1.3	1.3	5.6	
Pseudochironomus sp.								2.9
Pentaneura sp.				5.8	8.7	6.8		
Physa sp.						1.2		
Ferrissia sp.		3.8						
Corbicula sp.	3.7							
Dreissena polymorpha								18.7
Sphaeriidae				2.4				

NMDS ordination of 45 taxa indicated that three axes (stress = 15.44, n = 52 baskets) represented 79% of the total variation in the data set with axes 1 (26%) and 2 (33%) representing the most variability. Rock baskets collected in 2002 formed a distinct cluster at the negative end of axis 2 (Fig. 1) and were generally less variable than baskets collected in previous years. Axis 1 generally defined a gradient of taxa sensitivity and was positively correlated with oligochaetes (r = 0.65), *Cricotopus* sp. (r = 0.52), and nematodes (r = 0.31), while negatively correlated with *Cyrnellus* sp. (r = -0.48) *Stenonema* sp. (r = -0.30) and other insect taxa in the EPOT Orders. This Axis was also correlated with several metrics that again reflected the sensitivity gradient. Axis 1 was positively correlated with % oligochaete individuals (r = 0.72) and total number of individuals (r = 0.53) and was negatively correlated with EPT individuals (r = -0.57) and E and T / chironomid ratio (r = -0.43). Baskets samples collected in 2002 clustered at the negative end of Axis 2, which was negatively correlated with several taxa that were unique or more abundant that year, including *Gammarus* sp. (r = -0.63), *Stenacron* (r = -0.63), *Dicrotendipes* sp. (r = -0.72), *Hydroptila* sp. (r = -0.55), and *D. polymorpha* (r = -0.39). Axis 2 was negatively correlated with % tolerant individuals (r = -0.62) and with several richness metrics, including total number of taxa (r = -0.68), number of Diptera taxa (r = -0.66) and number of EPT taxa (r = -0.65).

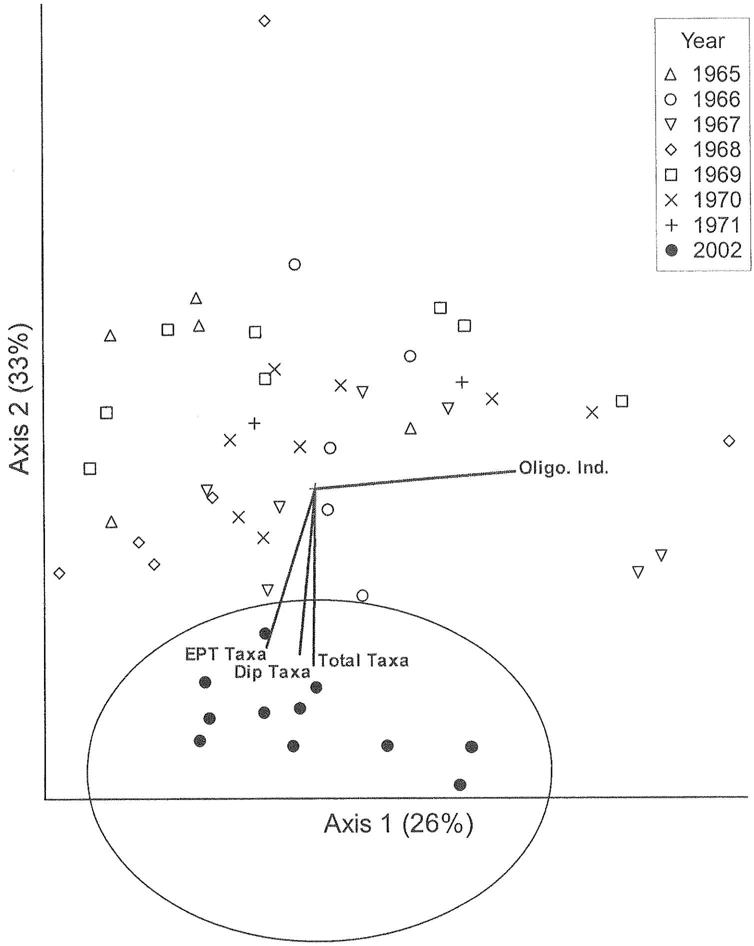

Figure 1. Non-metric multidimensional scaling ordination of historic and 2002 macroinvertebrate samples, including metric vectors ($r^2 \geq 0.4$).

The proportional index of community similarity also showed that 2002 benthic data were distinct from historic collections and supported NMDS results. Percent similarity values for 2002 with all past years ranged from14.9 to 32.8% with an average of 23.4% (Table 2). In contrast, all pairwise comparisons for the years 1965-1971 ranged from 25.7 to 76.7% with and average of 50.4%. Similarity among all historic years (1965-1971) was therefore more than double the similarity of 2002 with any other year, further indicating the differences that existed in the 2002 Ohio River benthic community.

Table 2. Proportional index of community similarity for Ohio River rock basket samples collected in 1965-1971 and 2002.

	1971	1970	1969	1968	1967	1966	1965
2002	21.7	20.2	14.9	31.1	20.0	32.8	22.3
1971		35.2	25.9	28.0	29.0	32.1	25.7
1970			76.7	62.2	61.7	61.4	55.2
1969				60.0	67.4	58.1	54.3
1968					55.5	48.1	62.2
1967						60.1	54.2
1966							46.2

All rock baskets collected within a year were grouped, and average metric values were compared across years. Eleven of the 16 benthic metrics tested indicated significant differences between 2002 and at least one of the previous years (Table 3). The number of taxa collected per basket for the period from 1965 to 1970 averaged 8.32, less than half the 2002 average of 17.2. Similarly, metrics for EPT and EPOT taxa richness, % Hydroptilidae, number of Diptera taxa, and % Amphipoda individuals were all significantly higher in 2002 than in all previous years. Metrics for EPT and EPOT taxa richness were significantly higher in 2002 because of generally higher proportions of the mayflies *Stenacron* sp. and *Caenis* sp. and the caddisfly *Cheumatopsyche* sp. Several members of these orders were also unique to 2002 samples, including the odonate *Enallagma* sp. and the caddisflies *Hydroptila* sp., *Ceraclea* sp., and *Oecetis* sp. Though the average % Hydroptilidae was low for 2002, hydroptilids had not been collected from rock baskets historically. *Cyrnellus* sp. dominated the historical EPT assemblage and comprised over half of all individuals from 1965 samples. Though still present in 2002, *Cyrnellus* sp. proportions have declined since the 1960s.

Chironomids represented the vast majority of dipteran taxa collected from rock baskets across all years. The average number of dipteran taxa collected per basket in 2002 was nearly double the average for 1965-1971. Many chironomid taxa were common to both 2002 and historic samples, but several midge taxa were also unique to 2002 samples, including *Nanocladius* sp., *Parakiefferiella* sp., *Thienemanniella* sp., *Cryptotendipes* sp., *Pseudochironomus* sp., and *Rheotanytarsus* sp. The metric % Tanypodinae was also higher in 2002 than in the years 1968-1971 when none were collected. The average percent amphipod metric was low in 2002 (mean = 2.63) yet was still significantly higher than in all previous years. All amphipods collected in 2002 samples were *Gammarus* sp., whereas all amphipods collected in earlier years were *Crangonyx* sp. The % oligochaete individuals metric was significantly lower in 2002 than in 1966, 1967, and 1969, while the metric for % tolerant taxa was significantly higher in 2002 than in the years 1965-1970.

In addition, 2002 metric values were plotted against values from all previous years by river mile location (Fig. 2). Graphical interpretation of metric values further demonstrates higher values in 2002 for total number of taxa, number of EPT taxa, number of Diptera taxa, % Hydroptilidae, number of chironomid individuals, % Amphipoda, number of EPOT taxa, and % tolerant taxa. Metric values clearly higher in historic rock basket samples include number of E and T individuals, E and T: chironomid ratio, and % Oligochaeta.

Table 3. Comparison of commonly used macroinvertebrate metrics between historic (1965-1971) and 2002 rock basket samples. Values are means; n = number of baskets per year. * indicates significant difference ($p<0.05$) from 2002 (one-way RM-ANOVA followed by Holm-Sidak test).

Metric	1965 n=5	1966 n=5	1967 n=7	1968 n=6	1969 n=8	1970 n=8	2002 n=11
Number of taxa	6.4*	9.6*	9.4*	6.3*	9.4*	10.1*	17.2
Number of EPT taxa	1.0*	2.0*	2.0*	1.2*	1.5*	1.25*	3.4
Number of Diptera taxa	2.8*	3.0*	3.6*	2.7*	3.9*	4.1*	6.6
Number of Individuals	62.2	278.8	2237.6	1058.8	372.3	706.3	856.0
% Tanypodinae	8.4	4.7	0.8	0*	0*	0*	4.1
% Hydroptilidae	0*	0*	0*	0*	0*	0*	1
Number of E and T individuals	36.8	17	135.3	110.2	32.1	92.5	39.6
Number of Chironomid individuals	11.4	132.4	85.1	115.4	34.5	75.6	132.8
E and T per Chironomid ratio	3.4	0.8	1.8	0.8	1.2	1.2	0.4
% Amphipoda individuals	0*	0*	0.1*	0.9*	0.2*	0*	2.6
% Oligochaeta individuals	12.5	29.8*	32.3*	16.8	26.2*	17.0	2.8
Number of EPOT taxa	1.6*	2.6*	2.4*	1.7*	2.3*	2.0*	4.4
% Chironomid individuals	25.4	33.3	25.0	36.0	27.1	34.1	40.3
% Tolerant taxa	6.3*	17.8*	16.8*	18.7*	5.6*	11.2*	37.5
% Dominant taxon	54.3	56.8	56.9	66.8	53.8	54.6	52.0

DISCUSSION

Our results indicate the Ohio River macroinvertebrate assemblage is markedly different and somewhat improved from historic conditions. Similarity analyses verified a temporal change in community composition driven by changes in taxa proportions and by several taxa unique to 2002 samples. The majority of calculated benthic metrics also indicated significant differences between 2002 and historic samples. Most apparent were the significant increases in total number of taxa, number of EPT and EPOT taxa, number of dipteran taxa, and % Amphipoda, along with a decline in % oligochaetes. These metric responses all indicate a general improvement in river condition. The average number of taxa per basket in 2002 was nearly double that of historic samples, indicating a more diverse and more evenly distributed benthic community. Taxa richness has often been included in multi-metric indices based on the principle that increasing diversity correlates with a healthier ecosystem capable of supporting survival and reproduction of many species (e.g., Resh et al. 1995, Barbour et al. 1996). Reductions in taxa richness have been observed in response to various types of disturbance, including acid mine drainage (e.g., Dills and Rogers 1974), organic enrichment (e.g., Hilsenhoff 1987, Ortiz et al. 2005), and insecticides (e.g., Wallace et al. 1996, Schulz and Liess 1999). In the Ohio River, which is subject to numerous point and non-point source pollutants, Beckett and Keyes (1983) demonstrated reductions in taxa richness in heavily polluted portions of the Ohio River. Reduced taxa richness has been frequently observed in benthic samples collected directly from the plume of a point source discharge (ORSANCO, unpublished data).

Members of the EPT insect orders are widely considered to be sensitive indicators and are often among the first groups to disappear in the presence of disturbance (e.g., Lenat 1988, Ohio EPA 1988). Though densities of taxa within these orders were typically low in 2002 basket samples and some taxa collected are considered the more tolerant members (e.g., *Cheumatopsyche* sp.), the increased richness within these orders

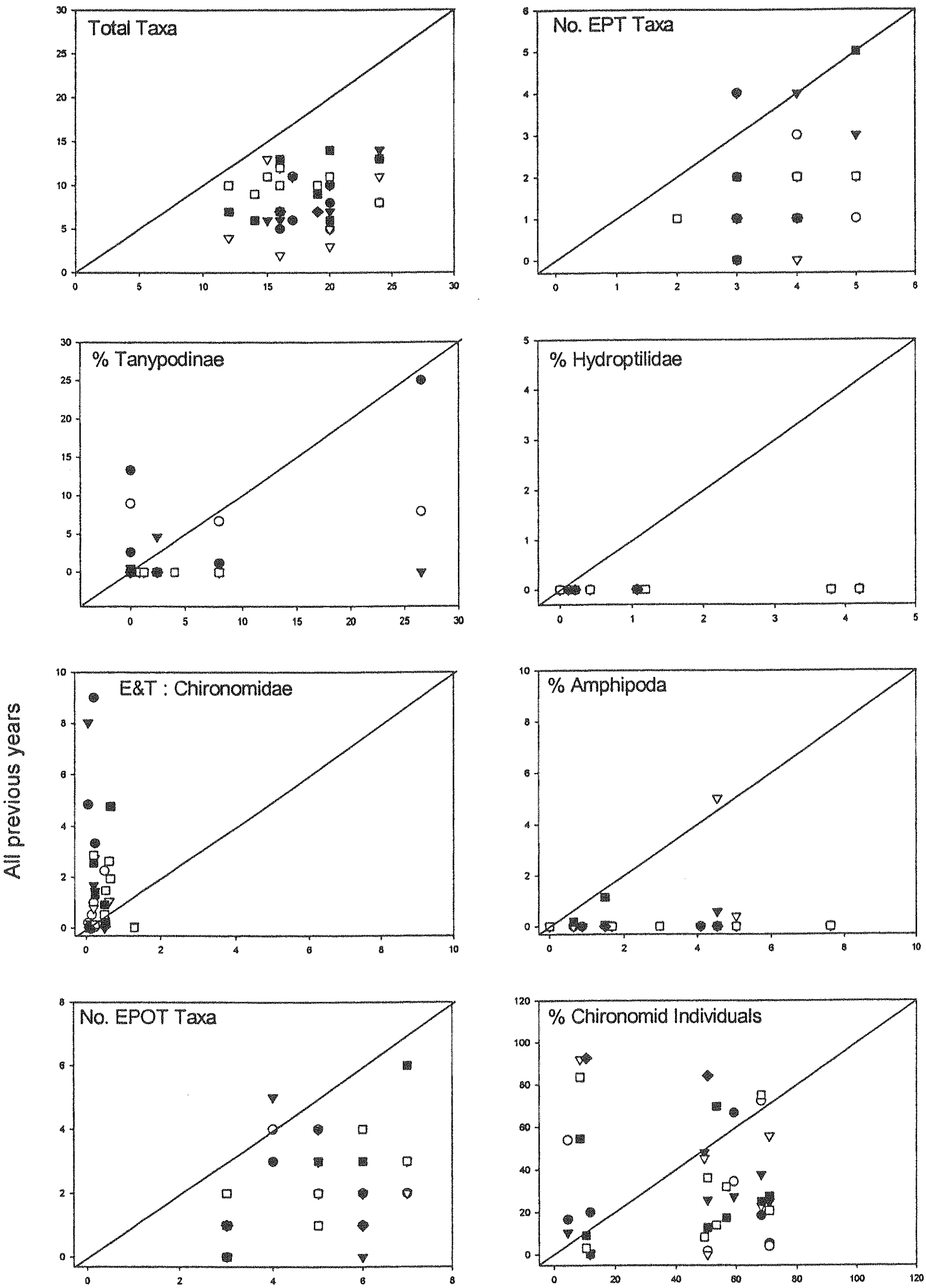

Figure 2. Benthic metrics for 2002 rock baskets paired with baskets from the same river kilometer groups for all historic years (1965-1971). Points within a year represent the different river kilometer groups. Values shown below the diagonal line indicate higher values for the 2002 samples. Symbols: ● 2002 vs 1965; ○ 2002 vs 1966; ▼ 2002 vs 1967; ▽ 2002 vs 1968; ■ 2002 vs 1969; □ 2002 vs 1970; ◆ 2002 vs 1971. (This figure continues on p. 569.)

does provide further evidence of improvement within the Ohio River benthic community. Chironomids represent a highly diverse group that exhibits a wide range of tolerance to disturbance and they may be considered a microcosm of the more inclusive macroinvertebrate community. Research by Losos (1984) indicates diversity of chironomid taxa decreased with increased pollution loads, which would be consistent with our findings. However, chironomid taxa richness might also increase under

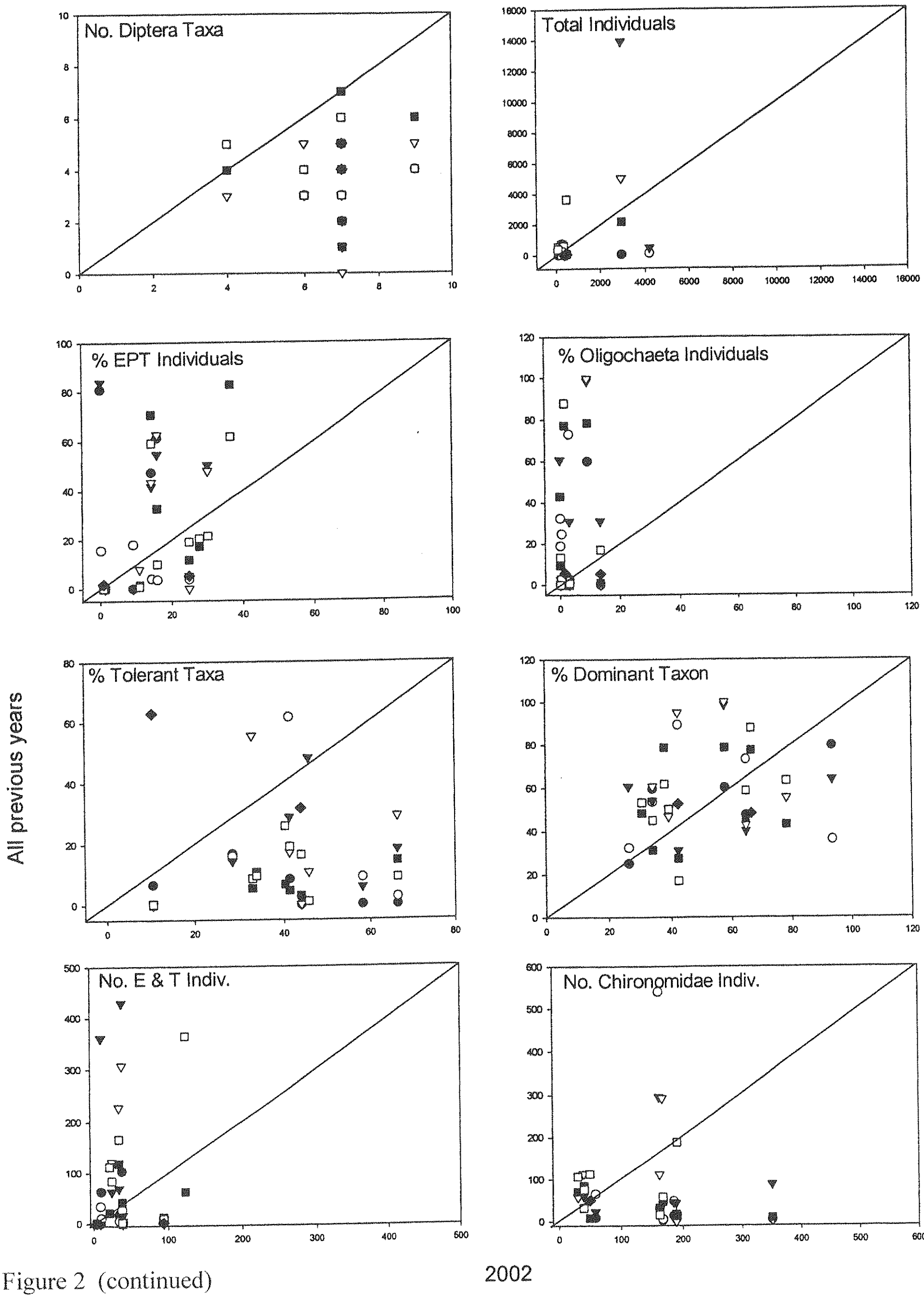

Figure 2 (continued)

moderate pollution, possibly responding to reduced competition with collector-gatherer EPT taxa (Lenat 1983). Oligochaetes are considered highly tolerant, and high densities have long been considered indicative of pollution problems within waterbodies (Kerans and Karr 1994, Barbour et al. 1996). Oligochaetes are particularly abundant in areas of organic enrichment (Pennak 1989) and can dominate the macroinvertebrate fauna of heavily polluted rivers (Eyers et al. 1978). The relative dominance of this group in the historical samples and their reduction within the 2002 samples may result from reduced organic loading from multiple point source discharges along the river.

Evaluation of metric values across years also indicated that the % tolerant taxa metric was higher in 2002 than in previous years, contradicting our general finding of improved river condition. The tolerant taxa metric was adopted from the Ohio EPA and consists of eight taxa generally considered highly tolerant in OH streams and rivers: *Polypedilum illinoense*, *Menetus dilatatus*, *Physella* sp., *Dicrotendipes lucifer*, *Cricotopus bicinctus*, *Chironomus* sp., *Tricladida* sp., and *Glyptotentipes* sp. The metric increase in 2002 is partly due to the collection of *Physella* sp. and *Menetus* sp. when neither of these taxa had been collected in previous years. The proportion of *Dicrotendipes* sp. collected in 2002 baskets was also more than double that collected in most previous years. However, this metric was developed primarily for wadeable streams of Ohio. The new appearance or increased proportion of some of these taxa may not necessarily indicate a degraded condition on a river the size of the Ohio Riverwhere the community is already generally tolerant and has a relatively depauperate fauna when compared with wadeable streams.

The zebra mussel, *D. polymorpha*, is an invasive species that was not collected in the historic rock basket samples. ORSANCO first collected *D. polymorpha* on Hester-Dendy samplers in 1994. Its distribution in the Ohio River is very patchy and this was evident in the 11 rock basket samples collected in 2002 where the number of individuals ranged from 0 to 3920 (mean = 396.7). Its density also varies greatly on an annual basis (M. Wooten, pers. obs.) and the factors controlling these spatial and temporal variations in the Ohio River remain poorly understood. Interestingly, numerous studies have demonstrated that *D. polymorpha* can facilitate other invertebrate taxa by increasing habitat complexity and altering interspecific interactions (Wisenden and Bailey 1995, Ricciardi et al. 1997, Stewart et al. 1998a, and 1998b, Gonzalez and Downing 1999, Greenwood et al. 2001, Kolar et al. 2002, Beekey et al. 2004). Thus, when *D. polymorpha* density is high, this species may be acting as an ecosystem engineer, and its presence may have contributed to the observed changes in the benthic community.

Several other taxa collected in this study were unique to 2002. Though every effort was made for a conservative taxonomic comparison among years, the presence of these taxa only in 2002 may indicate either a natural increase in the populations or differences with identification and sample processing criteria. Because identification of larval midges in particular can be difficult and larval keys were not well-developed at the time of historic sampling, we cannot discount taxonomic discrepancies. However, most of the midge taxa unique to 2002 (e.g., *Parakiefferiella* sp., *Thienemanniella* sp., *Cryptotendipes* sp.) were present in very low numbers (<1% of total individuals) and thus contributed little to observed differences among years in our similarity analyses.

Although many shifts within the macroinvertebrate community were observed during this study, one of the more intriguing aspects was the complete shift within the amphipod assemblage. All amphipod individuals from historic samples were *Crangonyx* sp., though their abundances were typically low. However, samples collected from 2002 were composed entirely of *Gammarus sp.*, with no occurrences of *Crangonyx sp.* This shift has been previously documented for the Ohio River (Beckett et al. 1997). Historical collections of the USEPA (some of which are the same utilized for this study) indicated that *Crangonyx pseudogracilis* was last collected from the Ohio River in 1973, and *Gammarus fasciatus* appeared soon thereafter (Beckett et al. 1997). Beckett et al. (1997) theorized that increased flow regulation was the primary reason for the species replacement, though some evidence also suggests that *Gammarus* sp. can reduce *Crangonyx* sp. densities through intraguild predation (MacNeil et al. 1999). Several studies have also shown that zebra mussels can specifically facilitate *Gammarus* sp. by increasing habitat complexity (e.g., Ricciardi et al. 1997, Gonzalez and Downing 1998, Stewart et al. 1998a, Greenwood et al. 2001); so the shift in the amphipod assemblage may also be partially due to zebra mussel invasion.

Another notable shift in the benthic community occurred within the EPT assemblage. The caddisfly *Cyrnellus fraternus* (Polycentropodidae: Trichoptera) has historically dominated rock basket collections. *C. fraternus* was not only the dominant caddisfly historically, but also the dominant taxon within the entire EPT assemblage. Even though trichopterans are generally considered pollution intolerant, *C. fraternus* has been designated as fairly tolerant (Bode et al. 1996, Mandaville 2002). Though this taxon still constituted a large portion of the EPT assemblage in 2002, there has been a significant decline in proportion compared to historic samples. *C. fraternus* is commonly found in large rivers and may inhabit still waters. Reasons for the general decline of this species are unclear, but Ricciardi et al. (1997) documented a reduction in *Cyrnellus* sp. following establishment of zebra mussel beds. They postulated that zebra mussels may compete with *Cyrnellus* sp. for substrate position and filtration currents.

We identified several major differences in the Ohio River benthic community in 2002 versus the late 1960s and early 1970s. Though our analyses were somewhat limited by the number of comparable samples that were collected historically, our results clearly indicate the benthic assemblage from 2002 was generally more diverse and was comprised of a greater number of relatively sensitive taxa. These changes in the benthic community have resulted from one or a combination of factors that include species introductions, altered competitive interactions, altered habitat due to increased flow regulation, and improved water quality. Our findings suggest that zebra mussels may indirectly affect other macroinvertebrate taxa in ways similarly to those documented for the Laurentian Great Lakes. Yet, further studies are required to measure the degree of zebra mussel facilitation in the Ohio River. The increased effluent regulations required by the 1972 CWA may have had the greatest influence on riverine biota given the large number of municipal and industrial wastewater dischargers located within the Ohio River drainage. Understanding how the benthic community has changed with improved water quality may also aid in development of a modern benthic multi-metric index for assessment of river condition. Metrics that reflect past conditions of assumed impairment (i.e., higher % oligochaetes) may indicate when disturbed conditions are present or possibly reoccurring. Conversely, appearance in 2002 of more sensitive EPOT taxa groups not found in historic samples are indicative of improved condition and may provide valuable assessment metrics. Though our results show general improvement in the benthic community since the late 1960s, it is difficult to establish biocriteria for a large regulated river subject to heavy commercial traffic and multiple point and non-point disturbances. Current assessment methods will likely utilize localized least-disturbed reaches to set an attainable level of expectation with the realization that the ecosystem is permanently altered by human activities

ACKNOWLDEGEMENTS

We extend special thanks to those individuals who assisted with field collections, especially Jeff Thomas, Dan Phirman, Erin Overholt, Stacey Cochran and Finn Pillsbury. We also thank Karen Blocksom for providing statistical expertise and valuable comments regarding the contents of the manuscript, Jim Green for providing details of the original sampling design, Randy Bruins and Florence Fulk of USEPA and ORSANCO's

biological water quality subcommittee for providing helpful comments. Although this work was reviewed by USEPA and approved for publication, it may not necessarily reflect official Agency policy.

LITERATURE CITED

Applegate, J.M. 2002. A preliminary multimetric macroinvertebrate index to assess biotic integrity in the Ohio River. Masters Thesis. The Ohio State University, Columbus, Ohio.

Barbour, M.T., J. Gerritsen, G.E. Griffith, R. Frydenburg, E. McCarron, J.S. White, and M.L. Bastian. 1996. A framework for biological criteria for Florida streams using benthic macroinvertebrates. J. N. Am. Benth. Soc. 15:185-211.

Beckett, D.C. and J.L. Keyes. 1983. A biological appraisal of water quality in the Ohio River using macroinvertebrate communities, with special emphasis on Chironomidae. Mem. Am. Ent. Soc. 34: 15-33.

Beckett, D.C., P.A. Lewis, and J.H. Green. 1997. Where have all the *Crangonyx* gone? The disappearance of the Amphipod *Crangonyx pseudogracilis*, and subsequent appearance of *Gammarus* nr. *fasciatus*, in the Ohio River. Am. Mid. Nat. 139: 201-209.

Beekey, M.A., D.J. McCabe, and J.E. Marsden. 2004. Zebra mussel colonization of soft sediments facilitates invertebrate communities. Freshwater Biol. 49:535-545.

Benke, A.C. 1990. A perspective on Americas vanishing streams. J. N. Am. Benthol. Soc. 9:77-88.

Blocksom, K.A. and J.E. Flotemersch. 2005. Comparison of macroinvertebrate sampling methods for nonwadeable streams. Env. Monit. Assess. 102:243-262.

Bode, R.W., M.A. Novak, and L.E. Abele. 1996. Quality assurance workplan for biological stream monitoring in New York State. NYS Department of Environmental Conservation, Albany, NY, 89p.

Dills, G.D. and D.T. Rogers. 1974. Macroinvertebrate community structure as an indicator of acid mine drainage. Env. Poll. 6: 239-262.

Emery, E.B., T.P. Simon, F.H. McCormick, P.L. Angermeier, J.E. Deshon, C.O. Yoder, R.E. Sanders, W.D. Pearson, G.D. Hickman, R.J. Reash, and J.A. Thomas, 2003. Development of a multimetric index for assessing the biological condition of the Ohio River. Trans. Am. Fish. Soc. 132: 791-808.

Eyers, J. P., N. V. Williams, and M. Pugh-Thomas. 1978. Ecological studies on Oligochaeta inhabiting substrata in the Irwell, a polluted English river. Freshwater Biol. 8: 25-32.

Fullner, R. W. 1971. A comparison of macroinvertebrates collected by basket and modified multi-plate samplers. J. Water Poll. Contr. Fed. 43: 494-499.

Gonzalez, M.J., and A. Downing. 1999. Mechanisms underlying amphipod responses to zebra mussel (*Dreissena polymorpha*) invasion and implications for fish-amphipod interactions. Can. J. Fish. Aquat. Sci. 56: 679-685.

Greenwood, K.S., J.H. Thorp, R.B. Summers, and D.L. Guelda. 2001. Effects of an exotic bivalve mollusk on benthis invertebrates and food quality in the Ohio River. Hydrobiologia. 462; 169-172.

Hester, F.E. and J.S. Dendy. 1962. A multiplate sampler for aquatic macroinvertebrates. Trans. Am. Fish. Soc. 91: 420-421.

Hilsenhoff, W.L. 1987. An improved index of organic stream pollution. Gr. Lakes Entomol. 20:31-39.

Johnson, Z.B., A.K. Riggs, and J.H. Kennedy. 1998. Microdistribution and secondary production of *Cyrnellus fraternus* (Trichoptera: Polycentropodidae) from snag habitats in the Elm Fork of the Trinity River, Texas. Ann. Entomol. Soc. Am. 91: 641-646.

Karr, J.R., R.C. Heidinger, and E.H. Helmer. 1985. Sensitivity of the index of biotic integrity to changes in chlorine and ammonia levels from wastewater treatment facilities. J. Water Poll. Contr. Fed. 57: 912-915.

Kerans, B.L. and Karr, J.R.. 1994. A benthic index of biotic integrity (B-IBI) for riversof the Tennessee Valley. Ecol. App. 4:768-785.

Kolar, C.S., A.H. Fullerton, K., M. Martin, and G.A. Lambert. 2002. Interactions among zebra mussel shells, invertebrate prey, and Eurasian ruffe or yellow perch. J. Gr. Lakes Res. 28: 664-673.

Kruskal, J.B. and M. Wish. 1978. Multidimensional scaling. Sage publications, Beverly Hills, CA. 93 pp.

Lenat, D.R. 1983. Chironomid taxa richness: natural variation and use in pollution assessment. Freshwater Inv. Biol. 2: 192-198.

Lenat, D.R. 1988. Water quality assessment of streams using a qualitative collection method for benthic macroinvertebrates. J. North Am. Benth. Soc. 7: 222-233.

Losos, B. 1984. The influence of pollution on the density and production of Chironomidae (Diptera) in running waters. Limnology 15:7-19.

MacNeil, C., R.W. Elwood, and J.T.A. Dick. 1999. Differential microdistributions and interspecific interactions in coexisting *Gammarus* and *Crangonyx* amphipods. Ecogr. 22: 415-423.

Mandaville, S.M. 2002. Benthic Macroinvertebrates in freshwaters-taxa tolerance values, metrics, and protocols. Soil and Water Conservation Society of Metro Halifax.

Mason, W.T. Jr., P.A. Lewis, and J.B. Anderson. 1971. Macroinvertebrate collections and water quality monitoring in the Ohio River Basin 1963-1967. Report by the Office of Technical Programs, Ohio Basin Region and Analytical Quality Control Laboratory, Water Quality Office, United States Environmental Protection Agency, Cincinnati, Ohio.

Mason, W.T. Jr., C.I. Weber, P.A. Lewis, and E.C. Julian. 1973. Factors affecting theperformance of multiplate macroinvertebrate samplers. Freshwater Biol. 3: 409-436.

Ohio EPA. 1988. Biological Criteria for the Protection of Aquatic Life: Volume II: Users Manual for Biological Field Assessment of Ohio Surface Waters. Ohio EPA. Ref Type: Report

ORSANCO (Ohio River Valley Water Sanitation Commission). 1994. Ohio River Fact Book. ORSANCO, Cincinnati, OH.

Ortiz, J.D., E. Marti, and M.A. Puig. 2005. Recovery of the macroinvertebrate community below a wastewater treatment plant input in a Mediterranean stream. Hydrobiologia. 545: 289-302.

Pennak, R.W. 1989. Fresh-water invertebrates of the United States, 3rd edition. John Wiley and Sons, New York.

Poff, N.L., J.D. Allan, M.B. Bain, J.R. Karr, K.L. Prestegaard, B.D. Richter, R.E. Sparks, and J.C. Stromberg. 1997. The natural flow regime: a paradigm for river conservation. BioSciince. 47: 769-784.

Resh, V.H., R.H. Norris, and M.T. Barbour. 1995. Design and implementation of rapid assessment approaches for water resource monitoring using benthic macroinvertebrates. Aus. J. Ecol. 20:108-121.

Ricciardi, A., F.G. Whoriskey, and J.B. Rasmussen. 1997. The role of the zebra mussel (*Dreissena polymorpha*) in structuring macroinvertebrate communities on hard substrata. Can. J. Fish. Aquat. Sci. 54: 2596-2608.

Schulz, R. and M. Liess. 1999. A field study of the effects of agriculturally derived insecticide input on stream macroinvertebrate dynamics. Aquatic Tox. 46: 155-176.

Stewart, T.W. J.G. Miner, and R.L. Lowe. 1998a. An experimental analysis of crayfish (*Orconectes rusticus*) effects on a *Dreissena*-dominated benthic community in western Lake Erie. Can. J. Fish. Aquat. Sci. 55:1043-1050.

Stewart, T.W. J.G. Miner, and R.L. Lowe. 1998b. Quantifying mechanisms for zebra mussel effects on benthic macroinvertebrates: organic matter production and shell-generated habitat. J. North Am. Benth. Soc. 17:81-94.

Thorp, J.H. 1992. Linkage between islands and benthos in the Ohio River, with implications for riverine management. Can. J. Fish. Aquat. Sci. 49: 1873-1882.

Wallace, J.B., J.W. Grubaugh, and M.R. Whiles. 1996. Biotic indices and stream ecosystem processes: results from an experimental study. Ecol. App. 6: 140-151.

Ward, J.V. and J.A. Stanford. 1995. Ecological connectivity in alluvial river systems and its disruption by flow regulation. Reg. Rivers 11: 105-119.

Whittaker, R.H. 1975. Communities and ecosystems. 2nd ed. MacMillan Publishing Co., New York.

Wisenden, P.A. and R.C. Bailey. 1995. Development of macroinvertebrate community structure associated with zebra mussel (*Dreissena polymorpha*) colonization of artificial substrates. Can. J. Zool. 73:1438-1443.

Received: 9 December 2005 Accepted: 19 February 2006