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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

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OFFICE OF WATER

Dear Colleagues:

The U. S. Environmental Protection Agency is pleased to transmit a copy of the document entitled *Combined Sewer Overflows and the Multimetric Evaluation of Their Biological Effects: Case Studies in Ohio and New York.* This document reports on a project undertaken to measure the biological effects of combined sewer overflows (CSOs). CSOs are discharges to surface waters of mixtures of untreated domestic sewage, industrial and commercial wastewaters, and stormwater runoff. Concern has grown in recent years over the possible adverse ecological effects of CSOs. This concern was reflected in the 1994 CSO Control Policy, which identified the need for characterization of impacts on aquatic life and designated uses.

Aquatic biological communities are exposed to many environmental stressors, which may include point and nonpoint source pollution and habitat alteration or destruction. How the biological communities respond to and integrate these impacts are often difficult to interpret. However, biological assessment methods exist which are designed to evaluate and characterize biological integrity and to identify possible causes of the biological impacts. One of these is an EPA method known as rapid bioassessment protocols (RBPs). RBPs include standardized procedures to assess the biological status and habitat condition of streams, in comparison with minimally impacted streams of the same type. The biological assessment calculates multiple statistics (known as metrics) measuring different attributes of the aquatic community, such as species diversity, food chain relationships, and pollution sensitivity. The metrics are combined into one score of the overall biological status of the community. Interpretation of individual metrics may provide clues to causes of any impairment. Habitat assessments are conducted to determine if habitat degradation is a cause of biological impairment, alone or in combination with water quality problems. It consists of standardized methods to evaluate stream and riparian features important to healthy aquatic communities.

These case studies were carried out in Ohio and New York, both of which have well-established biological monitoring and assessment programs and which use methods similar in approach to RBPs. The availability of historic data allowed comparison of results between studies. The report also explores whether different levels of effort within the RBP framework affected the results. The purpose of this was to determine if using smaller sample sizes or a lower level of detail in organism identification would be sufficient for some purposes such as screening studies and establishing priorities. A final objective was to address possible applications of the RBP methodology in other aspects of watershed protection.

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This document should not be construed as Agency guidance or policy, or as a requirement to use the RBP methodology. Rather, the intention of this document is to provide information on potential applications of RBPs and biological assessments. The document is aimed at state and local biologists and managers looking for potential tools to assess the biological effects of CSOs. It can be a tool to help prioritize limited resources where the CSO impacts are the greatest and where controls would do the most good.

Applications of RBPs are not limited to CSOs, however. Biological assessments have useful applications in various watershed protection approaches such as the TMDL process, 305(b) reporting, stormwater monitoring, and development of biological criteria. Bioassessments are useful screen tools for identifying and prioritization impaired waters. They may be able to provide an indication of causal relationships for different types of impairment such as habitat degradation, toxic loading, and organic enrichment. Finally, they may be useful in assessing how effective pollution control measures are in protecting aquatic life and biological integrity.

Requests for additional copies should be sent to U.S. Environmental Protection Agency, National Center for Environmental Publication and Information, 11029 Kenwood Road, Building 5, Cincinnati, Ohio 45242 (513-489-8190), or by email (Waterpubs@epamail.epa.gov.). Please refer to the EPA document number (EPA 823-R-96-002). For more information call Marjorie Coombs at 202-260-9821 (or via the Internet: coombs.marjorie@epamail.epa.gov).

We appreciate your interest in biological assessment and watershed management.

Sincerely,

Tudor T. Davies, Director
Office of Science and Technology

United States
Environmental Protection
Agency

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Washington, DC 20460

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April 1996

EPA

Combined Sewer Overflows and the Multimetric Evaluation of Their Biological Effects: Case Studies in Ohio and New York

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United States Environmental Protection Agency
Office of Water
Washington, DC 20460

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Contents

Figures iii
Tables v
Acknowledgments vii
Executive Summary ix
Abstract xiii

Introduction 1
1.1 Document Purpose 1
1.2 Environmental Effects of CSOs 1
1.3 Biological Assessments 2
1.4 Reference Conditions 3

Methods 5
2.1 Habitat Quality Assessment 5
2.2 Benthic Macroinvertebrate Sampling 5
2.2.1 Sampling and Sample Handling 5
2.2.2 Taxonomy 7
2.2.3 Counting 8
2.3 Data Analysis 8
2.3.1 Development of Bioassessment Scoring Criteria 8
2.3.2 Metrics 8
2.4 Quality Assurance/Quality Control 10

Evaluating the Biological Effects of Combined Sewer Overflows in Ohio 11
3.1 Site Selection and Location Description 11
3.2 ResultsTaxonomy and MetricsThe Scioto River at Columbus, Ohio 11
3.2.2 The Scioto River at Columbus, Ohio 11
3.2.2.1 Historical Information 15
3.2.2.2 Sampling Station Descriptions and Habitat Quality Assessments 15
3.2.2.3 Biological Assessments 17
3.2.2.4 Comparison to Historical Assessments 18
3.2.3 The Sandusky River at Bucyrus, Ohio 19
3.2.3.1 Historical Information 19
3.2.3.2 Sampling Station Descriptions and Habitat Quality Assessments 19
3.2.3.3 Biological Assessments 23
3.2.3.4 Comparison to Historical Assessments 23
3.2.4 The Little Cuyahoga River at Akron, Ohio 23
3.2.4.1 Historical Information 24
3.2.4.2 Sampling Station Description and Habitat Quality Assessments 25
3.2.4.3 Biological Assessments 27
3.2.4.4 Comparison to Historical Assessments 28

Evaluating The Biological Effects of Combined Sewer Overflows in New York 31
4.1 Site Selection and Location Description 31
4.2 Results 31
4.2.1 Taxonomy and Metrics 31
4.2.2 Canastota Creek at Canastota, New York 33

4.2.2.1 Historical Information 33
4.2.2.2 Sampling Station Description and Habitat Quality 34
4.2.2.3 Biological Assessments 36
4.2.3 Harbor Brook in Syracuse, New York 37
4.2.3.1 Historical Information 37
4.2.3.2 Sampling Station Description and Habitat Quality Assessments 38
4.2.3.3 Biological Assessments 40
4.2.4 Onondaga Creek in Syracuse, New York 41
4.2.4.1 Historical Information 41
4.2.4.2 Sampling Station Descriptions and Habitat Quality 41
4.2.4.3 Biological Assessments 43

Evaluation of Method Variation **45**
5.1 Adequacy of Screening Level (Rapid Bioassessment Protocol I) 45
5.2 Metric Performance with Variable Methods 45
5.2.1 Taxonomic Level Effects on Metric Performance 47
5.2.2 Subsampling Level Effects on Metric Performance 51
5.3 Summary of Results 57

Conclusions/Recommendations **61**
6.1 Historical Assessment Comparisons 61
6.2 Statistical Comparisons 61
6.2.1 Taxonomic Level — Conclusions 62
6.2.2 Subsample Size — Conclusions 62
6.3 Usefulness of RBPs in Assessing CSO Biotic Effects 62
6.4 The Place of Bioassessment in Watershed Protection 63

Literature Cited **65**

Figures

2-1 Habitat assessment field sheets; riffle/run prevalence.

3-1 State of Ohio; three river systems within which the CSO study occurred.

3-2 Cities of Columbus and Circleville, Ohio.

3-3 Linear comparison with Ohio EPA assessments on the Scioto River.

3-4 Percent Comparability of Biota and Habitat with Reference Conditions.

3-5 Cities of Bucyrus and Melmore, Ohio.

3-6 Linear comparison with Ohio EPA assessments on the Sandusky River.

3-7 City of Akron, Ohio.

3-8 Linear comparison with Ohio EPA assessments on the Little Cuyahoga River.

4-1 State of New York.

4-2 Locations of sampling stations on Canastota Creek.

4-3 Locations of sampling stations on Onondaga Creek, Harbor Brook, and Furnace Brook.

4-4 Location of sampling station on the Tioughnioga River (West Branch).

5-1 Correlational scatterplot (1:1) of bioassessment score, family vs. genus/species level taxonomy

5-2 Correlational scatterplot (1:1) of taxa richness, family vs. genus/species level taxonomy.

5-3 Correlational scatterplot (1:1) of Hilsenhoff Biotic Index, family vs. genus/species level taxonomy.

5-4 Correlational scatterplot (1:1) of percent contribution of dominant taxon, family vs. genus/species level taxonomy.

5-5 Correlational scatterplot (1:1) of Pinkham-Pearson Community Similarity Index, family vs. genus/species level taxonomy.

5-6 Correlational scatterplot (1:1) of bioassessment score, 100 vs. 300 organism subsample.

5-7 Correlational scatterplot (1:1) of taxa richness, 100 vs. 300 organism subsample.

5-8 Correlational scatterplot (1:1) of scraper/(scraper + filterer collector), 100 vs. 300 organism subsample.

5-9 Correlational scatterplot (1:1) of no. shredders/total sample, 100 vs. 300 organism subsample.

Tables

3-1 Biological sampling stations located in Ohio.

3-2 Scoring criteria developed for the benthic macroinvertebrate assemblage on each Ohio study river using USEPA's rapid bioassessment protocols.

3-3 Habitat assessments and physicochemical measurements of the Scioto River taken on 8 September 1992.

3-4 Calculated biological metrics based on 300-organism subsamples from double-composite 1-m^2 kicknet samples.

3-5 Habitat assessments and physicochemical measurements of the Sandusky River taken on 9 September 1992.

3-6 Habitat assessments and physicochemical measurements of the Little Cuyahoga River taken on 24 September 1992.

4-1 Biological sampling stations located in or near Syracuse, New York.

4-2 Scoring criteria developed for the benthic macroinvertebrate assemblage using RBPs in New York.

4-3 Metric values calculated from 300-organism subsamples.

4-4 Primary taxonomic composition of benthic macroinvertebrate samples taken by Preddice.

4-5 Canastota Creek habitat assessment scores.

4-6 Harbor Brook habitat assessment scores.

4-7 Onondaga Creek habitat assessment scores.

5-1 Narrative screening-level assessments of ten study stations in New York State.

5-2 Comparison of biological assessments between RBPI and RBPIII.

5-3 Family-level metric values calculated from 100-organism subsamples.

5-4 Genus/species level metric values calculated from 100-organism subsamples.

5-5 Significance of comparisons between taxonomic treatments of 100-organism subsamples at ten sampling stations in New York.

5-6 Significance of comparisons between treatments of samples from ten stations in New York.

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Executive Summary

Combined sewer overflows (CSOs) are direct discharges into wetlands, lakes, coastal waters, streams, and rivers of untreated domestic, commercial, and industrial waste and wastewaters, and urban storm water runoff. They have recently received increased national attention because they are recognized as a primary contributor to water quality degradation in some urban areas, as identified by the President's Clean Water Initiative.

CSOs may have deleterious effects both on the designated recreational uses because of the pathogens found in raw sewage, and on the designated aquatic life uses because of adverse impacts on the biological community. These case studies were initiated to examine the effects of CSOs on the biological integrity of some example streams, using an established EPA protocol for biological assessment.

These projects focused upon several objectives:

1. Evaluation of the effectiveness of rapid bioassessment protocols (RBPs) for detecting biological responses to combined sewer overflows;

2. Comparison with historical assessments performed by the Ohio Environmental Protection Agency and the New York Department of Environmental Conservation;

3. Comparison of results from different levels of assessment rigor, in particular, of taxonomic identification level and subsample size; and

4. Evaluation of the potential application of bioassessment methods to the Total Maximum Daily Load (TMDL) process and other watershed protection approaches.

These case studies are intended for use by state bioassessment personnel, CSO management and control staff, and regional watershed protection coordinators. However, this document should not be construed as Agency guidance or policy, or as a requirement to use the RBP methodology in any given situation.

RBPs were applied at a total of 23 sampling stations in 10 streams and rivers in Ohio and New York. In Ohio, a subsample (300 organisms) was taken from each of 11 benthic macroinvertebrate samples; in New York, two subsamples (100 organisms and 200 organisms) were taken from each of 12 samples.

RBPs include a procedure to assess habitat quality, which was employed at each location. The procedure evaluates stream and riparian habitat features important to healthy aquatic communities such as channel width, depth, and sinuosity; instream cover (variety of substrate sizes, woody debris); riparian vegetation and canopy cover; and bank stability. Habitat assessments are conducted in order to determine if habitat degradation is a limiting factor for aquatic communities in the absence of, or in addition to, water quality problems.

RBPs also include an assessment of biological condition, which is based on an aggregation of several metrics calculated from the sampling results. These metrics are attributes of the community of aquatic organisms being sampled and are used to characterize the status of a stream. When compared with reference values, the aggregated metrics are an indicator of ecological condition. The metrics used in these studies include: taxa richness; Hilsenhoff Biotic Index (HBI); ratio of scrapers to filterer collectors; ratio of Ephemeroptera, Plecoptera, and Trichoptera (EPT) to Chironomidae; percent contribution of dominant taxon; EPT index; percent shredders; ratio of Hydropsychidae to total Trichoptera; Pinkham-Pearson Community Similarity Index; Quantitative Similarity Index (QSI)-Taxa; Dominants-In-Common (DIC)-5; and QSI-Functional Feeding Group (FFG).

RBPs were found to be useful in determining biological impairment due to CSOs and additional urban effects. Adverse biological responses to CSOs were identified at all stations downstream from CSO input. Responses included increased abundance of Chironomidae, increased abundance of filterer collectors, decreases in taxa richness, and an increase in HBI values. All of these biological responses indicate a shift from a well balanced community structure to one of increased tolerance of pollution. The responses are characteristic of nutrient and/or toxic loading.

Study areas in Ohio were selected based on the availability of data from previous biological assessments conducted by

the Ohio Environmental Protection Agency (Ohio EPA) on rivers and streams impacted by CSOs. The three areas selected were the Scioto River at Columbus, the Sandusky River at Bucyrus, and the Little Cuyahoga River at Akron.

The Scioto River is a major tributary of the southern Ohio River and has a long history of degradation from a variety of sources including upstream water withdrawals, channel modifications, urban runoff, and input of organic matter, nutrients and toxics from CSOs. Historical monitoring by the Ohio EPA has generally resulted in biological assessment ratings as "poor" or "fair" in the Scioto near Columbus; assessment results from this study are consistent with the historical data. Habitat conditions at each station were judged to be similar so that any biological differences between stations should be due to water quality effects. The two stations within the zone of CSO influence were found to exhibit "moderate" and "slight" impairment relative to the regional reference station. Examination of the individual metrics indicate that the impairment may be due to organic enrichment and an increase in suspended organic particulates. The upstream reference station was found to have slight impairment relative to the regional reference. Review of individual metrics for the upstream station indicate that impairment was likely due to development, road runoff, and other human perturbations occurring upstream and adjacent to this station.

The Sandusky River is a major tributary to Lake Erie which runs through predominantly agricultural land in north central Ohio. Historical biological assessments of the Sandusky River at Bucyrus revealed significant impacts to the fish and macroinvertebrate communities from CSOs and the Bucyrus wastewater treatment plant (WWTP). In 1990, upgrades to the WWTP were made and corresponding improvements were reported in the biological condition. However, further historical assessments as well as current assessments indicate that slight impairment of the macroinvertebrate community remains downstream of CSO inputs. Impairment appears to be due to a combination of habitat degradation and water quality impacts associated with CSOs.

The Little Cuyahoga River flows through Akron in northeastern Ohio. The study area begins downstream of the Mogadore Reservoir. Historical assessments conducted by Ohio EPA indicate "fair" and "poor" biotic conditions due to a combination of urban runoff and organic enrichment problems from lake and wetland drainage. Current biological assessments indicate that the Little Cuyahoga has moderate biological impairment at the farthest downstream station; the upstream station was also assessed as having biological degradation. Habitat conditions were somewhat degraded at all stations along the Little Cuyahoga but were comparable at all three sites. Biological impairments at the downstream stations can thus be attributed to water quality. There was a distinct depression in overall biological condition at farthest downstream station, including decreased abundance and low diversity. This may possibly indicate the presence of toxicants contributed by CSO and/or industrial inputs. The middle station was originally expected to have been impacted by CSOs; however, the study results indicate improved conditions over the historical assessments. Further investigations revealed that the CSO outfalls upstream of the middle station had been recently eliminated. The biotic improvement over time shown at this station reflected their removal.

Three streams were also selected for the New York case study, Canastota Creek, Harbor Brook, and Onondaga Creek. These streams were selected by New York Department of Environmental Conservation for their known CSO-inputs and relevant historical assessment information.

Historical assessments of Canastota Creek indicate inputs of toxics as well as organic enrichment. Recent assessments (1990) indicate moderate impacts to the macroinvertebrate community in Canastota Creek. The current study found that the upstream station and the first CSO station were slightly to moderately impaired, likely due in part to organic enrichment occurring upstream of any CSO impacts. The downstream station was moderately impaired. Although the biological assessment score of the middle station was similar to that of the upstream station, examination of individual metrics found that the middle and downstream stations had a higher proportion of individual organisms considered to be pollution-tolerant, which is probably a response to CSO influence.

Habitat assessments on Harbor Brook indicated moderate impacts and severe impacts at the upstream and middle stations, respectively, as demonstrated by poor species richness and the high abundance of tolerant taxa. The results of the current study are consistent with these historical findings. Habitat conditions at the middle and downstream stations were very poor and the station farthest downstream on Harbor Brook was unable to be sampled due to severe habitat alterations (channelization), deep slow moving water, and a very soft bottom. The screening level assessment conducted at this site indicated severe biological impairment. Both the middle and downstream stations contained taxa considered to be tolerant to pollution and habitat degradation.

Historical assessments on Onondaga Creek correspond well to assessments conducted at the downstream station of the current study; both assessments indicated moderate to moderately-severe impairment. The upstream and middle stations on Onondaga Creek were found to be moderately impaired likely due to organic enrichment and habitat degradation.

The effectiveness of RBPs for detecting biological responses to CSOs was demonstrated through these case studies. Although "cause-and-effect" relationships are complicated by other problems associated with urbanization, such as habitat degradation and potential industrial discharges, reasonable support for attributing biological impairment to CSO effects was possible. Impairment due to CSO outfalls was noted in biological data in the historical assessments conducted by Ohio EPA and NYDEC, as well as in the current studies for all of the streams assessed. The upstream stations in the Scioto River, the Little Cuyahoga River, Canastota Creek, and Harbor Brook were all located in urbanized areas, yet the biological communities were of a high enough quality in comparison with the downstream stations to indicate that CSO outfalls had adverse effects on the macroinvertebrate communities.

Comparisons between the current studies and historical biological assessment results proved to be valuable; consistent comparisons were made with most historical assessments. In one instance where there were differences between historical and current results, i.e., the Little Cuyahoga River, the improvement in the biological assessment appears to be the result of removal of the CSO outfalls in that section of the river. Different sampling gears were used between the current and historical studies, therefore, only overall assessment results could be compared. Evaluation of how individual metrics or actual quantitative data differed among asessments was not possible.

Comparisons of individual metric values between different taxonomic levels showed some variability; however, total bioassessment scores (comparative ranking of sites) showed no difference. The appropriate level of taxonomic identification for a study is based on the study objectives; for other than screening-level assessments, the lowest possible level of identification is suggested. Several metrics use functional feeding group and tolerance value designations for their calculation (scraper-filterer collector ratio, percent shredders, QSI-FFG, and HBI). These are based on the knowledge of the ecology of macroinvertebrates at the species level. Therefore the uncertainty associated with the assignment of functional feeding group and tolerance value is greater the less detailed the identification is (e.g., genus, family, or order as opposed to species).

Subsample size had little effect on the rank order of total bioassessment scores. Metrics based on some form of taxa richness were variable with different subsample sizes, as expected, due to the increased probability of rare taxa being included in the larger subsample. However, as long as the test site and reference sites are treated in the same manner (i.e., same subsample size and taxonomic level), the biological assessment will be valid. Subsamples of 100 organisms are recommended in New York when using multimetric assessment approaches.

Biological assessments have useful applications in various watershed protection approaches such as the TMDL process, 305(b) reporting, stormwater monitoring, and development of biological criteria. Bioassessments are useful screening tools for identifying and prioritizing impaired waters. They may be able to provide an indication of causal relationships for different types of impairment such as habitat degradation, toxic loading and organic enrichment. Finally, they are useful in assessing how effective pollution control measures are in protecting aquatic life and biological integrity.

A limitation of this study is that, in nearly all cases, the farthest upstream stations showed some kind of impairment. Using impaired upstream stations as the control will often cause the downstream "affected" stations to appear better than they actually are. For increased accuracy, it is recommended that bioassessments use reference conditions composed of multiple reference sites, as opposed to single upstream reference sites.

Abstract

Combined sewer overflows (CSOs) are uncontrolled discharges, during wet and dry weather, of mixtures of untreated domestic sewage, industrial and commercial wastewaters, and stormwater runoff. There has been increasing interest in the effects of these discharges on the water quality and ecological integrity of surface waters receiving them. This document presents a discussion of the components of pollution produced by CSOs, the use of USEPA's rapid bioassessment protocols (RBPs) for evaluating instream community level effects on the benthic macroinvertebrate assemblage, and the potential for using bioassessment results in the total maximum daily load (TMDL) process, 305(b) reporting, biological criteria, and other watershed management efforts.

Application of the RBPs is presented in two case studies, in Ohio and New York, where assessments were completed and the results compared with historical assessments by the Ohio Environmental Protection Agency (Ohio EPA) and the New York Department of Environmental Conservation (NYDEC). Overall, the current assessments in Ohio are relatively consistent to Ohio EPA's assessments in 1986, 1988 and 1991; some assessment results varied slightly between the 1991 and 1992 surveys. The current assessments in New York are comparable to previous studies conducted by NYDEC in 1989 and 1990.

Also presented is an evaluation of the effects of the level of taxonomic identification and subsampling level on RBP results. When we compared two versions of the RBP methodology which employ different levelsof identification (family vs. genus or species), seven individual metrics showed variability with the changing taxonomic level while the total bioassessment scores were not affected. Results using family level identifications may be less sensitive than genus/species level for those metrics that depend on tolerance values and functional feeding group designation. Although the total bioassessment scores were not affected, the variability of the individual metrics, and lower taxonomic resolution, can lead to difficulties in interpreting the findings of the total bioassessment scores when family level identification is used. Comparisons between two different subsample sizes (100 and 300 organisms) also showed no differences in the total bioassessment scores; only two metrics (taxa richness and EPT index) performed differently between the subsampling efforts.

The results presented indicate that bioassessments, in general, and RBPs, specifically, are found to be effective in detecting the biological effects of CSOs.

1 Introduction

Combined sewer overflows (CSOs) are increasingly being recognized as significant sources of water quality impairment in some urban areas of the United States. Several factors have contributed to CSOs not being adequately controlled despite the fact that they are covered under the Clean Water Act's National Pollutant Discharge Elimination System (NPDES) permitting requirements. They are a highly complex, site-specific technical problem that is expensive to control, and the U.S. Environmental Protection Agency (USEPA) has historically focused on regulation of single chemical pollutants (Water Policy Report 1994).

Combined sewer systems are state or municipally-owned wastewater collection systems that channel sanitary wastewaters and stormwater to a treatment facility. CSOs are discharges from the sewer system prior to the treatment facility of mixtures of untreated domestic sewage, industrial and commercial wastewaters, and stormwater runoff. CSOs usually result from a lack of sufficient storage capacity at times of high precipitation. They often carry high concentrations of bacteria and other microorganisms, suspended solids, toxic pollutants, floatable solid wastes, oil and grease, nutrients, and oxygen-demanding organic compounds (USEPA 1994a).

1.1 Document Purpose

One of the purposes of this paper is to investigate a potential tool for characterizing the biological effects of CSOs. It is hoped that such a tool would aid in achieving the characterization and monitoring portion of the Long-Term Control Plans. Part of the Long-Term Control Plan is to use cost-effective screening procedures for identifying relative degrees of impairment to the ecosystem; biological monitoring provides a mechanism for this. Additional objectives of the paper are to present two case studies in which biological assessments were used to evaluate CSO impacts, to investigate the effects of variation in sampling and analysis methodology on assessment results, and to examine potential application of bioassessment methods to the total maximum daily load (TMDL) process and other watershed management efforts. These efforts may include development of biological criteria, storm water and wet weather monitoring, and preparation of 305(b) reports, which are biennial reports prepared by each state to report the status of the state's waterbodies. The audience for this document is intended to be state bioassessment personnel, programmatic staff overseeing CSO management and control, and regional watershed protection coordinators.

1.2 Environmental Effects of CSOs

Many of the limited existing data on CSOs are measurements of effluent levels of physicochemical water quality parameters (i.e., they measure stressors in the CSO directly). Stressors contained in CSOs may be physical (e.g., elevated temperatures, high velocity, heavy solids load), chemical (e.g., organic loading, biochemical oxygen demand, toxic pollutants), or biological (fecal coliforms) in nature. The high energy and intermittent flows characteristic of CSO discharges result in several physical effects in the receiving waterbody, among them scouring of the substrate, bank destabilization and erosion, and changes in the morphometry (shape) of the waterbody (e.g., increased channelization). The problems are probably most evident in lotic (flowing) waters, and particularly where there is a steep topographical gradient. The magnitude of the physical changes in the waterbody is dependent on the topography and geology of the area (e.g., how easily the substrate is eroded), the volume and flow of the discharge, the intensity of the storm event(s), and the amount of increase over "normal" flow. It should be noted that these physical effects are a function of the wet-weather flows and discharges, not CSOs in particular; storm water discharges can exert similar effects.

Numerous biological effects can occur in the aquatic ecosystem from the high flow. There might be an immediate, direct loss of organisms and their habitats. For example, in streams and rivers, plants and animals might not be able to withstand the greatly increased flows and might be swept downstream (Seager and Abrahams 1990), where they might or might not find suitable habitat. The high and intermittent flows could preclude the establishment or maintenance of vegetated areas once they have been uprooted or undermined by the flow, and curtail recolonization by benthic organisms after downstream

drift. Thus, the loss of habitat and organisms might be perpetuated.

CSO discharges are usually warmer than the receiving waterbody, especially in summer. Moreover, urban streams often lack shade, which raises ambient summertime temperatures. The heavy sediment load in CSOs can influence heat radiation in the water column (USEPA 1992), possibly by increasing heat retention by the particles in the water column, thus maintaining the elevated temperatures. Warm water cannot hold oxygen in solution as well as cold; therefore, an indirect result of elevated temperature is lower dissolved oxygen in the water column.

While suspended in the water column, particulate matter results in increased turbidity and reduced light penetration. Ambient light levels can be further lowered by color generated by materials in the discharges (or produced later by subsequent algal blooms). Much of the material in CSOs and storm water/runoff is relatively large (Field and Turkeltaub 1981). In such a case the majority of the material would settle out relatively quickly and light levels could return to normal. If there is a significant percentage of fine-grained silt and clays, however, the settling rates are much slower and the elevated turbidity levels can be more or less permanent. The high flows characteristic of CSOs can often cause a resuspension of potentially contaminated sediments (including microbes and pathogens, toxic substances, and metals) deposited from earlier storms.

CSOs have high levels of organic matter, which contribute to biochemical and chemical oxygen demand (BOD, COD) and thus to dissolved oxygen (DO) depletion in the water and sediments. There appear to be immediate and delayed stages in the high oxygen demand dynamics. There is an immediate (i.e., during the storm event) peak of COD (Ellis et al. 1992), due to the physical forces that scour, flush, and resuspend the sediment and associated material and due to the relatively rapid degradation of the dissolved organic compound portion of BOD. The delayed effects are due to the degradation of the BOD associated with the particulate matter (Lijklema et al. 1990; Hvitved-Jacobsen 1982), which is more refractory.

The toxic contents of CSOs are not well characterized because they are site-specific, storm-specific, and dependent on the relative proportions of the industrial waste, domestic waste, and storm water components along with the individual characteristics of each component. However, numerous constituents that are highly toxic to aquatic life have been documented in CSOs . These include heavy metals (copper, lead, zinc, etc.), PAHs, and pesticides. Non-priority pollutant toxic substances are also found. Ammonia might be present in the discharge itself, shown by peaks in instream NH_3-N concentrations during a storm event (Ellis et al. 1992). Ammonia might also be generated within the sediment and released to the water column. Also present are oil, grease, and gasoline, which have toxic effects of their own and might be further contaminated with various priority pollutants. There might be whole-effluent toxicity due to mixtures or unknown constituents as well.

1.3 Biological Assessments

Biological assessments provide integrated evaluations of water resource quality. They also can allow inferences to be drawn from a broad array of stressors based on both biological and physical habitat conditions. Impairments can be identified from a variety of sources including water column contamination, sediment contamination, nonchemical impacts, and alteration of physical habitat (Karr 1991). The instream communities act as continuous monitors of water quality, assimilating impacts from periodic spills, nonpoint source pollution, cumulative pollutants, and other sources that might be missed during sporadic chemical sampling (Ohio EPA 1987a; USEPA 1990a). Responses to natural habitat variability and impacts from intermittent physical habitat change precipitated by phenomena such as increased stormflows (e.g., sedimentation, scour, and modified flow characteristics) will also be reflected by the biological community (Heins 1991; Burton and Harvey 1990; Holomuzki 1991; Chambers et al. 1991; Jowett and Duncan 1990; Burns 1991; Plafkin et al. 1989; Barbour and Stribling 1991; Karr et al. 1986; Ohio EPA 1987b). Because of the unpredictable and fluctuating nature of storm events in urbanized watersheds (Schueler 1987), characterization of the biological community might provide a good measure of the cumulative instream effects caused by CSOs and stormwater discharge.

Rapid bioassessment protocols (RBPs) have been developed for determining the status of macroinvertebrate and fish community structure and function in streams and wadable rivers (Plafkin et al. 1989). These methods provide a relatively quick and cost-effective means of compiling and analyzing information on the impairment of aquatic communities from point or nonpoint source pollution. RBPs currently serve as the foundation of the bioassessment approach being adapted by many water quality agencies across the country. Forty-five states have implemented or are developing biological monitoring programs modeled after the RBPs or some other multiple-parameter (multimetric) approach for characterizing benthic macroinvertebrate communities in the context of habitat quality (Southerland and Stribling 1995). The RBP concept is well-founded in ecological principles and uses an information-gathering structure that categorizes and assimilates information into community parameters or metrics through the use of habitat and biological community assessments.

The biological community analysis consists of standardized field collection of benthic macroinvertebrates, and subsequent calculation of a series of "metrics," each measuring a different aspect of community structure and composition. The assessment integrates the metrics and compares them to reference values, allowing judgments to be made on what could be expected at the test site if habitat and pollutant impairments were corrected, as well as the current judgment of overall biotic impairment. The investigator can also evaluate the generic causes of impairments by examining the individual metrics (Yoder 1991; Yoder and Rankin 1995; Shackleford 1988). Different types of organisms have distinct reactions to various types of stresses. For example, metrics which focus on invertebrates that rely on particulate organic matter, such as leaf litter for food, could be used as a screening tool for assessing the impact of bound contaminants or degradation of the riparian vegetation.

Useful metrics for application of RBPs can vary by waterbody type and geographic region (Plafkin et al. 1989; Barbour et al. 1992). Ideally, they are selected based on criteria that would document relevance, sensitivity, responsiveness, and practicality (Barbour et al. 1995). Following pilot studies and evaluation of data and metrics, some might be discarded based on failure to meet pertinent criteria. Although the metrics used for the Ohio and New York studies were taken directly from Plafkin et al. (1989) and Barbour et al. (1992), their use does not necessarily imply that they are the most appropriate choices relative to desirable criteria for metrics, such as responsiveness to environmental degradation. Additional metrics might be more appropriate for assessing CSOs, but developing and testing metrics was beyond the scope of this project.

1.4 Reference Conditions

RBPs are based on the concept of comparison between a study area and a reference condition or site. A reference condition is the set of conditions of minimally impaired waterbodies characteristic of a waterbody type for a given region or subregion (Gibson 1994). The *reference condition* is made up of data from reference sites in a geographic area (or "ecoregion") for waterbodies of the same class and serves as the benchmark for determining the biological potential of test sites in that geographic region and of the same class; it gives more accurate description of expected conditions and the natural variability than do site-specific reference sites. Regional calibration of metrics allows for fine tuning of biological information so that the most appropriate metrics are used for each specified ecological stratum (e.g., type of waterbody) and the regional boundaries for metric variability are recognized.

A *reference site* is a specific locality on a waterbody that represents the expected biological integrity for other sites on the same (site-specific reference site) or nearby waterbodies (regional reference site). Site-specific reference sites have the potential to be affected by stressors affecting the watershed. For that reason, we currently recommend that several reference sites be used for comparisons if reference conditions have not yet been developed for the region and site class. As more site-specific reference sites are sampled and metrics tested and calibrated, they will serve as the foundation for building a reference condition database for waterbodies in the same class and region. Further discussion on the topic of ecological reference conditions and site-specific reference data can be found in Hughes (1995).

The current study used one site-specific (upstream) reference site and one regional reference site as the benchmark to determine the biological impairment of the test sites. In some cases the regional reference site was determined to be unsuitable for use as a reference due to impaired biological condition; in these cases the site-specific reference site was used for comparison. For the current Ohio study, single regional reference sites were used in addition to the upstream reference sites; however, the historical assessments for Ohio are based on the regional reference condition. Two of the three rivers in the current Ohio study (Scioto and Sandusky) are in the same ecoregion (Eastern Cornbelt Plains) and thus might not have required separate scoring criteria if regional calibration had been performed. This could be the source of some differences in the biological assessment for some sites between historic and current assessments. For the most part, regional reference conditions provide more general criteria for acceptable biological integrity.

2 Methods

2.1 Habitat Quality Assessment

Habitat quality assessment is an essential part of any assessment of ecological integrity (Karr et al. 1986; Plafkin et al. 1989). The quality of the physical habitat at a site identifies constraints on the attainable biological potential of that site and provides information for interpreting biosurvey results (Barbour and Stribling 1991). Numerous components of the physical structure of stream environments and riparian habitat are critical to the ecological integrity of lotic water resources, including channel morphology (width, depth, and sinuosity); floodplain shape and size; channel gradient; instream cover (boulders, woody debris); substrate type and diversity; riparian vegetation and canopy cover; and bank stability.

Specific habitat parameters and narrative descriptions of the condition categories for which visual assessments of condition are made are shown in Figure 2-1. Some scoring systems have some habitat characteristics weighted more heavily than others. For instance, the parameter condition scoring framework (Barbour and Stribling 1991) used for the 1992 Ohio study had differential weighing for the primary, secondary, and tertiary parameters with a maximum of 20, 15, and 10 points, respectively. However, with the testing of habitat assessment consistency among multiple observers (Barbour and Stribling 1994), it became evident that the weighing could be a substantial source of variability. The habitat scoring systems currently recommended have all parameters weighted equally (Figure 2-1); that is, on a 20-point scale. The scoring system used in New York used equal weighing.

Parameters are visually inspected at each sampling location and assigned scores within the continuum of conditions ranging from optimal to poor based on the narratives. The scores assigned to each parameter are totalled for a station. That score is compared to the reference score to provide a relative assessment of habitat quality that will assist in the interpretation of biological condition. The total score for each sampling station is used in classifying the station, based on the percent comparability to the reference condition ("expected" condition) and the station's apparent potential to support the same level of biological community development as that observed at the reference station. Basic water quality data (temperature, dissolved oxygen, pH, and conductivity) are also collected to allow for further comparison among sites. Further discussion of the logic and justification for the approach can be found in several other documents (Plafkin et al. 1989; Barbour and Stribling 1991, 1994).

2.2 Benthic Macroinvertebrate Sampling

For the benthic macroinvertebrate studies, a standardized collection procedure based on RBPs (Plafkin et al. 1989) was used to obtain samples of the macroinvertebrate fauna from comparable habitat types at all stations. Sampling, according to RBPs for high-gradient streams, is focused on what is generally considered to be the most productive of stream systems, riffles and runs. For the New York study, three different RBP level assessments were conducted at each station in order to compare assessment results from the differing levels of effort (RBPI, RBPII, RBPIII).

2.2.1 Sampling and Sample Handling

Samples were obtained using a 1-m^2 kick net (no. 30 mesh, 600 µm openings). Two 1-m^2 samples were collected at each station: one from a fast-water riffle and one from a slow-water riffle. Sampling from both the fast and slow riffle current velocities allows for a broader coverage of variability within the riffle habitat. For those sampling sites which lacked riffles, run areas with cobble or gravel substrate were sampled instead. The two kick net samples from each station were composited in the field, concentrated in a no. 30 (600 µm) sieve bucket, and emptied into a gridded sorting pan for subsampling. For the Ohio portion of the study, the gridded pan was a metal, porcelain-covered pan with numbered grid squares drawn on the bottom. For New York, a change in subsampling methods was made to minimize movement of organisms among grids and increase the standardization of the subsampling effort. The standardized gridded screen (Caton 1991) contains 30 clearly marked squares, each a uniform 6 cm x 6 cm. The gridded screen fits into another slightly larger tray so that water can be added to the sample to allow for even distribution. When the screen is lifted out of the tray, the sample contents settle onto the screen, effectively restricting organism mobility.

Figure 2-1. Habitat scoring system for streams with riffle/run prevalence.

HABITAT ASSESSMENT FIELD DATA SHEET

RIFFLE/RUN PREVALENCE

Habitat Parameter	Category: Optimal	Suboptimal	Marginal	Poor
1. Instream Cover	Greater than 50% mix of boulder, cobble, submerged logs, undercut banks, or other stable habitat.	30-50% mix of boulder, cobble, or other stable habitat; adequate habitat.	10-30% mix of boulder, cobble, or other stable habitat; habitat availability less than desirable.	Less than 10% mix of boulder, cobble, or other stable habitat; lack of habitat is obvious.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
2. Epifaunal Substrate	Well-developed riffle and run; riffle is as wide as stream and length extends two times the width of stream; abundance of cobble.	Riffle is as wide as stream but length is less than two times width; abundance of cobble; boulders and gravel common.	Run area may be lacking; riffle not as wide as stream and its length is less than 2 times the stream width; gravel or large boulders and bedrock prevalent; some cobble present.	Riffles or run virtually nonexistent; large boulders and bedrock prevalent; cobble lacking.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
3. Embeddedness	Gravel, cobble, and boulder particles are 0-25% surrounded by fine sediment.	Gravel, cobble, and boulder particles are 25-50% surrounded by fine sediment.	Gravel, cobble, and boulder particles are 50-75% surrounded by fine sediment.	Gravel, cobble, and boulder particles are more than 75% surrounded by fine sediment.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
4. Velocity/Depth Regimes	All four velocity/ depth regimes present (slow-deep, slow-shallow, fast-deep, fast-shallow).	Only 3 of the 4 regimes present (if fast-shallow is missing, score lower than if missing other regimes).	Only 2 of the 4 habitat regimes present (if fast-shallow or slow-shallow are missing, score low).	Dominated by 1 velocity/depth regime (usually slow-deep).
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
5. Channel Alteration	No channelization or dredging present.	Some channelization present, usually in areas of bridge abutments; evidence of past channelization, i.e., dredging, (greater than past 20 yr) may be present, but recent channelization is not present.	New embankments present on both banks; and 40 to 80% of stream reach channelized and disrupted.	Banks shored with gabion or cement; over 80% of the stream reach channelized and disrupted.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
6. Sediment Deposition	Little or no enlargement of islands or point bars and less than 5% of the bottom affected by sediment deposition.	Some new increase in bar formation, mostly from coarse gravel; 5-30% of the bottom affected; slight deposition in pools.	Moderate deposition of new gravel, coarse sand on old and new bars; 30-50% of the bottom affected; sediment deposits at obstruction, constriction, and bends; moderate deposition of pools prevalent.	Heavy deposits of fine material, increased bar development; more than 50% of the bottom changing frequently; pools almost absent due to substantial sediment deposition.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
7. Frequency of Riffles	Occurrence of riffles relatively frequent; distance between riffles divided by the width of the stream equals 5 to 7; variety of habitat.	Occurrence of riffles infrequent; distance between riffles divided by the width of the stream equals 7 to 15.	Occasional riffle or bend; bottom contours provide some habitat; distance between riffles divided by the width of the stream is between 15 and 25.	Generally all flat water or shallow riffles; poor habitat; distance between riffles divided by the width of the stream is between ratio >25.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0

Figure 2-1. (continued)

Habitat Parameter	Category			
	Optimal	Suboptimal	Marginal	Poor
8. Channel Flow Status	Water reaches base of both lower banks and minimal amount of channel substrate is exposed.	Water fills >75% of the available channel; or <25% of channel substrate is exposed.	Water fills 25-75% of the available channel and/or riffle substrates are mostly exposed.	Very little water in channel and mostly present as standing pools.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
9. Condition of Banks	Banks stable; no evidence of erosion or bank failure.	Moderately stable; infrequent, small areas of erosion mostly healed over.	Moderately unstable; up to 60% of banks in reach have areas of erosion.	Unstable; many eroded areas; "raw" areas frequent along straight sections and bends; on side slopes, 60-100% of bank has erosional scars.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
10. Bank Vegetative Protection	More than 90% of the streambank surfaces covered by vegetation.	70-90% of the streambank surfaces covered by vegetation.	50-70% of the streambank surfaces covered by vegetation.	Less than 50% of the streambank surfaces covered by vegetation.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
11. Grazing or Other Disruptive Pressure	Vegetative disruption, through grazing or mowing, minimal or not evident; almost all plants allowed to grow naturally.	Disruption evident but not affecting full plant growth potential to any great extent; more than one-half of the potential plant stubble height remaining.	Disruption obvious; patches of bare soil or closely cropped vegetation common; less than one-half of the potential plant stubble height remaining.	Disruption of streambank vegetation is very high; vegetation has been removed to 2 inches or less in average stubble height.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0
12. Riparian Vegetative Zone Width (Least Buffered Side)	Width of riparian zone >18 meters; human activities (i.e., parking lots, roadbeds, clear-cuts, lawns, or crops) have not impacted zone.	Width of riparian zone 12-18 meters; human activities have impacted zone only minimally.	Width of riparian zone 6-12 meters; human activities have impacted zone a great deal.	Width of riparian zone <6 meters: little or no riparian vegetation due to human activities.
SCORE ____	20 19 18 17 16	15 14 13 12 11	10 9 8 7 6	5 4 3 2 1 0

Total Score _____

For subsampling, individual grid squares were randomly selected, then organisms were removed from each selected grid until the desired subsample number (300 organisms) was reached. Then any large organic material (whole leaves, twigs, algal or macrophyte mats) was rinsed, visually inspected, and discarded. Randomly selected grid squares were completely sorted regardless of whether the number of organisms was greater than that needed for the subsample. For the Ohio study, organisms were removed from selected girds until the 300-organism subsample was reached. For the New York study, a series of grids were chosen to constitute a 100-organism subsample and a 200-organism subsample for each sample. These subsamples were maintained separately for identification and storage, then the data were totaled to create the 300-organism subsample. Specimens for both studies were placed in a pre-labeled sample container containing 70 percent ethanol and shipped to Monticello Ecological Research Station (University of Minnesota, Monticello, Minnesota) for identification.

2.2.2 Taxonomy

For the RBPIII assessments, all specimens were identified to the lowest practical level, generally genus or species; RBPII assessments used family-level identifications. Both utilized primarily Merritt and Cummins (1984), Wiederholm (1983), Brinkhurst (1986), and Thorpe and Covich (1991). RBPI assessments consisted of field identifications generally to the family level; some identifications were to order.

2.2.3 Counting

For metrics calculated from taxa counts, pupae and adults were not included in the calculations if larvae or nymphs of the same taxon were identified in the sample. For those metrics which use counts of individuals, pupae and adults were included in the calculations. Exceptions are described for the Ohio and New York data in Appendix A and B, respectively.

2.3 Data Analysis

The data were analyzed using the multimetric approach advocated by Karr (1986), Ohio EPA (1987a; b), Plafkin et al. (1989), and Barbour et al. (1995). Metrics were calculated using the 300-organism subsamples from the Ohio study. For the New York study, metrics were calculated based on both 100- and 300-organism subsamples at both family-level and genus/species-level taxonomy. Further rationale for each of these study designs is presented in Sections 3 and 4 of this document.

2.3.1 Development of Bioassessment Scoring Criteria

Bioassessment values derived from each metric are normalized into *bioassessment scores* so that multiple metrics, which yield a wide range of values, can be aggregated. *Scoring criteria* are developed for each class of test sites, stratified by geographic region and stream order, by dividing the metric value range into equal quadrisections ranging from the lowest possible value of a metric (usually zero) to either the maximum value obtained or the 95th percentile. The scoring criteria categories for Ohio were equal quadrisections from the lowest possible value to the maximum obtained. In most cases, the maximum value of a metric was exhibited at regional reference stations or at specific upstream stations. In the New York study, the upper end of the range used was the 95th percentile, which was used to control for outliers.

Using the appropriate scoring criteria table (Sections 3.2.1, 4.2.1), all calculated or enumerated metric *values* were normalized into bioassessment *scores* (0, 2, 4, 6), which were summed for a total bioassessment score. The total bioassessment scores of test sites were then compared to the regional reference sites for each station. The test sites were evaluated on the basis of their percent comparability to the reference values. For two sites, the regional reference site was found to be impaired (e.g., Furnace Brook, New York) or unable to be sampled (flooded) (e.g., Breakneck Creek, Ohio). Therefore, the upstream reference site (station CC1 and CR1, respectively) served as the baseline for comparison. The suitability of both sites for reference were further examined by deriving information from individual metrics and habitat assessment parameters, and the site CC1 was found to be slightly to moderately impaired. CR1 also had a degraded biological condition but was not given a rating. This illustrates the problems which can arise when relying on a single reference site, and therefore that the comparison should, when possible, be made to reference conditions rather than to single reference sites.

Some metrics include data from the reference site in their calculation; these are known as "paired" metrics. For those sites that used the impaired upstream reference sites as a baseline for comparison, paired metrics were not included in the final assessment. When biological scores are summed using paired metrics, the site designated as the reference site receives an automatic score of 6 (the highest score) for each paired metric, which can artificially raise the overall bioassessment score for that site. Therefore, if the reference site is not minimally impaired (i.e., has some degradation as does CR1 and CC1), the site assessment is given a score that indicates better biological condition than it actually has, or would have if compared to a truly minimally impaired site.

In any biological assessment, comparison of total bioassessment scores to reference is but the first step, which is followed by inspection of individual parameters that allow one to identify potential cause-and-effect relationships. The severity of impairment (slight, moderate, etc.) is determined by comparison with minimally impaired conditions. The thresholds for impairment categories are typically some portion of the distribution of the conditions of all sites. For example, the 75th percentile of the range of scores can be considered the cutoff for nonimpairment. To do this correctly, multiple (at least three) reference sites should be used. However, these studies were designed with only an upstream reference site and a regional reference site. Thus, the assignment of narrative impairment categories, in general, is based on those found in Plafkin et al. (1989). However, because the reference sites in New York appeared to have organic enrichment, it was decided that the actual impairment category should be interpreted as one category less than those listed in Plafkin et al. (1989).

2.3.2 Metrics

The metrics used in the biological evaluation of sites include eight "individual" metrics and four "paired" metrics (Barbour et al. 1992). The paired metrics are those which compare the test site to the upstream reference site for the initial calculations. The following is a brief description of the metrics and their calculations. It is worth noting that some descriptions indicate what we expect to find for "good" or "bad" situations for these assessments (based on ecoregions or stream orders). However, the metric value is actually scored good or bad as compared to the reference condition or reference site(s).

1. **Taxa Richness.** Taxa richness reflects the health of the community through a measurement of the total number of taxa present. Taxa richness is calculated by counting the total number of distinct taxa identified in the sample. Generally, taxa richness increases as water quality, habitat diversity, and habitat suitability increase.

2. **Hilsenhoff Biotic Index (HBI).** The HBI was developed by Hilsenhoff (1982) to summarize the various tolerances of the benthic arthropod community with a single value; tolerance values range from 0 to 10, with 10 being assigned to those taxa usually detected in the most degraded situations (i.e., the most tolerant taxa). Only those taxa for which the tolerance values were available were included in these calculations. The formula for calculating the HBI is:

$$HBI = \sum \frac{x_i t_i}{n}$$

where x_i = number of individuals within a taxon,

t_i = tolerance value of a taxon, and

n = total number of individuals in the sample.

Following the Plafkin et al. (1989) document, the HBI was modified to assess the total benthic community not just arthropods and regional development of tolerance values for various environmental pollutants, in addition to organic pollution (Hilsenhoff 1982, 1987; New York State Department of Environmental Conservation, Albany, New York, *in litt* 2/27/89; Illinois Environmental Protection Agency, Marion, Illinois, *in litt* 6/25/86; and Huggins and Moffett 1988). The primary sources for tolerance values and functional feeding group designations were regional when possible (New York State Department of Environmental Conservation, Albany, New York, *in litt* 2/27/89) and USEPA (1990, draft report). Those stations with a lower HBI value are interpreted as being in better condition, having a lower abundance of individuals within tolerant taxa than individuals in sensitive taxa.

3. **Scraper Functional Feeding Group to Scrapers plus Filterer Collectors (Scr/[Scr + Fil] x 100).** The relative abundance of scrapers and filterer collectors reflects the riffle/run community foodbase. When compared to a reference site, shifts in the dominance of a particular feeding type indicate that a community is responding to an overabundance of a particular food source. Scrapers generally increase with increased diatom abundance and decrease as filamentous algae and aquatic mosses increase. However, filamentous algae and aquatic mosses provide good attachment sites for filterer collectors, which may then increase in abundance. The organic enrichment often responsible for overabundance of filamentous algae can also provide fine organic particles used by filterers. This metric reflects biotic response to nutrient overenrichment. Higher values are considered to indicate better conditions.

4. **Individuals of Ephemeroptera, Plecoptera, and Trichoptera (EPT) Taxa to EPT Taxa Plus Chironomidae (EPT/[EPT + Chironomidae]).** This ratio is used as an indication of community balance and compares the number of individuals of Ephemeroptera, Plecoptera, and Trichoptera (mayflies, stoneflies, and caddisflies, respectively) to the number of individuals of EPT taxa plus Diptera: Chironomidae (midges). A relatively even distribution of all four groups indicates a good biotic condition, as does substantial representation of the sensitive groups Ephemeroptera, Plecoptera, and Trichoptera. Environmental stress is indicated by a disproportionately high number of the generally tolerant Chironomidae, reflected by lower values of this metric.

5. **Percent Contribution of Dominant Taxon ([number of individuals of dominant taxon/total number of individuals of all taxa in sample] x 100).** The percent contribution of the dominant taxon uses the abundance of the numerically dominant taxon, relative to the rest of the sample, as an indication of community balance. The lowest practical taxonomic level (assumed to be genus or species in most instances) yields a more accurate assessment value for this metric. A community dominated by only a few species would indicate environmental stress; thus, lower values for this metric are taken to reflect better conditions.

6. **EPT Index.** The EPT Index is the total number of distinct taxa within the Ephemeroptera, Plecoptera, and Trichoptera (mayflies, stoneflies, and caddisflies, respectively) and summarizes the taxonomic richness of three groups of insects that are generally considered to be pollution-sensitive. This value increases with improving water quality.

7. **Shredder Functional Feeding Group to the Total Number of Individuals Collected ([Shr/Total] x 100).** The abundance of the shredder functional feeding group relative to all other individuals allows evaluation of potential impairment to the riparian zone. Higher ratios generally indicate better conditions. Shredders should decrease in abundance if their food source is reduced through habitat alterations or contaminated by toxins.

8. **Hydropsychidae to Total Trichoptera ([H/T] x 100).** Though caddisflies (Trichoptera) as a group are usually considered to be pollution-sensitive, a number of taxa within the Hydropsychidae often greatly increase in abundance and density in degraded and organically-enriched waters. This metric is calculated as the number of individuals of Hydropsychidae to the total number of individuals of Trichoptera in the sample. Higher values reflect a dominance of the hydropsychids (low caddisfly diversity), which indicates poorer water quality.

9. **Pinkham-Pearson Community Similarity Index.** This metric measures the degree of similarity in taxonomic composition between the reference sample and the test sample (Pinkham and Pearson 1976). A higher calculated value reflects a higher degree of similarity to the reference sample and presumably better conditions. It is calculated as:

$$PP = \sum \frac{\text{minimum}(x_{ia}, t_{ib})}{\text{maximum}(x_{ia}, t_{ib})}$$

where x_{ia} = number of individuals in the ith species in sample A

and

x_{ib} = number of individuals in the ith species in sample B.

10. **Quantitative Similarity Index - Taxa (QSI-Taxa).** This measure of comparative similarity in taxonomic composition combined with relative abundance between two sampling stations is based on the concept of "percent similarity" (Whittaker 1952; Bray and Curtis 1957). It has been applied by Shackleford (1988) in Arkansas streams and by others in several individual studies in the mid-Atlantic states. It compares two samples in terms of presence/absence of taxa and relative abundances and is calculated as:

$$S_{ab} = \sum \min(p_{ia}, p_{ib})$$

where p_{ia} = the relative abundance of species i at station A,

p_{ib} = the relative abundance of species i at station B,

and

$\min(p_{ia}, p_{ib})$ = the minimum value of species i at station A or B in terms of relative abundance.

Relative abundance is the percentage of individuals in the total sample that are of species i. Values for these calculations range from 0 to 100. Samples that are identical have a score of 100; those which have nothing in common have a score of 0. Thus, those test stations which are more similar to selected reference conditions have higher index values and are inferred to have better biological condition.

11. **Dominants in Common - 5 (DIC-5).** The DIC-5 compares the five dominant taxa (as in greatest abundance) between the reference station samples and test station samples. For this metric, the top five taxa (numerically) for each of the two samples are listed. The number of taxa shared in the top five list is the metric value. Values for this metric range from 0 to 5 with 5 being most similar to reference and 0 least similar.

12. **Quantitative Similarity Index - Functional Feeding Group (QSI-FFG).** The QSI-FFG compares the relative abundance of functional feeding groups between two samples with the goal of showing changes in the function of a community. This metric is calculated in the same way as QSI-Taxa except that the numbers of individuals are those within functional feeding groups: filterer collectors, gatherer collectors, shredders, scrapers, miners, predators, and parasites.

2.4 Quality Assurance/Quality Control

The quality control elements for the Ohio and New York case studies are provided in Appendix C.

3

Evaluating the Biological Effects of Combined Sewer Overflows in Ohio

A demonstration project was initiated to examine the utility of biological assessment in general and RBPs specifically for evaluating impairment due to CSOs. The study objectives were to:

- Evaluate the impact of CSOs on the benthic macroinvertebrate assemblage at test sites by identifying changes in taxonomic structure, composition, and trophic function;
- Determine the usefulness of RBPs in detecting those effects; and
- Evaluate the agreement of RBPs with historical assessments produced by Ohio EPA.

3.1 Site Selection and Location Description

Three sites that have a history of CSO study were selected for this investigation: the Scioto River at Columbus, the Sandusky River at Bucyrus, and the Little Cuyahoga River at Akron (Figure 3-1). These sites were selected because they represent different regions of the state and are therefore likely to exhibit different biological expectations, and because historical biological data are available. The sites were located with the intention of having one station upstream of any CSO effects, one downstream of all CSO inputs, another far enough downstream to perhaps be in a recovery zone, and a fourth to represent regional reference conditions for each stream (Table 3-1). However, the regional reference site for the Little Cuyahoga River could not be sampled due to flooding; that assessment was based on an upstream condition.

3.2 Results

3.2.1 Taxonomy and Metrics

Taxonomic results and counting exceptions are presented in Appendix A; the results of the metric calculations are shown within the section for each CSO site.

Separate bioassessment scoring criteria were developed for each river under study based on metric values acquired. The scoring criteria are based on equal quadrisections of the value range from the lowest possible value for a metric (usually zero) to the maximum observed, usually observed at the regional reference. The scoring criteria used for each of the three sites are summarized by metric in Table 3-2.

3.2.2 The Scioto River at Columbus, Ohio

The Scioto River is a major tributary of the southern Ohio River (Figure 3-1). It originates in northwestern Ohio in Hardin County in what is known as Scioto Marsh (Ohio EPA 1979). It flows east 60 miles and then south 175 miles to its confluence with the Ohio River at Portsmouth. The Scioto River drainage area, approximately 6,500 square miles, displays a branching stream pattern with tributaries flowing through gorges north of Columbus (Ohio EPA 1986). Flows in the river channel are regulated by two major impoundments and three low-head dams in the central Ohio stretch of the river. Channelization with concrete reinforcement and levees occurs in some of the municipal areas; these channel modifications continue to just upstream of the Jackson Pike Waste Water Treatment Plant (WWTP) (River Mile [RM] 127.1). CSO outfalls are concentrated between RM 132.3 and 129.8 upstream of Jackson Pike. South of Jackson Pike, evidence of impoundment and other channel modifications disappears. The channel is typical of a lotic environment with good sinuosity and riffle-pool sequences. The river is situated over a buried valley filled with glacial outwash material (sand and coarse gravel). Therefore, the substrate ranges from limestone bedrock and silt/muck north of Columbus to coarse sand and gravel/cobble south of Columbus. Flooding in this area has been known to cover extensive areas of the floodplain. This study covers the area of the Scioto from 5 miles upstream of the confluence with the Olentangy River (RM 132.3) to approximately 20 miles south of Columbus at Circleville (RM 100.0) (Figure 3-2). At the northernmost sampling station the drainage area of the Scioto River is approximately 980 square miles; at the southernmost site it is 3,849 square miles.

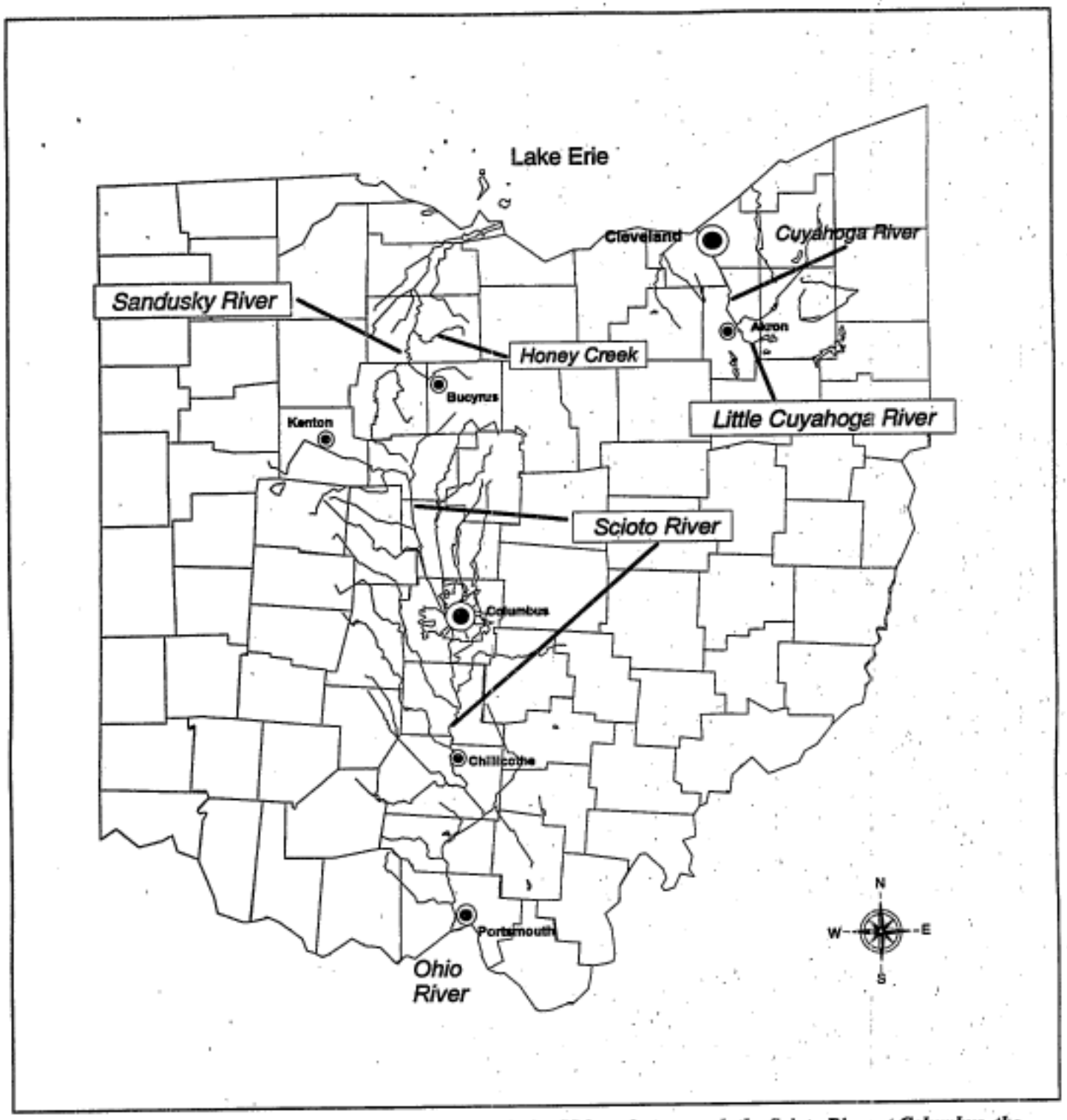

Figure 3-1. State of Ohio; three river systems within which the CSO study occurred: the Scioto River at Columbus, the Sandusky River at Bucyrus, and the Little Cuyahoga River at Akron. Honey Creek serves as a regional reference stream for the Sandusky River.

Table 3-1 Biological sampling stations in Ohio. Sampled 8-9, 24 September 1992.

STATION LABEL	STATION TYPE	RIVER MILE	LOCATION OF SAMPLING STATION
S1	Upstream Reference	136.4 Scioto	200 meters downstream from the Fifth Avenue bridge in western Columbus
S2	CSO Impact	129.5 Scioto	200 meters downstream of the Greenlawn Avenue bridge in Columbus about 2 miles upstream of the Frank Road station (S3)
S3	CSO Impact	127.7 Scioto	50 meters downstream of Frank Road bridge (Hwy. 104) in Columbus
S4	Regional Reference	99.9 Scioto	200 meters upstream of Hwy. 22 bridge (20 miles south of Columbus at Circleville)
SA1	Upstream Reference	115.0 Sandusky	150 meters upstream of Hwy. 30 bridge at the Fish Hatchery in Bucyrus
SA2	CSO Impact	111.5 Sandusky	700 meters upstream of Bucyrus WWTP at the downstream edge of Aumiller Park (RM 114.4)
SA3	CSO Impact	111.1 Sandusky	50 meters upstream of Bucyrus WWTP
SA4	Regional Reference	12.5 Honey Creek	50 meters upstream of Hwy. 100 bridge (20 miles north of Bucyrus on Hwy. 100 at Melmore)
CR1	Upstream Reference	11.3 Little Cuyahoga	500 meters upstream of Gilchrist Road bridge in Mogadore
CR2	Upstream*	7.1 Little Cuyahoga	Immediately downstream of Massillon Rd. bridge in Akron
CR3	CSO Impact	0.3 Little Cuyahoga	At the Police Firing Range off Cuyahoga Street in Akron

*In selecting this sampling location, it was originally thought that it could be considered a CSO receiving station. According to Ohio EPA (J. DeShon, pers. comm.), the outfall(s) upstream of Massillon Road had been eliminated some time prior to our sampling; thus, impacts from CSOs were expected to be evident only at CR3.

Table 3-2 Scoring criteria developed for the benthic macroinvertebrate assemblage on each Ohio study river using USEPA's rapid bioassessment protocols. For a description of the development of scoring criteria, see Section 2.3.1.

ASSESSMENT SCORES	SCORING CRITERIA											
	0	2	4	6	0	2	4	6	0	2	4	6
METRIC	*SCIOTO RIVER*				*SANDUSKY RIVER*				*LITTLE CUYAHOGA RIVER*			
1. Taxa richness	0-5	6-10	11-15	≥16	0-6	7-13	14-20	≥ 21	0-4	5-8	9-13	≥14
2. HBI	≥5.3	5.2-3.9	3.8-2.5	≤2.4	≥5.1	5.0-3.7	3.6-2.3	≤2.2	≥6.3	6.2-4.6	4.5-2.9	≤2.8
3. Sc/(Sc+Fc) x 100	0-7.3	7.4-14.7	14.8-22.1	≥22.2	0-19.3	19.4-38.7	38.8-58.1	≥58.2	0-15	16-30	31-45	≥46
4. EPT/(EPT+Chir) x 100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100
5. % Contribution Dominant Taxon	100-76	75-51	50-26	≤25	100-76	75-51	50-26	≤25	100-76	75-51	50-26	≤25
6. EPT Index	0-3	4-6	7-9	≥10	0-3	4-6	7-9	≥10	0-3	4-6	7-9	≥10
7. Shredders/Tot x 100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100
8. H/T x 100	100-76	75-51	50-26	≤25	100-76	75-51	50-26	≤25	100-76	75-51	50-26	≤25
9. Pinkham-Pearson	0-1.8	1.9-3.7	3.8-5.6	≥5.7	0-1.6	1.7-3.3	3.4-5	≥5.1	0-1.2	1.3-2.5	2.6-3.8	≥3.9
10. QSI-Tax	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100
11. DIC-5	≤1	2	3	≥4	≤1	2	3	≥4	≤1	2	3	≥4
12. QSI-FFG	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100	0-25	26-50	51-75	76-100

3.2.2.1 *Historical Information*

The Scioto River mainstem downstream from Columbus has been monitored frequently over a distance of approximately 40 miles (Ohio EPA 1992) from 1974 to the present. The most current biological data from Ohio EPA are from macroinvertebrate surveys conducted in 1988 and 1991 and fish surveys conducted from 1985 to 1991. The 1988 macroinvertebrate results showed the most severe impacts in the CSO-impacted area (RM 132.3 to 129.8) of the Scioto River study area (Ohio EPA 1992) (Figure 3-3). Invertebrate community index (ICI) values were in the "poor" range and reflected the impact of CSO inputs combined with extremely low river flows due to withdrawals for drinking water and drought. The ICI is Ohio EPA's multiple metric approach for assessing the biological integrity of streams and rivers, and is based on benthic macroinvertebrate samples taken from artificial substrates (Ohio EPA 1987b). Ohio EPA believes that the CSO impacts, at least in part, extended downstream for a distance of 15 to 20 miles. In 1991, another low flow year, the ICI improved somewhat but remained in the lower "fair" range. The combined effects of upstream water withdrawals and drought, old channel modifications, urban runoff, and the input of organic matter and nutrients from CSOs account for the degradation.

The three most upstream sampling sites surveyed in the present study (1992) were also sampled in 1991. These stations were RM 136.3 (136.4 in the present study), RM 129.0 (129.5 in the present study), and RM 127.8 (127.7 in the present study). The farthest downstream station at RM 100 (99.9 in the present study) was sampled in 1989 and 1992. The ICI rated the upstream station (RM 136.3) and farthest downstream station (RM 100) as very good and exceptional, respectively, for macroinvertebrate assemblage condition. RM 129.0 was considered fair in 1991, and RM 127.8 was considered poor; both are in the downstream end of the area of CSO inputs.

3.2.2.2 *Sampling Station Descriptions and Habitat Quality Assessments*

The four sampling stations on the Scioto selected for this study are described in Figure 3-2 and Table 3-1; habitat assessment rating scores, along with measurements of dissolved oxygen, temperature, and conductivity, are presented in Table 3-3.

Scioto River below the Fifth Avenue bridge - Station S1 (upstream reference). Station S1 is a site-specific reference used by Ohio EPA for the Scioto. The river receives no CSO input at or above this location. This station has a large riffle area stretching across the channel; the substrate is composed primarily of gravel and cobble along with some small boulders. Channel stability here appears to be good from observations of bank form and the riparian vegetation. There was an abundance of leaf litter along with some green algae in the kick net samples. The habitat assessment rating score was 145 (Table 3-3).

Scioto River at Greenlawn Avenue - Station S2 (CSO impact). The gravel bars at this station did not support the colonization by grasses, and there was also a broader floodplain that lacked any undergrowth. The habitat score at this station was 131.

Scioto River at Frank Road (Hwy. 104) - Station S3 (CSO impact). This station seemed to have considerable stability in features of channel morphology such as banks and riparian vegetation. There were also relatively low levels of siltation and embeddedness and abundant growths of filamentous green algae growing on rocks. Several gravel bars had developed here in mid- and off-center sections of the channel and had become vegetated by grasses. At this station there was a strong sewage odor during sampling. All CSO input from the city of Columbus is present in the flow by this level (RM 127.7) in the river. This station received a habitat assessment score of 136.

Scioto River at Circleville - Station S4 (regional reference). This sampling station serves as the regional reference site for Ohio EPA. The river here exhibits active bank erosion, formation of large gravel bars, perhaps reflecting increased bedload, and relatively heavy sedimentation. There was also indication of recent out-of-bank flows on one side of the channel floodplain. Habitat rated 117 points at this station. Although the habitat score at station S4 might appear low, it is likely a "natural" artifact of the large river; i.e., the drainage area at this site is 3,200 square miles compared to 1,600 at station S2 and ~1,000 square miles at S1. This river size exceeds the usual applications of the habitat assessment approach used in this study; therefore some of the physical attributes of S4 might be unfairly penalized. Ohio EPA found that ICI metric scores are "lower" at higher drainage areas. Therefore, the habitat quality might be "natural" and comparisons between S1 and S4 might be inappropriate.The quality of the overall habitat structure was best at Station S1 (the upstream site-specific reference) and relatively similar at Stations S2 and S3 (the middle stations). The worst rated condition was observed at Station S4 (Table 3-2), which is considered to be an ecoregional reference station by Ohio EPA. However, the relative magnitude of similarity (i.e., percent comparability) in habitat quality was 80 percent between S1 and S4,which is not a significant difference. Individual components of the physical habitat structure at Station S4 that were rated as suboptimal or marginal were related to erosion and sedimentation (producing embeddedness and siltation of the substrate) and to alteration in channel morphology and bank structure. These problems in habitat structure are most likely a result of the agricultural land use

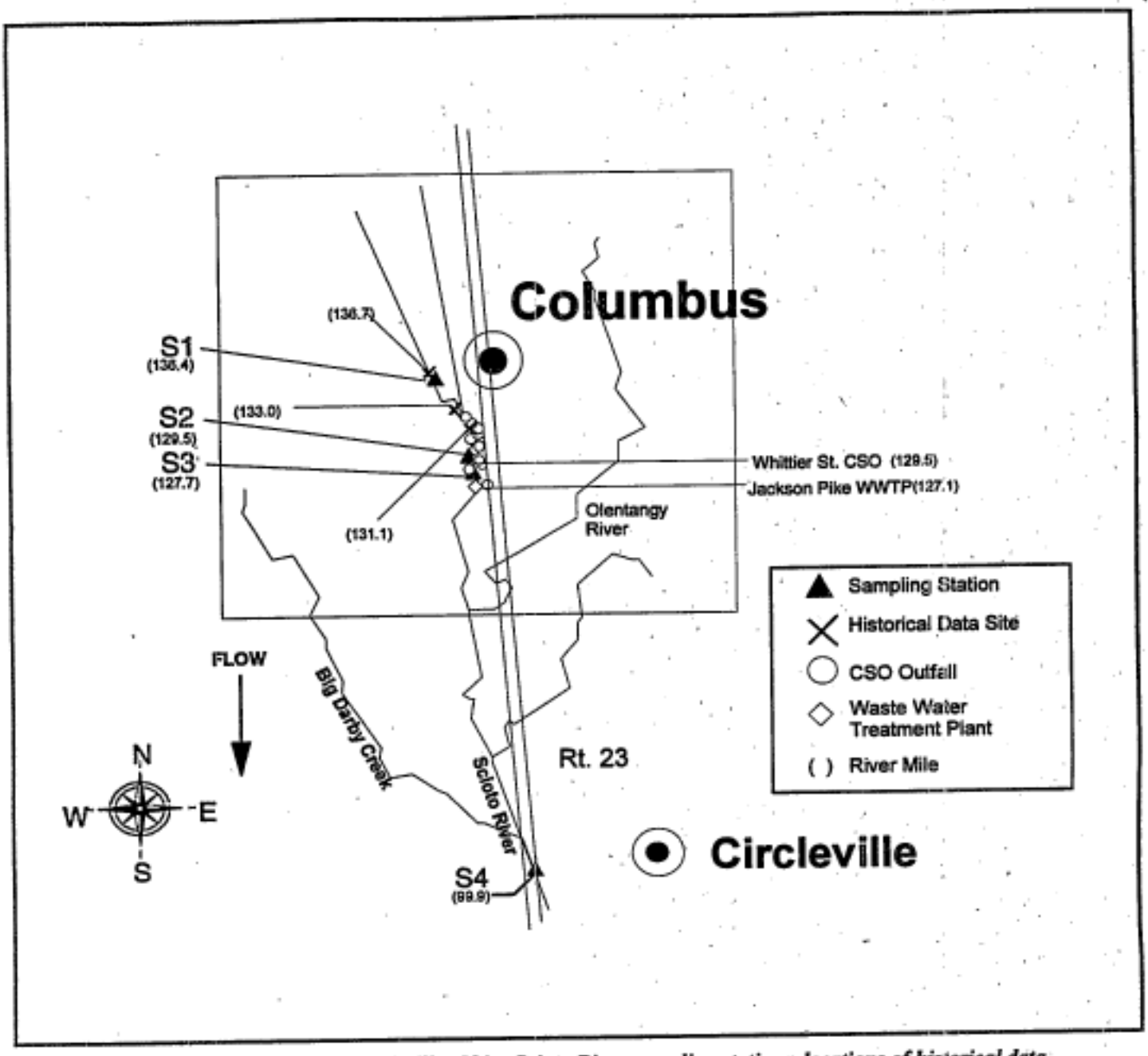

Figure 3-2. Cities of Columbus and Circleville, Ohio; Scioto River sampling stations, locations of historical data collection, CSO outfalls, WWTP, and river mile designations (approximate scale 1 inch = 8.2 miles).

patterns in this area of the Scioto River, located south of Circleville. It is also an area characterized by glacial outwash (C. Yoder, Ohio EPA, January 1993, pers. comm.), a geological condition that contributes to the "degraded" appearance of many large river channels. Stations S2 and S3 are apparently subjected to substantial bedload movement along with dense growths of filamentous algae. Gravel bars were present at Station S3, but were vegetated with grasses, indicating that increases in flow were not frequent enough to flood or destabilize the bars. However, the broader floodplain at Station S2 was not vegetated, which indicated frequent flooding.

In spite of the sedimentation and bedload at the middle stations, habitat should not be limiting to development of the biological community. Differences in biological condition among Stations S1, S2, and S3 may, therefore, be assessed in the context of differences in water quality. Habitat quality might be limiting at Station S4 compared to the site-specific reference. However, Station S4 is consid-

Table 3-3 Habitat assessments and physicochemical measurements of the Scioto River taken on 8 September 1992. For a description of the stations, see Table 3-2 and Section 3.2.2.2.

HABITAT PARAMETERS		SCORES			
		SCIOTO RIVER SAMPLING STATIONS			
		S1	S2	S3	S4
Primary					
Substrate Instream Cover Flow Canopy (0-20)	Bottom Substrate/Instream Cover	18	16	15	17
	Embeddedness	15	14	16	11
	Flow or Velocity/Depth	18	16	16	16
	Canopy Cover (Shading)	10	10	14	8
Secondary					
Channel-Morphology (0-15)	Channel Alteration	13	11	13	9
	Bottom Scouring and Deposition	13	12	11	9
	Pool/Riffle, Run/Bend Ratio	14	11	8	8
	Lower Bank Channel Capacity	11	8	10	8
Tertiary					
Riparian and Bank Structure (0-10)	Bank Stability	9	8	9	6
	Bank Vegetative Stability (Grazing)	8	8	8	7
	Streamside Cover	8	8	8	8
	Riparian Vegetative Zone Width	8	9	8	10
TOTAL SCORE		145	131	136	117
Physicochemical Parameters	Dissolved Oxygen (mg/L)	6.7	8.9	8.9	8.4
	Temperature (C)	23.5	24.9	24.8	23
	Conductivity (μMhs)	600	590	600	750

ered to be an appropriate ecoregional reference by Ohio EPA; therefore, the biological condition is expected to be of a reasonably high quality.

3.2.2.3 Biological Assessments

Even though habitat quality was rated lower at the ecoregional reference station at Circleville (S4) due to the river size and the habitat parameters used (Figure 3-4), biological metrics indicated good conditions (Table 3-4). The upstream station (S1) scored only 79 percent of the ecoregional reference, which indicated that the benthic assemblage was slightly impaired before exposure to the CSO discharge. There is an increased abundance of midges at the two middle stations (S2 and S3), resulting in low values of the EPT/Chironomidae ratio (metric 4), a result often seen in stressed situations. Also, lower calculated values of the scraper/filterer collector ratio (metric 3), seen in these same two stations, indicate increased suspended organic particulates in the flow, perhaps resulting from organic enrichment.

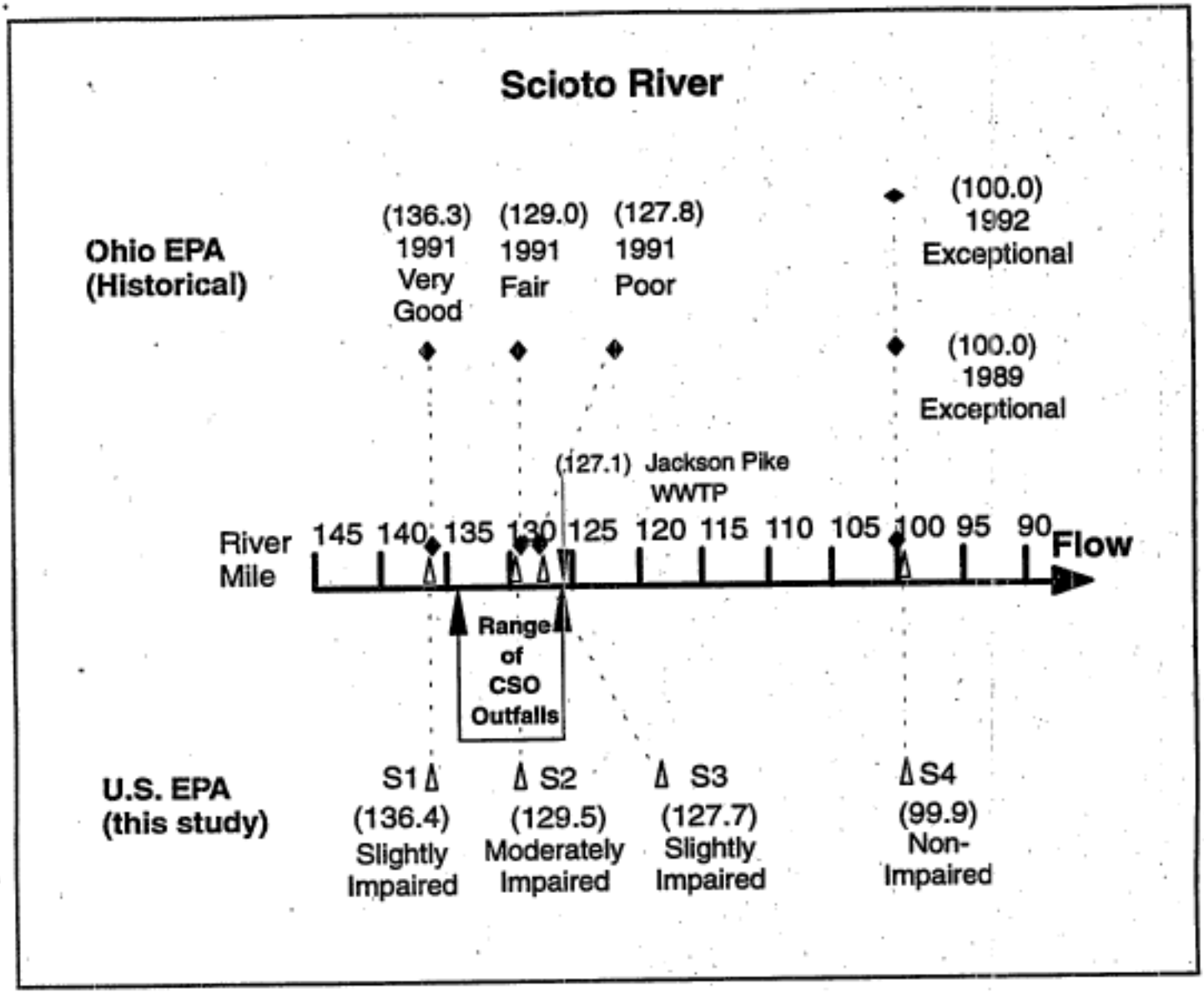

Figure 3-3. Linear comparison with Ohio EPA assessments on the Scioto River.

Station S2, located approximately 4.5 miles downstream of initial CSO outfalls and exactly at the location of the Whittier Street CSO outfall (RM 129.5), received a bioassessment score 50 percent of the reference, indicating moderate impairment. Downstream 2 miles, Station S3 had a bioassess-ment rating that indicated slight impairment (69 percent of reference) and some recovery from the conditions at S2.

3.2.2.4 Comparison to Historical Assessments

The Scioto River has the most extensive history of biological monitoring and assessment of the three CSO sites under investigation in this project (Ohio EPA 1986). The results from Ohio EPA seem to be comparable to those of the present study in which Stations S1 (RM 136.4) and S4 (RM 99.9) were found to be of the best biological quality (Figure 3-3). Stations S2 (RM 129.5) and S3 (RM 127.7) were found to be moderately and slightly impaired. Ohio EPA found its two nearest stations, RM 129.0 and 127.8, to be fair and poor, respectively. The largest discrepancy in the results between the present study and the 1991 Ohio EPA study was between the farthest downstream station within the zone of CSO outfalls, RMs 127.8 (Ohio EPA) and 127.7 (present). The former was found in 1991 to be in "poor" condition by the ICI and in "slightly impaired" condition by the RBPs. This difference may be a sign of improvement in water quality during the time between the two sampling events. However, an alternative explanation is that the differences in the macroinvertebrate communities were due to the differences in flow between 1991 (a very dry year) and 1992 (a very wet year). Ohio EPA data (Ohio EPA 1992) suggest that more severe degradation in areas of

CSO releases are experienced in the dry years. This may be due to the material deposited by CSOs in previous years which may exert strong effects on biologoical factors such as 0_2 demand. If a community is already stressed from low flow, changes in 0_2 demand would more quickly cause an impact on it.

3.2.3 The Sandusky River at Bucyrus, Ohio

The Sandusky River is a major tributary to Lake Erie, its drainage area occupying 1,420 square miles of predominantly agricultural land in north-central Ohio (Figure 3-1). It flows east to west from its headwaters to Upper Sandusky, where it turns north and discharges into Sandusky Bay, the largest embayment on the southern shore of Lake Erie. The major urban areas in the basin include Fremont, Tiffin, Upper Sandusky, and Bucyrus.

Within the study area, the Sandusky River is predominantly unmodified and free-flowing. Minor channel modifications have occurred at RM 110.8 downstream from the Bucyrus WWTP. The majority of the Sandusky River is predominated by bottom substrates of cobble, gravel, and boulders.

3.2.3.1 Historical Information

A survey of the Sandusky in 1980 revealed significant impacts by CSOs, particularly downstream of Bucyrus. A study done in 1990 compared assessments after modifications were made to the Bucyrus WWTP (Ohio EPA 1991) with results from 1980. Trend assessment data showed that there was a general improvement in fecal coliform bacteria since 1979, though high counts still occurred downstream of CSO outfalls (Ohio EPA 1991). The WWTP was upgraded in 1988 and was successful in reducing, but not eliminating, CSO loadings. An improvement in the condition of the benthic macroinvertebrate assemblage downstream of the WWTP outfall (comparing 1990 samples to 1979 samples) reflects this plant upgrade. CSOs within Bucyrus were identified in 1979 as a significant source of organic degradation; moderate impacts to the invertebrate assemblage continued as recently as 1990.

Upstream of Bucyrus, a marginal decline in the condition of the fish assemblage was detected in 1990 as compared to that of 1979. As of 1990, the fish assemblage had shifted to more tolerant species, resulting in nonattainment of the state biocriteria for this river. Downstream of the WWTP, slight improvement in the fish assemblage was detected between 1979 and 1990. As with the macroinvertebrates, this increase in biological condition can be partially attributed to the WWTP upgrade in 1988. Additional improvement in the fish assemblage is expected since it is not unusual for recovery in fish populations to lag behind improvements in water chemistry and macroinvertebrate community structure (C.O. Yoder, personal communication).

3.2.3.2 Sampling Station Descriptions and Habitat Quality Assessments

Four sampling stations on the Sandusky River were selected for this study of Bucyrus CSOs (Table 3-1); habitat assessment rating scores, along with measurements of dissolved oxygen, temperature, and conductivity, are presented in Table 3-5.

Sandusky River upstream of Hwy. 30 bridge - Station SA1 (upstream reference). No habitat problems are evident at this station. The riparian zone is in an undisturbed condition, and there was little obvious sedimentation occurring. However, the riffle from which the samples were taken appeared as if it had been constructed, perhaps in an effort to enhance fish habitat with larger and deeper pools upstream and downstream. The riffle was composed of various-sized boulders, some very large. The upstream and downstream pools were too deep to wade in, and it appeared that the rocks had been removed from them for placement in the riffle. There was no indication of how long the riffle had been in place to allow for colonization. Nonetheless, habitat quality was unquestionably in the best condition of the Sandusky sampling stations, as it received an RBP habitat score of 153 (Table 3-5, Figure 3-4).

Sandusky River at Aumiller Park - SA2 (CSO impact). This station is located approximately 700 meters upstream of the Bucyrus WWTP at the downstream edge of Aumiller Park. Ohio EPA has indicated that the majority of CSO input is at this park. Here the river is experiencing severe physical disruptions apparently unrelated to CSOs. Heavy sedimentation is occurring due to the activity of heavy machinery approximately 150 meters upstream and bank failure at the station. Habitat quality ratings were in the marginal or poor category for embeddedness, all of the channel morphology parameters, and riparian vegetative buffer zone width. This station received a habitat quality rating score of 81.

Sandusky River downstream of Bucyrus, upstream of WWTP - SA3 (CSO impact). Station SA3 is located approximately 50 meters upstream of the Bucyrus WWTP and is downstream of most CSO outfalls. The station could not be located farther downstream of the CSOs due to the WWTP. According to Ohio EPA, there are numerous outfalls along the 700- to 750-meter stretch of the river between Aumiller Park and the WWTP. The river here rated suboptimal and marginal for embeddedness, width of riparian zone, bottom scouring, and deposition, and it had a low pool/riffle, run/bend ratio. (Throughout the entire reach of the river walked, approximately 750 meters, only three riffle areas were found.) One bank is part of an old landfill and is composed of soil completely interspersed

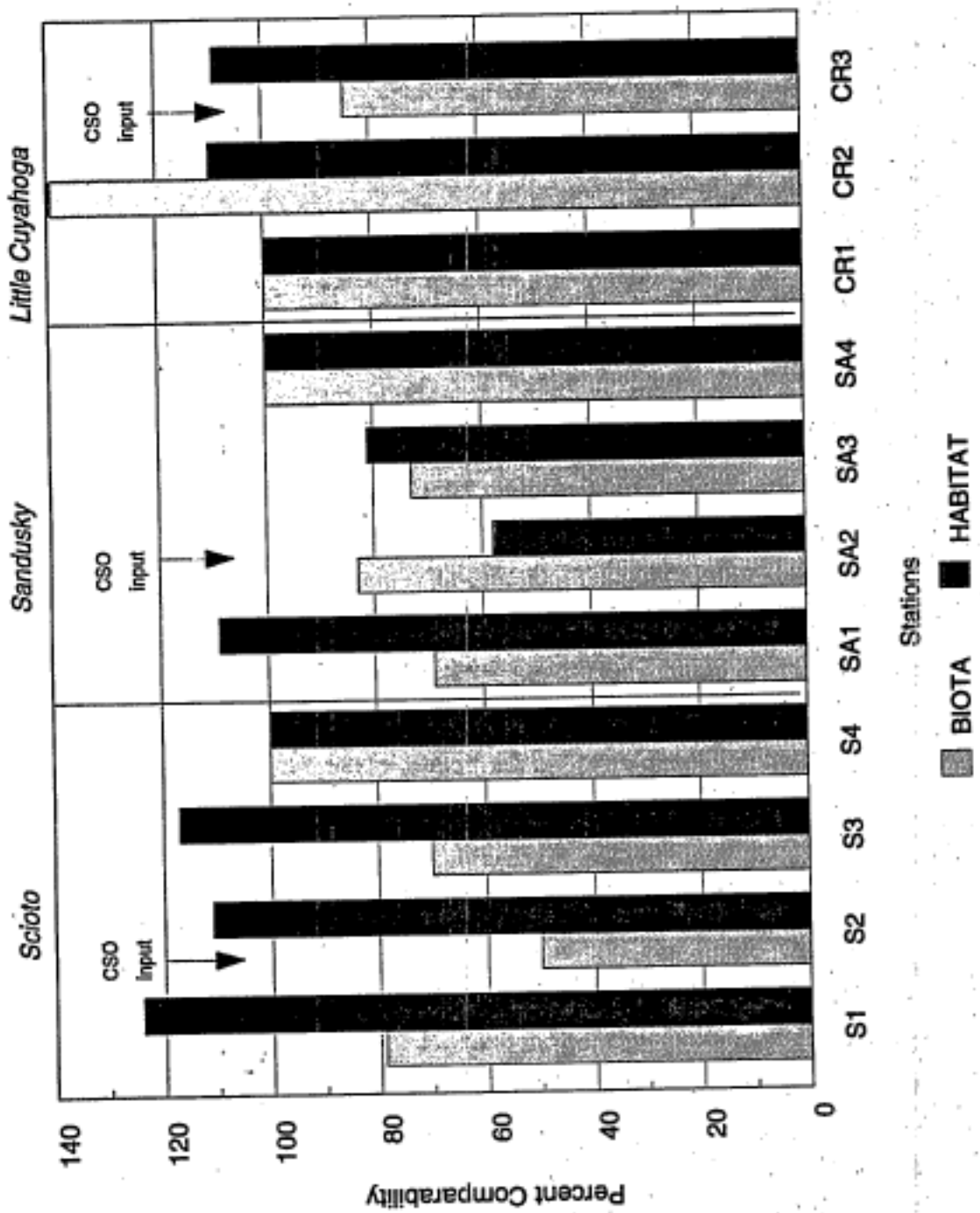

Figure 3-4. Percent comparability to reference sites for Ohio sampling stations.

Table 3-4 Calculated biological metrics for Scioto River, Sandusky River, and Little Cuyahoga River, Ohio; September 1992[a].

METRIC	S1	S2	S3	S4	SA1	SA2	SA3	SA4	CR1	CR2	CR3
1. Taxa Richness	18 (6)	19 (6)	22 (6)	20 (6)	18 (4)	22 (6)	21 (6)	21 (6)	14 (6)	16 (6)	11 (4)
2. HBI	5.1 (2)	4.7 (2)	5.3 (0)	4.9 (2)	4.9 (2)	5.1 (2)	5 (2)	4.4 (2)	5.2 (2)	6.3 (0)	5.9 (2)
3. Scr/(Scr + Fc) x 100	24 (6)	16 (4)	18 (4)	29 (6)	12 (0)	25 (2)	77 (6)	54 (4)	1.5 (0)	60 (6)	0 (0)
4. EPT/(EPT + Chir) x 100	83 (6)	42 (2)	54 (4)	87 (6)	97 (6)	92 (6)	80 (6)	98 (6)	98 (6)	43 (2)	28 (2)
5. % Contr. Dominant Taxon	28.4 (4)	28.2 (4)	29.9 (4)	17 (6)	57 (4)	37 (4)	27 (4)	23 (6)	78 (0)	18 (6)	35 (4)
6. EPT Index	8 (4)	6 (2)	12 (6)	12 (6)	7 (4)	10 (6)	8 (4)	11 (6)	2 (0)	3 (0)	2 (0)
7.CPOM: (Shredders/total) x 100	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Kick: (Shredders/total) x 100	0.003 (0)	0 (0)	0 (0)	0 (0)	0.2 (0)	0.5 (0)	1.3 (0)	0 (0)	0 (0)	0.03 (0)	0 (0)
8. (Hydropsychidae/total Trich) x 100	99 (0)	99 (0)	99 (0)	95 (0)	25 (6)	39 (4)	68 (2)	22 (6)	100 (0)	100 (0)	0 (0)[b]
9. Pinkham-Pearson	5.6 (4)	4.6 (4)	7.1 (6)	RR (6)	4.7 (4)	6.4 (6)	3.5 (4)	RR (6)	UR (6)	4.8 (6)	3.6 (4)
10. QSI-Taxa (%)	48 (2)	6.5 (0)	41.1 (2)	RR (6)	45.5 (2)	57 (4)	41.4 (2)	RR (6)	UR (6)	6.3 (0)	4.1 (0)
11. DIC - 5	3 (4)	1(0)	1 (0)	RR (6)	3 (4)	4 (6)	1 (2)	RR (6)	UR (6)	2 (2)	2 (2)
12. QSI-FFG (%)	82.5 (6)	73.7 (4)	83 (6)	RR (6)	53.8 (4)	71.1 (4)	79.3 (6)	RR (6)	UR (6)	14.3 (0)	22.9 (0)
Total bioassessment scores with paired metrics	44	28	38	56	40	50	44	60	38	30	18
Biology (with paired) % comparison to reference	79	50	69	----	67	83	73	----	----	95	47
Total bioassessment scores without paired metrics	----	----	----	----	----	----	----	----	14	22	12
Biology (without paired) % comparison to reference	----	----	----	----	----	----	----	----	----	157	86
Habitat scores	145	131	136	117	153	81	116	143	107	116	115
Habitat - % comparison to reference	124	112	116	---	107	57	81	----	----	108	107

For station and metric descriptions, see Table 3-2 and Section 2.3.2. Bioassessment scores (in parentheses) are derived by comparing metric values to scoring criteria (Table 3-1). (Paired metrics [9-12] used data from the regional reference (RR) station for calculations.)

[b] no Trichoptera collected.

Table 3-5 Habitat assessments and physicochemical measurements of the Sandusky River taken on 9 September 1992. For a description of the stations, see Table 3-2 and Section 3.2.3.2.

HABITAT PARAMETERS		SCORES			
		SANDUSKY RIVER SAMPLING STATIONS			
		SA1	SA2	SA3	SA4
Primary					
Substrate Instream Cover Flow Canopy (0-20)	Bottom Substrate/Instream Cover	17	10	10	18
	Embeddedness	16	5	8	17
	Flow or Velocity/Depth	19	16	16	18
	Canopy Cover (Shading)	18	16	18	10
Secondary					
Channel-Morphology (0-15)	Channel Alteration	12	3	13	13
	Bottom Scouring and Deposition	11	2	7	12
	Pool/Riffle, Run/Bend Ratio	14	4	7	13
	Lower Bank Channel Capacity	12	8	11	11
Tertiary					
Riparian and Bank Structure (0-10)	Bank Stability	9	5	8	9
	Bank Vegetative Stability (Grazing)	8	6	8	9
	Streamside Cover	8	5	8	8
	Riparian Vegetative Zone Width	9	1	2	5
TOTAL SCORE		153	81	116	143
Physicochemical Parameters	Dissolved Oxygen (mg/L)	8.6	7.7	6.9	9.5
	Temperature (C)	19	20	16	21
	Conductivity (μMhs)	750	700	650	450

with broken glass and rusted pieces of metal. However, enough soil is present to have been colonized by some woody and herbaceous vegetation. This station rated 116.

Honey Creek at Melmore (Hwy. 100) - SA4 (regional reference). This station is an Ohio EPA regional reference site. Even though the weather conditions were sunny and warm, the water level seemed to be up and, in fact, slightly rising while on-site. The water also appeared somewhat turbid. There might have been some rainfall upstream in the watershed causing these conditions. Aspects of the habitat that rated in the suboptimal or marginal ranges were related to channel capacity and the vegetated buffer zone. Water appeared to have intermittently escaped the channel on the side with a low bank. Also, the width of the riparian vegetative zone is reduced on one side by agricultural fields and on the other by mowing. In particular, the zone on the mowed side had a buffer zone of woody vegetation only approximately 3 to 6 meters wide. The habitat assessment rating score was 143.

The condition of the instream habitat and channel morphology at the Aumiller Park station (SA2) is indicative of considerable physical degradation. It might prove to be

limiting to the development of the benthic macroinvertebrate assemblage. Station SA3, just upstream of the WWTP and the downstream-most station on this river, has substantial riparian degradation and embeddedness with some evidence of scour, but it should provide habitat that will allow development of the benthic assemblage to a level comparable to that of the reference conditions. The best habitat encountered on the Sandusky was at the Fish Hatchery station (SA1), the Ohio EPA upstream reference station; the regional reference station habitat scored slightly less than SA1 but was comparable.

3.2.3.3 Biological Assessments

In the Sandusky River system, the regional reference (Figure 3-5; Honey Creek at Melmore, SA4) produced a total bioassessment score of 60. Station SA2, the upstream CSO-impact station at Aumiller Park, was most comparable to the regional reference at 83 percent comparability for biology (Figure 3-4), indicating nonimpairment; the slight reduction in biological condition was likely due to problems in habitat quality at this station. The downstream impact station, SA3, was slightly impaired, producing a habitat assessment score 73 percent comparable to the regional reference (Table 3-4). The bioassessment score least comparable to the regional reference was 67 percent at Station SA1, the upstream reference; this could be due to the habitat at SA1 being somewhat different with an apparently human-constructed riffle. Though this station was rated higher in habitat quality, the substrate composition might have had an effect on comparisons with the downstream stations, the substrate of which was primarily embedded cobble and gravel. Overall, the slight decrease in biological condition from SA2 to SA3 is attributed to additional CSOs and urban runoff, which further impaired the biological community in an area of increased habitat quality. These findings concur with the 1990 Ohio EPA survey of the Sandusky River (Ohio EPA 1991).

3.2.3.4 Comparison to Historical Assessments

The most recent Ohio EPA macroinvertebrate sampling on the Sandusky River, in 1990, categorized the macroinvertebrate assemblages at RMs 115.0 and 111.4 as "exceptional" and RM 111.1 as "marginally good" (Ohio EPA 1991) (Figure 3-6). The current study shows station SA1 (RM 115.0), the farthest upstream station, to be slightly impaired at 67 percent comparability to the regional reference station at Honey Creek (SA4) due to an apparent habitat alteration. Differences between the current study and that of Ohio EPA (Figure 3-6) might be attributed to gear differences (artificial substrate samplers by Ohio EPA and instream substrate in the current study). It is likely that sampling the bottom substrate directly with the kick net is demonstrating the difference in the habitat quality (substrate) at the two different stations, whereas use of artificial substrate samplers might have masked that difference by providing suitable "habitat" for colonization. Therefore, effects on the biological community observed when using artificial substrate might better reflect pure water quality differences.

Another factor could be the use of Honey Creek as a site-specific reference in the current study; Ohio EPA uses Honey Creek as one of the 133 reference streams that make up its reference condition for this class of stream. As stated earlier, the use of multiple reference sites (or reference conditions) are preferable to single reference sites. It should also be noted that the habitat disturbance at station SA1 noted in 1992 might have occurred after the 1990 sampling was conducted, but it was not possible to be certain. RBP samples were taken at RMs 111.5 and 111.1 (SA2 and SA3, respectively), bracketing the station found to be "exceptional" by Ohio EPA (1991). Comparability to the regional reference at SA2 was at 83 percent or "nonimpaired"; SA3 was 73 percent or "slightly impaired." Even with habitat problems at SA2 (RM 111.5), there was little indication of biological impairment compared to the regional reference.

At the downstream station (SA3, RM 111.1), there was slightly less habitat degradation in the form of scour and embeddedness but a further decrease in biological condition. Habitat problems at SA3 compared to SA2 were not as severe as those seen at SA2. Therefore, the slight biological impairment noted at SA3 can be attributed to influence from additional CSOs and urban runoff rather than habitat.

This assessment of slightly impaired biological condition at SA3 (RM 111.1) is similar to the Ohio EPA 1990 assessment (marginally good), which was also attributed to CSO inputs. These results seem to be compatible with those included in the most recent historical assessment reports (Ohio EPA 1991) (Figure 3-6). Additionally, SA2 might have experienced organic or fertilizer loading that caused a positive response of the benthic community (nonimpaired assessment). The initial phase of nutrient loading (organic enrichment) can mask the effects of habitat degradation by elevating the biological community (plants and animals). As organic enrichment increases, however, the bloom in the biological community begins to have adverse effects on the waterbody. For instance, algal blooms cause reduced light penetration below the water's surface and the bottom-dwelling plants die. As the abundant plant material decays, oxygen is used up rapidly, which causes further stress, and eventual more severe impairment of the biological community. Thus, while organic enrichment in the initial phase has a positive effect on the biology, it cannot be sustained over a longer periods of time.

3.2.4 The Little Cuyahoga River at Akron, Ohio

The Little Cuyahoga River flows through Akron in northeastern Ohio. The study area begins just downstream of Mogadore

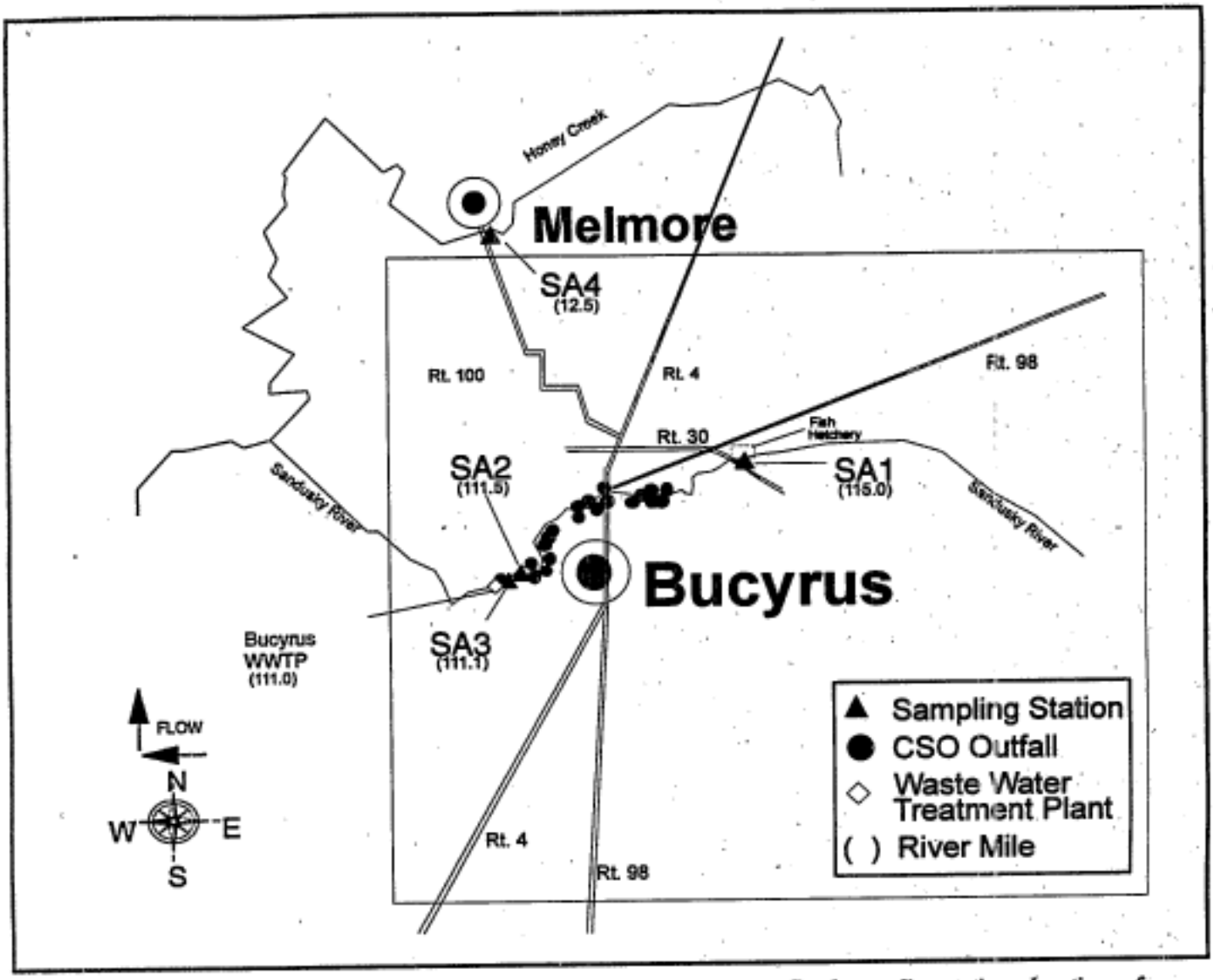

Figure 3-5. Cities of Bucyrus and Melmore, Ohio; Sandusky River and Honey Creek sampling stations, locations of historical data collections, CSO outfalls, WWTP, and river mile designations (approximate scale 1 inch = 6.0 miles).

Reservoir. Of the three stations sampled, the two downstream stations (CR2 and CR3) were expected to be receiving CSO input. It was later discovered that the outfalls upstream of the middle station had been eliminated in the past 5 years, leaving only the lower station to provide biological data expected to reflect response to pollutant input. This situation might allow the middle station to yield information on biological recovery following removal of CSO outfalls.

3.2.4.1 Historical Information

A benthic survey was conducted in 1986 on the Little Cuyahoga River. The ICI results indicated a combination of urban runoff and enrichment problems from lake and wetland drainage. These impacts resulted in a fair to poor ICI rating for most of the river between RMs 9.6 and 1.8.

The three sampling stations in the present study were also sampled in 1986: RMs 11.2 (RM 11.3 in present study), 7.1, and 0.3. However, of these three stations, only RM 0.3 was sampled in 1991 by Ohio EPA. In 1991, at RM 0.3 (upstream of the confluence with the Cuyahoga), the ICI reached the "fair" range and was essentially unchanged from 1986 (Ohio EPA 1994). In 1986, however, the condition of macroinvertebrate assemblage at RM 0.3 was lower than sites well upstream. The poor conditions were characterized by reductions in taxa richness, mayfly and caddisfly richness and abundance, and sharp increases in the percentage of tolerant invertebrate populations. These results were attributed to CSOs, urban runoff, and industrial point sources in Akron. Only a slight improvement (from poor to fair) was noted in 1986 at RM 0.3 when compared to the next upstream site at RM 3.8.

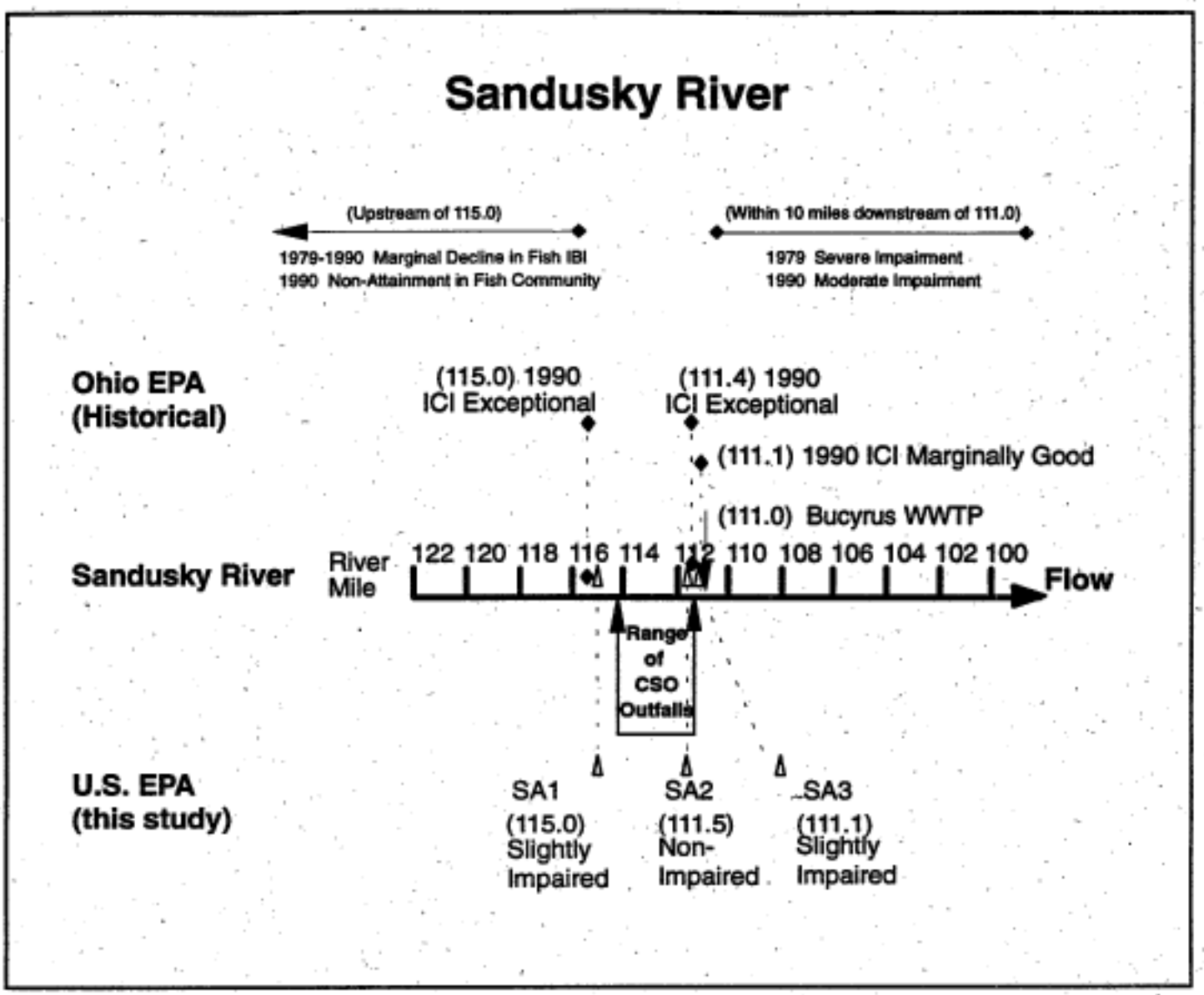

Figure 3-6. Linear comparison with Ohio EPA assessments on the Sandusky River.

3.2.4.2 Sampling Station Description and Habitat Quality Assessments

The three sampling stations on the Little Cuyahoga River selected for this study are presented in Table 3-1 and Figure 3-7. On visiting the regional reference stream used by Ohio EPA for the Little Cuyahoga (Breakneck Creek at Kent), it was found to be flooded out of its banks. Sampling could not be completed; therefore, the upstream reference station was used for comparison. Habitat assessment rating scores are provided in Table 3-6.

Little Cuyahoga River at Mogadore, Ohio - Station CR1 (upstream reference). This station is located approximately 2 miles downstream of releases from the dam of Mogadore Reservoir, well within the range within which physical channel alterations have been observed as a result of dam operations (Gordon et al. 1992; Rochester et al. 1984). However, this location was about 0.3 mile upstream from the station recommended by Ohio EPA as the reference station, which was inaccessible due to high flows. The station sampled contained no riffles; therefore, the samples were taken from runs. There was minimal variability of depths in the channel, a very strong flow, and substrate particles of mostly large cobble and small boulders with considerable embeddedness due to sand deposition. The sand was apparently coming from a sand and gravel pit upstream several hundred meters on one side of the stream channel. Station CR1 received marginal or poor scores on scouring/deposition; pool/riffle, run/bend ratio; and those parameters related to the riparian zone. This degradation is consistent with that expected downstream of dams

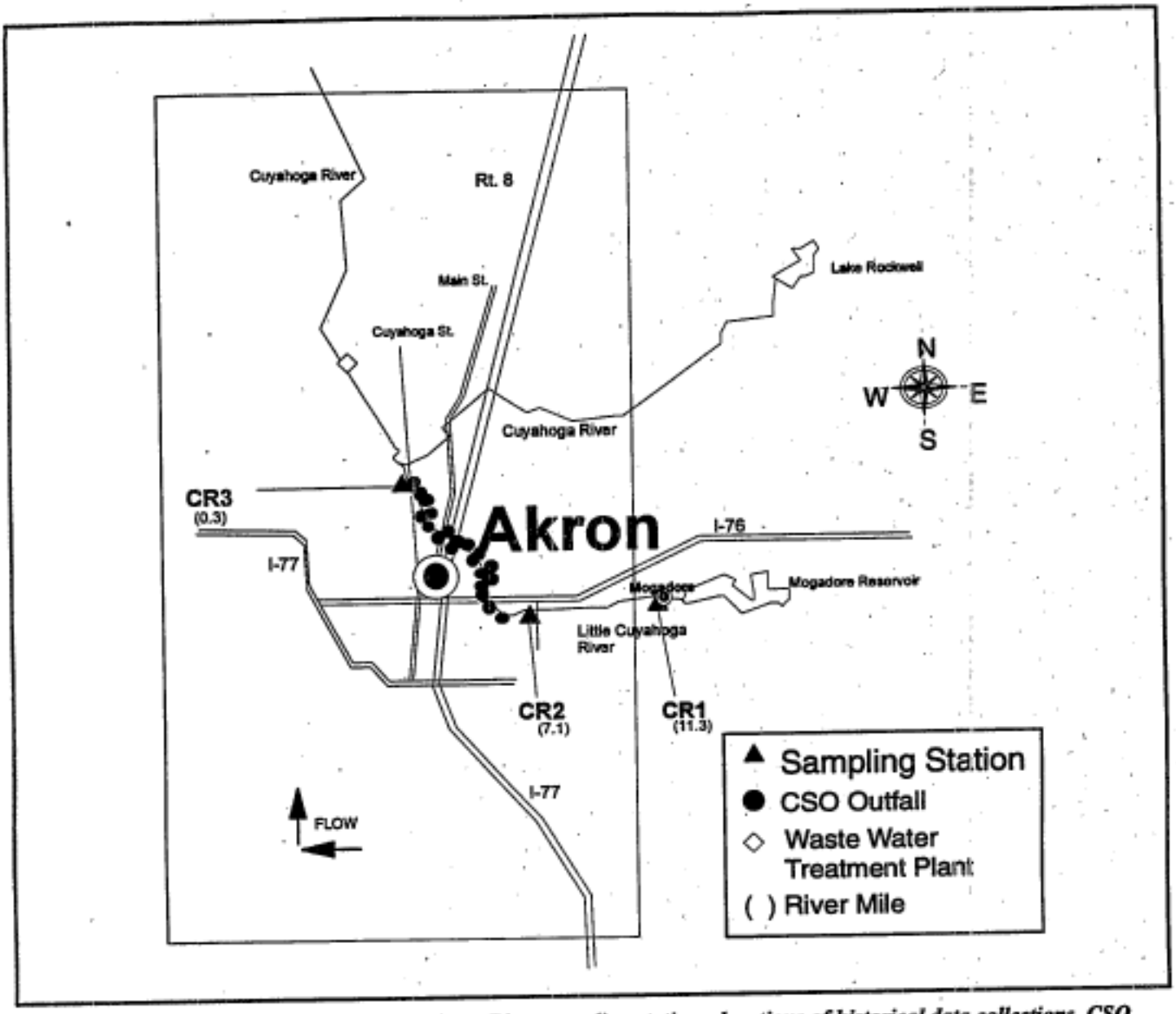

Figure 3-7. City of Akron, Ohio; Little Cuyahoga River sampling stations, locations of historical data collections, CSO outfalls, WWTP, and river mile designations (approximate scale 1 inch = 4.5 miles).

(Rochester et al. 1984). The overall habitat assessment score for CR1 was 107 (Table 3-6).

Little Cuyahoga River at Massillon Road bridge (State Rte. 241) - Station CR2 (upstream). Station CR2 is located in a heavily urbanized area of Akron (commercial/industrial/transportation). Components of habitat structure that were rated as suboptimal to poor included parameters related to a reduction in riparian vegetation and lack of variability in bottom contours, though some deep pools were present and there was diversity of substrate particle size. Riffles were at a minimum and samples were taken from runs. There was a stability of bank structure normally unexpected in such heavily urbanized areas. Habitat received an assessment score of 116, comparable to that of the reference station.

Little Cuyahoga River at the Police Firing Range off Cuyahoga Street - Station CR3 (CSO impact). The Little Cuyahoga River at Station CR3 experienced some sedimentation reflected in the rating scores for embeddedness, and scour and deposition. At this level the river is a straight channel without much variability in bottom contours, and substrate particle sizes were limited mostly to sand with some cobble and gravel. As at CR2, there were no true riffles; samples were taken from run areas. The station scored 115 on the assessment of habitat quality and was considered comparable to the reference station.

Table 3-6 Habitat assessments and physicochemical measurements of the Little Cuyahoga River taken on 24 September 1992. For a description of the stations, see Table 3-2 and Section 3.2.4.2.

HABITAT PARAMETERS		SCORE		
		LITTLE CUYAHOGA RIVER SAMPLING STATIONS		
		CR1	CR2	CR3
Primary				
Substrate Instream Cover Flow Canopy (0-20)	Bottom Substrate/Instream Cover	14	17	14
	Embeddedness	11	15	15
	Flow or Velocity/Depth	11	15	10
	Canopy Cover (Shading)	8	11	10
Secondary				
Channel-Morphology (0-15)	Channel Alteration	14	14	14
	Bottom Scouring and Deposition	7	11	8
	Pool/Riffle, Run/Bend Ratio	9	10	4
	Lower Bank Channel Capacity	11	7	13
Tertiary				
Riparian and Bank Structure (0-10)	Bank Stability	9	9	8
	Bank Vegetative Stability (Grazing)	5	2	6
	Streamside Cover	6	4	8
	Riparian Vegetative Zone Width	2	1	5
TOTAL SCORE		107	116	115
Physicochemical Parameters	Dissolved Oxygen (mg/L)	8	7.9	4
	Temperature (C)	17	15	2
	Conductivity (μMhs)	320	320	400

Overall, the Little Cuyahoga River, in the reaches of this study, has had considerable habitat degradation mostly from sedimentation and alteration of the riparian zone. However, the components of habitat quality that exhibited degradation were relatively consistent throughout the study area, and the resulting habitat scores were comparable at all three stations (Table 3-6). Thus, direct comparisons of the biological data among these stations should be possible and any observed differences can be interpreted to be the result of water quality problems.

3.2.4.3 Biological Assessments

Examination of metric values for the upstream reference station CR1 revealed a degraded biological condition. An increase in filterer collectors resulted in a low scraper to scraper + filterer ratio indicating potential organic pollution problems. The percent contribution of dominant taxon (78 percent Hydrosphychids), indicate poor community balance and account for the increase in filterers. Therefore, with no regional reference for comparison, this site (CR1) was not given a rating. The percent comparisons to reference (CR1) for stations CR2 and CR3 were made using metric totals without paired metrics; each assessment category was interpreted as one category less than those listed in Plafkin (1989) since the comparison was made using an impaired reference site.

The condition of the benthic community at station CR2 was considerably better than either the upstream or downstream

stations (CR1 and CR3, respectively). Although many taxa at this station were relatively tolerant, the taxa richness was the highest among the three stations and the percent contribution of dominant taxon was low.

A slight difference in condition of the benthic community was detected at the downstream station (CR3), which was 86 percent comparable to the upstream reference (Figure 3-4, Table 3-4). Because the habitat assessment was within the same range as that at CR1, the difference should be attributable to water quality. Specifically, there was a distinct depression in biological condition at CR3 (as exhibited by the metrics taxa richness, EPT-Chironomidae ratio, Pinkham-Pearson Community Similarity Index, DIC-5, and QSI-taxa), indicating the potential presence of toxicants from the CSO input. Abundance of invertebrates at both the middle and downstream stations was unexpectedly low (Appendix A): at CR3 a total of only 60 specimens were collected; at CR2, 133 specimens were in the total sample. At CR2 and CR3, a complete removal of organisms was required from the double-composite kick net samples in contrast to CR1, where a 300-organism subsample was taken. CR2 is considered to have a slightly impaired biology; CR3 is considered severely impaired.

3.2.4.4 Comparison to Historical Assessments

There are considerable habitat and discharge problems upstream of RM 11.0 along the Little Cuyahoga River (C. Yoder, pers. comm.). During low flow years, DO problems lead to decreased ICI values and thus lower bioassessment ratings. Ohio EPA found the upstream station of the Little Cuyahoga River (RM 11.2) to be in "fair" condition in assessments in 1986.

The upstream site assessment for the current study, (CR1 at RM 11.3) could not be rated due to evidence of biological impairment at the site and the lack of an accessible regional reference site to sample for baseline comparisons. Comparison to a degraded reference site falsely elevates the test site assessments. Thus, due to the degraded biological condition at CR1, the upstream reference site, assessments for CR2 and CR3 were lowered by one category.

Just above RM 11.0, a tributary from a natural and relatively undisturbed lake (Wingfoot Lake) enters the Little Cuyahoga River. This tributary entering above RM 11.0 is at least as large as the Little Cuyahoga upstream. This flush of clean water likely accounts for the Ohio EPA ratings of "good" and "very good" at the RM 11.0 station from 1986 to 1991 (Figure 3-8).

Just upstream of the CSO zone at RM 7.1 (Station CR2), the current RBP assessment found the stream to be "slightly-impaired", apparently somewhat improved over the 1986 ICI rating of "fair." This finding might reflect improvement following the removal of CSOs. While the biological condition along the entire reach of the Little Cuyahoga (RM 0.3 - RM 11.3, excluding RM 11.0), exhibits degradation, the station at RM 7.1 seems to have rebounded slightly since the removal of the upstream CSO outfall. At RM 0.3, the ICI (Ohio EPA 1986 and 1991) and RBP assessments were in agreement, with macroinvertebrate community evaluations of "fair" and "moderately impaired," respectively.

Results from the present study are consistent with those obtained by Ohio EPA in previous surveys (1986 and 1991). The macroinvertebrate assemblage at RM 0.3 (Station CR3) reflects an impaired condition that has been present since at least 1986 probably attributable to the combined influence of CSOs and industrial input. One station upstream of the CSO outfalls (CR1) was in similar condition to that indicated from a 1986 assessment; Station CR2 apparently improved following CSO removal.

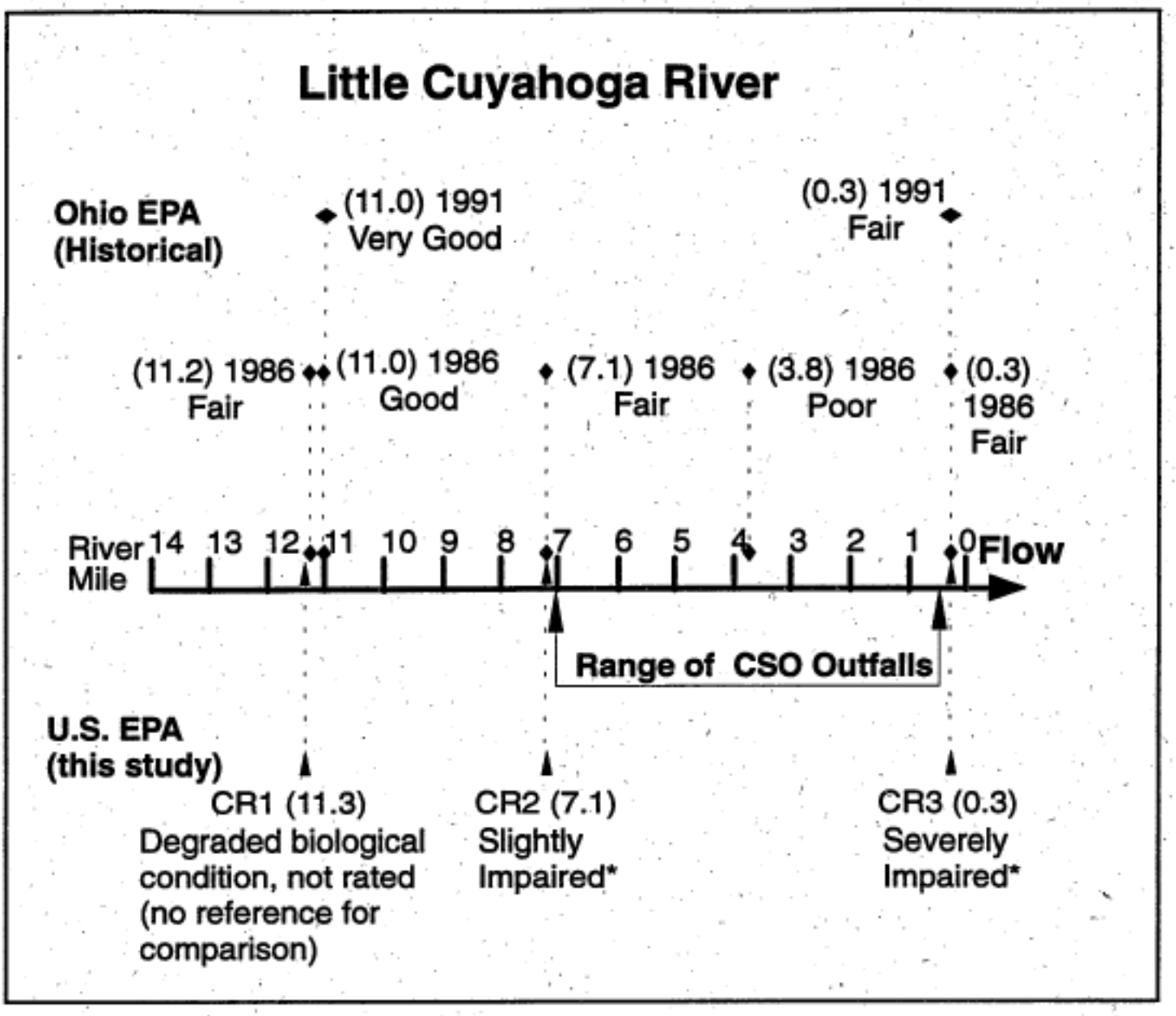

Figure 3-8. Linear comparison with Ohio EPA assessments on the Little Cuyahoga River.
**It should be noted that if an appropriate (non-impaired) reference condition was used as a baseline for comparison, all test sites for this study would likely receive lower biological assessment ratings.*

4

Evaluating the Biological Effects of Combined Sewer Overflows in New York

As a followup to the Ohio study, a design for an additional CSO bioassessment case study was developed. For this study, the State of New York was selected for the following reasons:

- Active CSOs are known to exist in several cities of the state.
- Historical data and assessments would likely be available.
- The state is dedicated to biological monitoring and assessment.

In addition to the objectives of the Ohio project (Section 3.0), the New York study was also designed to evaluate the effect of method variation on RBP performance: specifically, when varying the level of method intensity and rigor (screening level assessment, subsample size and taxonomic level), are the same conclusions reached regarding impairment of water resource integrity? The different levels of taxonomy are meant to roughly correspond to RBPII (family-level) and RBPIII (genus/species-level). RBPI has no standardized sampling and is based primarily of hand-turning of the substrate (cobble and gravel) and an estimate of relative abundance of higher taxonomic groups (i.e., family or order). Thus, the additional objectives for this study are meant to examine the effects of these differences on assessments; they are:

- To evaluate the ability of RBPI to detect CSO effects on the aquatic biota;
- To evaluate the effects of taxonomic level (family vs. genus/species) on metric behavior and overall assessments; and
- To evaluate the effects of subsample size (100-organism vs. 300-organism) on metric behavior and overall assessments.

RBPIII results are presented in this section; the evaluations of method variation are presented in Section 5.

4.1 Site Selection and Location Description

Eleven sampling stations were selected for this investigation: three stations each for the CSO-affected streams, Canastota Creek, Harbor Brook, and Onondaga Creek, and one station each for two regional reference sites on the Tioughnioga River (West Branch) and Furnace Brook (Table 4-1). Sampling stations on CSO receiving streams had the same general placement as in the Ohio study, with one location upstream of CSO outfalls, another downstream of at least initial CSO outfalls, and the third well downstream of any outfalls. The stations on Onondaga Creek, Harbor Brook, and Furnace Brook are located in Syracuse; those on Canastota Creek and the Tioughnioga River are in Canastota and Homer, respectively (Figure 4-1). The regional reference site selected by the New York Department of Environmental Conservation (NYDEC) for Onondaga Creek was the West Branch Tioughnioga River. Furnace Brook, south of Syracuse, was selected as the regional reference for Canastota Creek and Harbor Brook but was subsequently dropped after the evaluation of the biological metrics indicated impairment; the upstream reference site on Canastota Creek was used for reference instead. Table 4-1 presents detailed descriptions of sampling locations.

4.2 Results

4.2.1 Taxonomy and Metrics

Taxonomic results and counting exceptions are presented in Appendix B. Bioassessment scoring criteria were developed by dividing the metric value range into equal quadrisections, from the lowest possible value of a metric score (usually 0) to the 95th percentile of the maximum value observed for each metric. The scoring criteria for the genus/species-level, 300-organism subsample, which were used for the biological assessments, are presented in Table 4-2. Note that separate criteria were developed (Table 4-2) based on least-impaired conditions in Canastota Creek (for assessment of stations in that stream

Table 4-1 Biological sampling stations located in or near Syracuse, New York. Sampled 20-23 September 1993.

SITE	STATION TYPE	WATERBODY	GEOGRAPHIC DESCRIPTION OF SITE
CC1	Upstream Reference	Canastota Creek	Canastota Creek, off of Rt. 5 in Canastota, 1 block east of Mt. Pleasant Rd., ~50 meters upstream of the Rt. 5 bridge.
CC2	CSO Impact	Canastota Creek	Canastota Creek, downstream of Canastota Village, ~60 meters upstream of the Main St. bridge (across from Joe A. Stagnitti La.).
CC3	CSO Impact	Canastota Creek	Canastota Creek, perpendicular to Main St., behind the STP, ~12 meters upstream of the agricultural ditch on the left (upstream) side of the entrance field (behind STP).
HB1	Upstream Reference	Harbor Brook	Harbor Brook, ~150 meters upstream of the flow dissipator and ~350 meters upstream of the Velasko Rd. bridge.
HB2	CSO Impact	Harbor Brook	Harbor Brook, ~5 meters downstream of the State Fair Blvd. bridge, Syracuse.
HB3	CSO Impact	Harbor Brook	Harbor Brook, ~10 meters upstream of the Hiawatha Blvd. bridge, downstream of the pipe. RBP I only at this station.
CHR4*	Regional Reference	Furnace Brook	Furnace Brook, off of McDonald and Reinman Sts., behind Corcoran High School gym, just upstream of the asphalt bank.
OC1	Upstream Reference	Onondaga Creek	Onondaga Creek, in Kelly Park off of Dorwin Ave., ~75 meters upstream of the USGS gauging station.
OC2	CSO Impact	Onondaga Creek	Onondaga Creek, ~10 meters downstream of the Newell St. bridge between Vale and Onondaga Creek Blvd.
OC3	CSO Impact	Onondaga Creek	Onondaga Creek, ~30 meters upstream of footbridge behind the Utility Life Bldg.at the end of Plum St.; ~0.5 miles upstream of Onondaga Lake.
OC4	Regional Reference	Tioughnioga River (West Branch)	Tioughnioga River, in Durkee Memorial Park, Homer, NY, ~15 meters downstream of the Hwy. 11 bridge.

CC = Canastota Creek OC = Onondaga Creek HB = Harbor Brook CHR4 = Furnace Brook

* This station was not used in development of the scoring criteria due to biological impairment. Station CC1 was used for comparison for Canastota Creek and Harbor Brook.

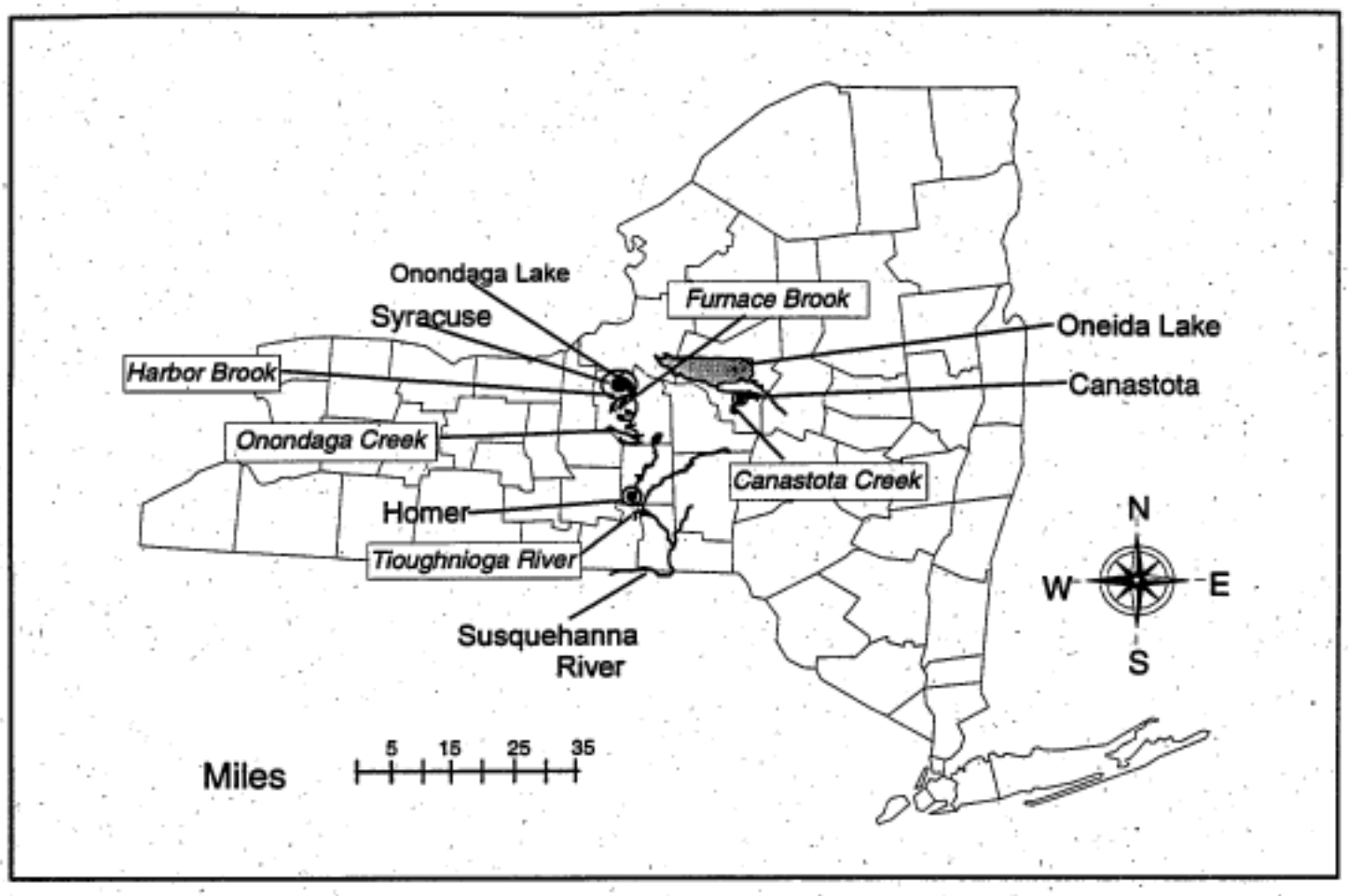

Figure 4-1. State of New York. Rivers and streams sampled for the biological assessment of combined sewer overflow effects, September 1993.

and Harbor Brook) and in the Tioughnioga River (for assessment of Onondaga Creek stations). The calculated or enumerated values for each of the metrics are given in Table 4-3.

4.2.2 Canastota Creek at Canastota, New York

4.2.2.1 Historical Information

The Canastota Creek watershed covers a drainage area of approximately 8.5 square miles and includes Cowaselon and Canaseroga Creeks. The drainage area encompasses Canastota, Lakeport, and agricultural lands. Canastota Creek flows through the town of Canastota and joins Cowaselon Creek on the northwest side of town. Before the construction of the WWTP, sewage was discharged directly into the lower part of Canastota Creek. At the time of the initial biological survey of these streams (Preddice 1975), the WWTP discharge was directly into Cowaselon Creek upstream of the Canastota Creek confluence.

A sample collected just downstream of the Main Street (Canastota) bridge in 1975, comprised a relatively tolerant macroinvertebrate assemblage (Preddice 1975) (Table 4-4). Several of these groups are indicative of potential organic enrichment; they were also found in low density. Other organisms at this site were *Cladophora* (Chlorophycophyta [green algae]) and some blue-green algae (Cyanophyophyta), and several species of bottom-feeding fishes (blacknose dace, longnose dace, creek chub, and white sucker). In spite of the appearance of suitable substrate quality and flow conditions, the low number of benthic macroinvertebrates found, combined with their relative tolerance, indicated a potential of simultaneous toxic input and nutrient enrichment (Preddice 1975). This assessment was considered consistent with the presence of both green and blue-green algae. The upstream source of toxicants was not determined; however, it was learned that an herbicide, atrazine, had been used. At the time, atrazine was considered to have only limited toxic effects on insects (Weed Science Society 1974) and, therefore, was not considered the source of the problem (Preddice 1975).

Table 4-2 Scoring criteria developed for the benthic macroinvertebrate assemblage, 300-organism subsample. For a description of the development of scoring criteria, see Section 2.3.1.

ASSESSMENT SCORES	SCORING CRITERIA							
	0	2	4	6	0	2	4	6
METRIC	Canastota Creek, Harbor Brook				Onondaga Creek, Tioughnioga River			
1. Taxa Richness	≤10	21-11	32-22	≥33	≤10	21-11	32-22	≥33
2. HBI	≥6.3	4.2-5.2	2.1-4.1	0-2.0	≥5.1	3.4-5.0	1.7-3.3	0-1.6
3. Sc/(Sc+Fil) x 100	7.7-0	15.5	23.3-15.6	≥23.4	≥13.3	26.7-13.4	40.1-26.8	≥40.2
4. EPT/(EPT+Chir) x 100	≤21.5	43.1-21.6	64.7-43.2	≥64.8	≤22.9	45.9	68.9	≥69
5. % Cont. Dom. Taxon	45.9	30.6-45.8	15.3-30.5	0-15.2	≥17.7	11.8-17.6	5.9-1.7	0-5.8
6. EPT Index	3-0	7-4	11-8	≥12	4-0	9-5	14-10	≥15
7. Shredders/Total x 100	2.2-0	4.5-2.3	6.8-4.6	≥6.9	3.7-0	7.5-3.8	4.3-7.6	≥11.4
8. H/T x 100	≥71.4	47.6-71.3	23.8-47.5	0-23.7	≥71.4	47.6-71.3	23.8-47.5	0-23.7
9. Pinkham-Pearson	2.3-0	4.7-2.4	7.1-4.8	≥2.7	2.0-0	4.1-2.1	6.2-4.2	≥6.3
10. QSI-Taxa	13.3-0	26.7-13.4	40.1-26.8	≥40.2	7.8-0	15.7-7.9	23.6-15.8	≥23.7
11. DIC-5	≤1	2	3	≥4	≤0	1	2	≥3
12. QSI-FFG	20.7-0	41.5-20.8	62.3-41.6	≥62.4	13.6-0	27.3-13.7	41-27.4	≥41.1

It was also discovered that a sewage/stormwater bypass pipe was present in the channel. This pipe was acknowledged as the probable source of nutrient loadings during storm flow (Preddice 1975).

More recently, at another site downstream of the Main Street bridge, Canastota Creek was found to be moderately impacted (Bode et al. 1993). Samples taken in early summer (19 June 1990) produced 24 percent midges (Chironomidae) and 69 percent aquatic earthworms (Oligochaeta), both of which are considered tolerant to severe pollution including conventional toxics, eutrophication, and habitat degradation. No mayflies, stoneflies, or caddisflies (Ephemeroptera, Plecoptera, and Trichoptera, respectively) were found, and the HBI fell in the "moderate impact" category.

4.2.2.2 Sampling Station Description and Habitat Quality

Three sampling stations were selected on Canastota Creek (Figure 4-2). The regional reference site for this stream was located on Furnace Brook (Station CHR4). Habitat assessment scores, along with measurements of dissolved oxygen, temperature, and conductivity, are presented in Table 4-5.

Furnace Brook, Station CHR4. This station was recommended by NYDEC for the regional reference for both Canastota Creek and Harbor Brook. The habitat quality rated optimal in all parameters except riparian vegetative zone width, which rated 15. The substrate was composed of cobble and small boulders with well-developed riffles. The riparian vegetation was very good, and the banks were stable. The habitat assessment score was 212. However, a

Table 4-3 Metric values calculated from 300-organism subsamples; based on genus/species level taxonomy. Bioassessment scores (in parentheses) are derived by comparing metric values to scoring criteria (Table 4-2).

METRIC	SAMPLING STATIONS								
	CC1	CC2	CC3	HB1	HB2	OC4	OC1	OC2	OC3
1. Taxa richness	31(4)	43(6)	31(4)	21(2)	18(2)	42(6)	41(6)	26(4)	23(4)
2. HBI	4.5(4)	5.1(2)	5.1(2)	5.7(2)	8.4(0)	4.4(2)	6.1(0)	5.5(0)	6.6(0)
3. Scr/(Scr + Fc) x 100	32.4(6)	26(6)	17(4)	0(0)	0(0)	56(6)	10.8(0)	0(0)	16.7(2)
4. EPT/(EPT + Chir) x 100	80.8(6)	60.4(4)	90.4(6)	56.3(4)	0(0)	92.3(6)	26.2(2)	58.6(4)	7.4(0)
5. % Contr. Dom. Taxon	28.1(4)	14.1(6)	25.8(4)	63.9(0)	60.1(0)	22.5(0)	11(4)	17(2)	24.4(0)
6. EPT Index	11(4)	10(4)	8(4)	7(2)	1(0)	18(6)	7(2)	6(2)	1(0)
7. (Shred/Tot) x 100	7.7(6)	9.2(6)	2.8(2)	7.5(6)	0.6(0)	4.5(2)	11(4)	13.1(6)	15.8(6)
8. (Hydro/Trich) x 100	94.1(0)	99.1(0)	100(0)	98.3(0)	0(6)	69.3(2)	97.8(0)	99.2(0)	100(0)
9. Pinkham-Pearson index	UR(6)	9.7(6)	6.6(4)	1.6(0)	0.7(0)	RR(6)	6.7(6)	8.4(6)	5.9(4)
10. QSI Tax x 100	UR(6)	55(6)	56(6)	25(2)	12(0)	RR(6)	19(4)	33(6)	11(2)
11. DIC-5	UR(6)	3(4)	3(4)	2(2)	1(0)	RR(6)	0(0)	1(2)	0(0)
12. QSI-FFG x 100	UR(6)	87.4(6)	77.6(6)	45.1(4)	28(2)	RR(6)	55.7(6)	57.2(6)	25.9(2)
Total (with paired) metrics	58	56	46	24	10	54	34	38	22
Biology (with paired) % comparison to reference	--	96	79	41	17	--	63	70	41
Total (without paired) metrics	34	34	26	16	8	30	18	18	14
Biology (without paired) % comparison to reference	--	100	76	47	23	--	60	60	47
Habitat score	139	132	92	182	57	191	86	114	118
Habitat % comparison to reference	--	95	66	131	41	--	45	60	62

No scrapers or filterer-collectors taken in sample.
UR = upstream reference
RR = regional reference (regional reference was not used for Canastota Creek and Harbor Brook, Section 4.2.2.2).
Note: station HB3 was not included in the biological assessment due to severe habitat alterations.

Table 4-4 Primary taxonomic composition of benthic macroinvertebrate samples taken by Preddice (1975). Canastota Creek, Canastota, New York, 29 July 1975.

TAXON	HIGHER LEVEL CATEGORY	COMMON NAME
Physa	Gastropoda: Physidae	snails
Asellus	Isopoda: Asellidae	sowbugs
Lumbricidae	Oligochaeta	aquatic earthworms
Tendepedidae	Diptera	true flies
Heptageniidae	Ephemeroptera	mayflies
Tricladida	Turbellaria	planarians (flatworms)
Hirudinea	Oligochaeta	leeches
Elmidae	Coleoptera	riffle beetles

hyperabundance of amphipods was found (nearly 76 percent of the 300-organism subsample was Gammarus); therefore, this station was not used as a representative of reference conditions. Furnace Brook was not used to establish scoring criteria.

Canastota Creek, Station CC1 (upstream reference). The station farthest upstream does not receive CSO input at or above this location. At this location the stream is approximately 2.5 meters wide with a dense canopy cover and variable bank stability. Where the samples were taken, the banks were in relatively stable condition with little evidence of accelerated erosion; however, areas of active bank erosion were observed upstream. The riparian vegetative zone on either side of the stream was less than 6 meters due to human activity. The substrate available for benthic fauna at this site consisted of a good mix of boulder and cobble, and well-developed riffles were prevalent. However, there were some problems with sedimentation and substrate embeddedness. The habitat assessment rating score was 149 (Table 4-5).

Canastota Creek, Station CC2. This station was the first station on Canastota Creek below CSO outfalls. Relative to the upstream reference station, CC2 had a reduced canopy cover, a predominantly sand and gravel bottom with a high degree of embeddedness, and less well-defined riffles. The stream here was approximately 3 meters wide and had moderately unstable banks with very poor riparian vegetation zones. The habitat assessment rating score was 132 (Table 4-5).

Canastota Creek, Station CC3. Station CC3 is behind the Sewage Treatment Plant; the stream in this area is approximately 3 meters wide. Sampling took place approximately 12 meters upstream from the agricultural ditch that enters on one the side of the stream. The habitat structure at this station was also more degraded than that of CC1. The substrate consisted almost entirely of sand; there was substantial sediment deposition and evidence of past channelization. The riparian buffer zone and the condition of the banks were both scored very poor. The habitat assessment rating score for this station was 92 (Table 4-5).

Overall, the best habitat quality on Canastota Creek was found at the upstream reference site, station CC1. The two downstream stations both experienced degradation in channel characteristics and poor riparian vegetative protection. The individual components of the physical habitat structure that were rated in the poor and marginal ranges, at both stations CC2 and CC3, were related to the lack of riparian buffer zone and the high degree of embeddedness. In addition, at CC3 the condition of the banks and increase in sediment deposition related to erosion were rated poor. Station CC3 rated consistently lowest in most habitat parameters, which is reflected in the percent comparability (66 percent) to the reference station. Habitat condition should be considered degraded at station CC3; habitat quality should not be limiting to the biological condition at Stations CC1 or CC2, despite some problems at CC2.

4.2.2.3 Biological Assessments

The stations on Canastota Creek were assessed for the RBPI as slightly to moderately impaired (CC1) and moderately impaired (CC2, CC3). A further description of this screening-level assessment and how it compares to the more rigorous RBPIII assessment can be found in Section 5.1.

For biological assessments using CC1 as the reference site, metric totals without paired metrics were used for percent comparisons. The upstream (CC1) and middle (CC2) stations were very similar in their biological condition, the latter having the same assessment score as the former (Table 4-3). However, more detailed interpretation of individual metric values shows substantial differences in number of taxa. Twelve additional taxa were found at CC2, eight of which were genera of the Chironomidae (Appendix B), a group generally considered to be pollution-tolerant. All of the additional midge genera have designated

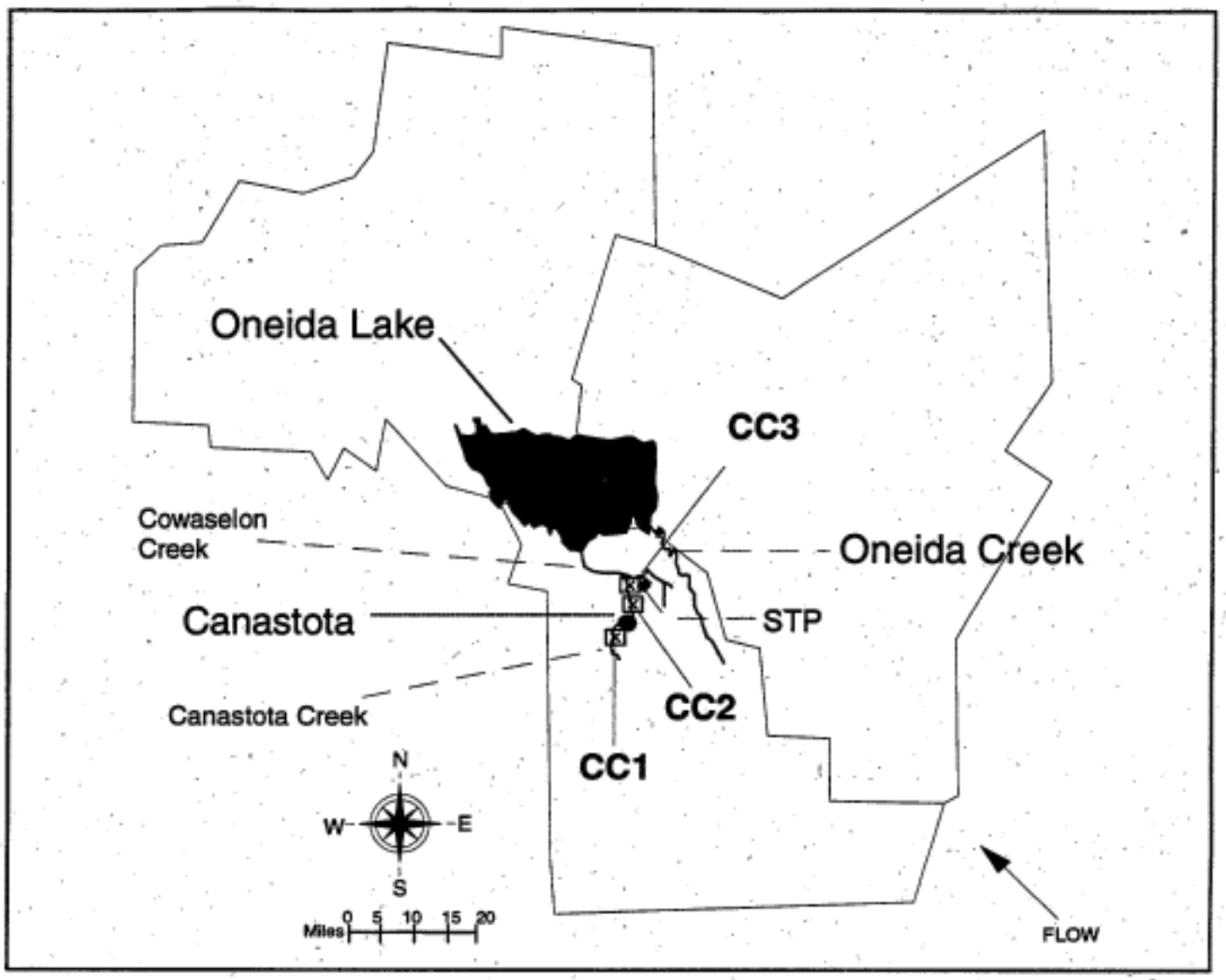

Figure 4-2. Locations of sampling stations on Canastota Creek. STP = Sewage Treatmant Plant.

tolerance values of 5 or above, indicating their high tolerance of or potential for positive response to pollution. The high tolerance values caused an increase in the HBI. There was also a higher proportion of Stenelmis (Coleoptera: Elmidae: riffle beetles) at CC2 than was seen upstream. This might have been due to a combination of increased growths of periphyton and filamentous green algae responding to removal of some of the canopy (providing increased light), and upstream organic enrichment.

Station CC3 had habitat that was further degraded and a biota that compared at 76 percent to that at the upstream-most site, substantiating what was seen at the upstream stations. That is, there is likely some organic enrichment of Canastota Creek occurring upstream of any CSO effects, possibly from agricultural activities. Though none of the three stations had excessively high values for the metric "percent contribution of dominant taxon," samples from each were dominated by the Hydropsychidae, often seen in high numbers in organically enriched streams. The degree of habitat degradation between CC1 and CC2 (11 percent change) is less than that between CC2 and CC3 (30 percent change), but both indicate either nonimpairment (CC2) or only slight impairment. However, in the absence of a suitable (nonimpaired or minimally impaired) regional reference site for comparison, CC1 and CC2 should be considered slightly to moderately impaired; CC3, moderately impaired.

4.2.3 Harbor Brook in Syracuse, New York

4.2.3.1 Historical Information

Historical data on Harbor Brook (11.8 square miles drainage area) are from NYDEC sampling at stations near Highway

Table 4-5 Canastota Creek habitat assessment rating score.

HABITAT PARAMETERS (0 - 20)		SCORES			
		Canastota Creek Sampling Stations			
		CC1	CC2	CC3	CHR4[a]
Substrate and Instream Cover	Instream Cover (Fish)	12	13	8	19
	Epifaunal Substrate	16	11	10	19
	Embeddedness	13	8	7	16
	Velocity/Depth Regimes	18	12	7	19
Channel Morphology	Channel Alteration	17	10	14	16
	Sediment Deposition	4	10	5	18
	Frequency of Riffles	19	12	8	19
	Channel Flow Status	13	15	8	19
Riparian	Conditions of Banks	12	10	5	18
	Bank Vegetative Protection	15	14	9	17
	Grazing or Other Disruptive Pressure	8	14	10	17
	Riparian Vegetative Zone Width (Least Buffered Side)	2	3	1	15
TOTAL SCORE		149	132	92	212
Physicochemical Parameters	Temp (°C)	12	11.8	14.2	10.5
	pH	8	8.23	8.88	8
	Conductivity	16.38	15.91	---[b]	---[b]
	DO	10.3	8.5	8.1	9.8

[a] Furnace Brook, regional reference site, not used in biological assessments due to impairment.
[b] Lack of physicochemical data is due to equipment failure.

173 at Split Rock and another station approximately 0.8 kilometer upstream from the mouth into Onondaga Lake (Bode et al. 1989). The upstream station at Split Rock is approximately 1.5 miles south (upstream) of HB1 of this study. At the Split Rock station the benthic macroinvertebrate assemblage was assessed as moderately impacted. This stream is known to be intermittent (Bode et al. 1989), and the abundance of taxa tolerant to temporary desiccation influenced this assessment. Bode et al. stated that the chemical water quality might actually have been nonimpacted.

The second station assessed by Bode et al. was located between HB2 and HB3 (Bode et al. 1989). The channelized and degraded habitat produced a sample made up mostly of Chironomidae, Oligochaeta, Hirudinea, and Amphipoda (midges, earthworms, leeches, and scud, respectively). All four parameters rated as poor (species richness, 8; biotic index, 9.7; EPT value, 0; and percent model affinity, 15) and resulted in an assessment of "severely impacted." Descriptions of these parameters are presented in Bode et al. (1989, 1993) and Bode and Novack (1995).

4.2.3.2 *Sampling Station Description and Habitat Quality Assessments*

Three sampling stations were selected on Harbor Brook (Figure 4-3). It was not possible, however, to include HB3 in the biological assessments due to the deep, soft bottom, which is not suitable for a wadable kick net sample. The length of Harbor Brook within this study was completely channelized. Approximately 150 meters downstream from the farthest upstream station (HB1) is a flow dissipator, through which water enters an emergent-macrophyte-filled retention basin. Further downstream, the water is subjected

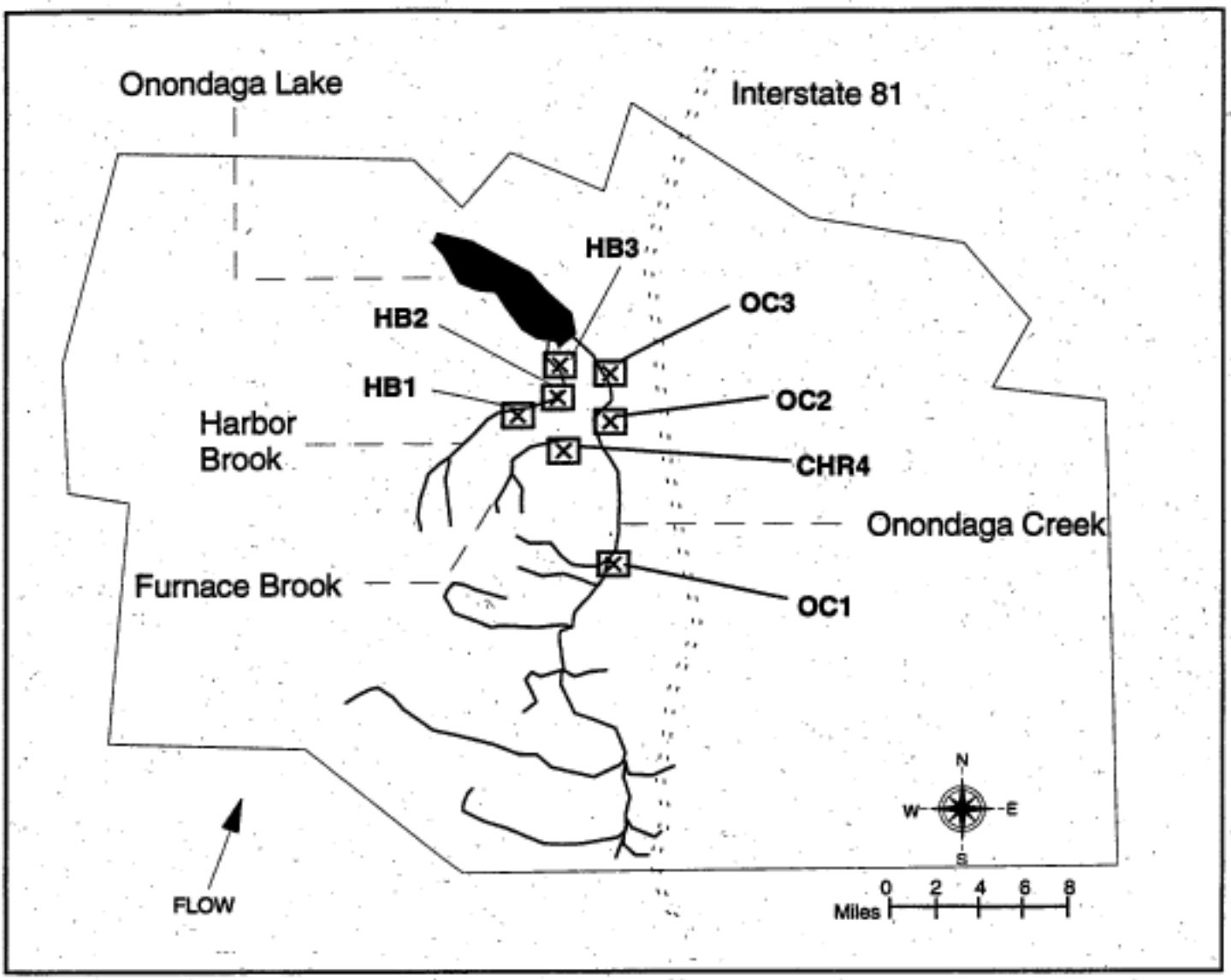

Figure 4-3. Locations of sampling stations on Onondaga Creek, Harbor Brook, and Furnace Brook.

to a flow splitter; the flow then enters a cement- and rock-sided channel; this type of channel with armored sides continues for the rest of the length of Harbor Brook. For some intermediate distance in the study length, the stream has been closed on the top, making it essentially a subsurface channel. The second and third sampling stations (HB2 and HB3, respectively) were located just downstream from where the channel was no longer covered; that is, in the section between State Fair Boulevard and Hiawatha Street. Thus, there is a major difference in habitat quality between the sampling site farthest upstream (HB1) and the two downstream sites, which should be recalled in these comparisons. The regional reference site for this stream (Furnace Brook [Station CHR4]) was dropped as the regional reference site; therefore the upstream station on Canastota Creek (CC1) was used for reference. Habitat assessment scores, along with measurements of dissolved oxygen, temperature, and conductivity, are presented in Table 4-6.

Harbor Brook, Station HB1. This station was located approximately 150 meters upstream of the flow dissipator and about 350 meters upstream of Velasko Road. The stream was approximately 2.5 meters wide, and the substrate was composed of cobble and gravel with very little embeddedness. The frequency of riffles was optimal as was the condition of the banks and the riparian vegetative protection. Even though the stream was located among relative heavy urbanization, its physical quality was very good. This site scored high on the habitat assessment at 182.

Harbor Brook, Station HB2. HB2 was located approximately 5 meters downstream of the State Fair Boulevard bridge off Hiawatha Street. The stream was completely channelized with no riffles and very slow-moving water; the width was approximately 2 meters. The substrate at this station was mostly sand with a little gravel. The only parameters scored above poor or marginal were channel

Table 4-6 Harbor Brook habitat assessment scores.

HABITAT PARAMETERS (0 - 20)		SCORES			
		Harbor Brook Sampling Stations			
		HB1	HB2	HB3	CHR4[a]
Substrate and Instream Cover	Instream Cover (Fish)	16	2	2	19
	Epifaunal Substrate	19	1	0	19
	Embeddedness	12	5	1	16
	Velocity/Depth Regimes	18	1	1	19
Channel-Morphology	Channel Alteration	13	1	1	16
	Sediment Deposition	17	1	1	18
	Frequency of Riffles	17	1	1	19
	Channel Flow Status	12	18	19	19
Riparian	Conditions of Banks	17	18	19	18
	Bank Vegetative Protection	18	7	10	17
	Grazing or Other Disruptive Pressure	18	1	1	17
	Riparian Vegetative Zone Width (Least Buffered Side)	5	1	1	15
TOTAL SCORE		182	57	54	212
Physicochemical Parameters	Temp (°C)	13	12.5	---[b]	10.5
	pH	7.95	7.86	---[b]	8
	Conductivity	---[b]	---[b]	---[b]	---[b]
	DO	9	7.3	---[b]	9.8

[a] Furnace Brook, regional reference site, not used in biological assessments due to impairment.
[b] Lack of physicochemical data is due to equipment failure.

flow status and condition of banks; the banks were armored. This station had severely degraded habitat and scored only 57.

Harbor Brook, Station HB3. This station, located about 5 meters upstream of the Hiawatha Street bridge, was extremely degraded. All habitat parameters scored in the poor range except for the channel flow status and condition of banks (due to the armored sides). The substrate at this site was a grayish-black muck; the stream was too deep here to be considered "wadable" for sampling. The habitat assessment score for HB3 was 54.

The poor habitat at both HB2 and HB3 can be considered to be extremely limiting to the biological condition of this stream.

4.2.3.3 Biological Assessments

Screening-level assessments (RBPI) indicate moderate (HB1) to severe (HB2, HB3) impairment on Harbor Brook. Comparisons between the screening-level assessment and the RBPIII assessment are discussed in Section 5.1.

Because of the extreme habitat alteration, samples could not be taken from what was to be the downstream-most station (HB3). As discussed in Section 4.2.2.2, the regional reference site for Harbor Brook (Furnace Creek) was dropped as a baseline comparison due to biological impairment; station CC1 was used for reference. HB1 and HB2 scored 41 percent and 17 percent comparability, respectively, to the upstream site on Canastota Creek (CC1). The upstream station (HB1) had a metric score for "percent contribution of dominant taxon" of 63.9 that represented

246 *Gammarus* (Crustacea: Amphipoda: Gammaridae); the second most dominant, comprising another 15 percent of the individuals, was of the caddisfly family Hydropsychidae (Trichoptera: *Cheumatopsyche*, *Hydropsyche*). Both of these groups are considered to be tolerant to some level of habitat degradation, positively responsive to nutrient enrichment (Hydropsychidae, *Gammarus*), and tolerant to some potentially toxic inputs (*Gammarus*). Compared to CC1, this station had a lower taxa richness, higher HBI, higher percent dominant taxon, and lower EPT index. Each shows the expected direction of the metric value change when exposed to physical and chemical degradation.

The downstream station (HB2) had completely different habitat and flow regime. Here, the two most dominant taxa were *Chironomus* (Diptera: Chironomidae) (60 percent, 194 individuals of 385 total) and *Gammarus* (66 individuals). The genus *Chironomus* is one of the taxa more tolerant of chemical pollution and habitat degradation. Also found were 13 specimens of a cyclopoid copepod (Crustacea: Copepoda) normally found in lakes and reservoirs. At this station, water was deep and slow-moving, perhaps making it suitable for copepods. There were six different genera of Oligochaeta (aquatic earthworms) that are as a whole considered to be tolerant of a range of severity in habitat degradation. The HBI was 8.4 in contrast to the 5.7 and 4.5 of HB1 and CC1, respectively. This is indicative of a sample dominated by individuals of pollution-tolerant taxa.

Even though the habitat and most of the benthic metrics differed substantially between HB1 and HB2, the "taxa richness" of the stations was nearly identical. This is an illustration of why single measurement parameters should not be relied upon for performing biological assessments; rather, single parameters should be used to interpret overall multimetric assessment scores and aid in determining causes of impairment. Station HB1 should be considered moderately impaired and HB2 severely impaired.

4.2.4 Onondaga Creek in Syracuse, New York

4.2.4.1 Historical Information

The Onondaga Creek drainage covers approximately 111 square miles. It traverses rural agricultural communities, a Native American reservation, and downtown Syracuse. Bode et al. (1989) sampled Onondaga Creek at two locations, one about 1 mile upstream of Onondaga Lake and the other near Cardiff just off Webster Road and about 15 miles upstream of the first site. The site upstream of Onondaga Lake was assessed as "severely impacted" in 1989 (Bode et al. 1989) and 1990 (Bode et al. 1993). In 1993 Bode et al. found only Chironomidae and Oligochaeta, both considered to be strongly pollution-tolerant. Other characteristics of the sample were eight species (poor), a biotic index of 9.7 (poor), EPT value 0 (poor), and percent model affinity 15 percent (poor).

Tissue analysis of caddisflies collected at the site indicated no elevated levels of metals above background levels; crayfish had elevated levels of the PCB aroclor 1254 (0.42 µg/g, which is below the U.S. Food and Drug Administration action level of 2 ppm). Parameters of concern in the water column were aluminum, iron, lead, mercury, zinc, dissolved solids, and both total and fecal coliform; manganese was borderline. Bottom sediments contained levels of copper, zinc, lead, mercury, PCBs, and DDE above background levels but below assessment criteria levels. Toxicity testing indicated that significant mortality and reproductive impairment occurred in assays during 1990.

It should also be noted that 17 miles of Onondaga Creek upstream of Syracuse are affected by mud boils. These geomorphic reactions to excessive groundwater drawdown result in periodic episodes of hyperturbidity. Also, sections of this creek have been closed to fishing due to brine discharges and mining operations. In spite of the hyperturbidity, the macroinvertebrate community at the NYDEC upstream station located in Cardiff was found to be in "slightly impacted" condition in 1989 and 1990, with 24 species, a biotic index of 6, EPT value of 4, and 68 percent model affinity (Bode et al. 1993).

4.2.4.2 Sampling Station Descriptions and Habitat Quality

Tioughnioga River, West Branch, Station OC4. The Tioughnioga River was selected as the regional reference for Onondaga Creek (Figure 4-4). This station was located in Homer, New York, just downstream of the Highway 11 bridge and upstream from potential backwater influence resulting from a slow segment of the river that probably widened during construction of Interstate 81. Habitat quality was rated optimal for most of the parameters. The substrate was composed of cobble, and riffles were well-developed and prevalent. The lowest habitat score was for the riparian buffer zone width. The overall habitat assessment score was 191 (Table 4-7). Habitat quality would not limit the biological communities at this regional reference site.

Three sampling stations were selected on Onondaga Creek (Figure 4-3). The entire length of Onondaga Creek downstream of the Onondaga Tribal Reservation is channelized, and the state of the streambanks differs at all three stations. At the upstream station, the banks are mown and grassy; at the middle station, they are armored; and at the farthest downstream station, they consist of rubble and debris with some weedy vegetation. Habitat

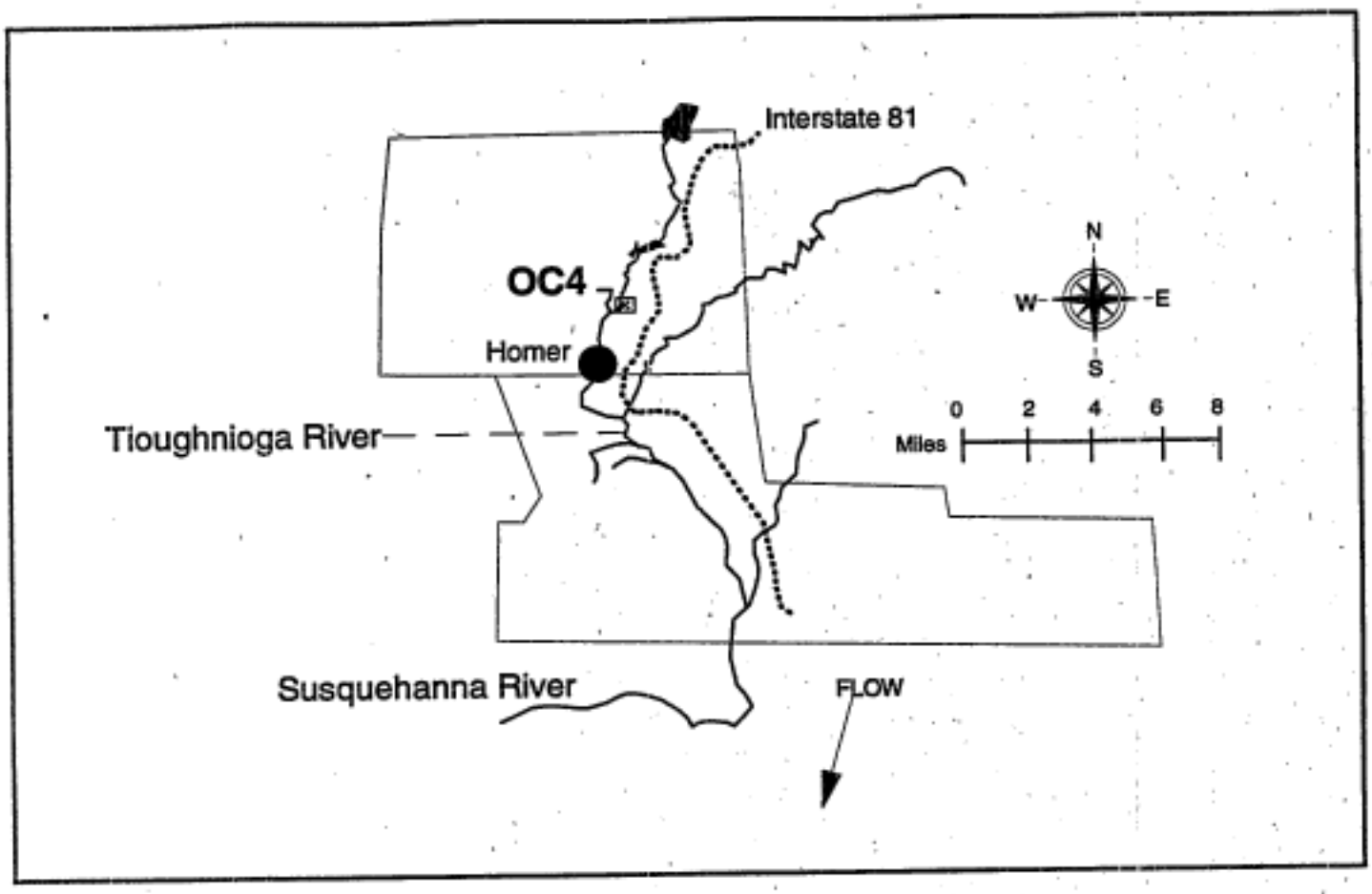

Figure 4-4. Location of sampling station on the Tioughnioga River (West Branch).

assessment scores along with measurements of dissolved oxygen, temperature, and conductivity are presented in Table 4-7.

Onondaga Creek, Station OC1. Station OC1 was estimated as approximately 50 meters wide and is channelized. This station was used as the upstream reference station because it was located above any CSO outfalls. The substrate was composed of mainly gravel with a high degree of embeddedness, there were no fast water riffles present at this site, and the velocity/depth regime was rated "very poor." The habitat variability at this site was minimal. The banks were stable but very little riparian buffer existed. According to DEC staff water was unusually clear at this station (S. Cook, personal communication, September 1993), at the time of sampling. The habitat assessment score was 86.

Onondaga Creek, Station OC2. The stream at this station was approximately 8 meters wide and was completely channelized with concrete armored sides. The substrate was composed of cobble with intermittent riffles; there was little embeddedness and sediment deposition. There was no riparian vegetative buffer zone, but the condition (i.e., stability) of the banks was rated optimal due to the armored sides. The habitat assessment score was 114.

Onondaga Creek, Station OC3. This station was located approximately 0.8 kilometer upstream of Onondaga Lake. The stream was approximately 10 meters wide with cobble substrate and intermittent, well-developed riffles. Riparian zone scores were the lowest rated at this station, which had little to no buffer zone and little bank vegetative protection. The overall habitat assessment score was 118.

The upstream reference station, OC1, was rated the poorest in the habitat assessment. The habitat at this station would seem to be the limiting factor for the development of the biological community. Although the bank vegetative stability at this site was rated in the optimal range, upstream erosion caused a marked increase in embeddedness and sediment deposition, which decreases the amount and variety of epifaunal substrate habitat available for colonization. The habitat assessments for the two CSO receiving stations were scored higher than that for OC1 in all instream habitat characteristics because there was a lack of apparent sedimentation and embeddedness. In spite of the absolute channelization with

Table 4-7 Onondaga Creek habitat assessment scores.

HABITAT PARAMETERS (0 - 20)		SCORES			
		Onondaga Creek Sampling Stations			
		OC1	OC2	OC3	OC4[a]
Substrate and Instream Cover	Instream Cover (Fish)	5	10	11	16
	Epifaunal Substrate	7	18	16	19
	Embeddedness	3	17	11	15
	Velocity/Depth Regimes	2	7	10	16
Channel-Morphology	Channel Alteration	1	1	3	13
	Sediment Deposition	6	16	13	16
	Frequency of Riffles	2	5	8	19
	Channel Flow Status	18	17	16	18
Riparian	Conditions of Banks	19	19	16	19
	Bank Vegetative Protection	18	1	7	17
	Grazing or Other Disruptive Pressure	4	3	6	15
	Riparian Vegetative Zone Width (Least Buffered Side)	1	0	1	8
TOTAL SCORE		86	114	118	191
Physicochemical Parameters	Temp (°C)	12.5	15	13.5	13
	pH	8.14	7.85	7.89	8.03
	Conductivity	---[b]	---[b]	---[b]	---[b]
	DO	10.6	9.8	8.3	10

[a] Tioughnioga River, regional reference site.
[b] Lack of physicochemical data is due to equipment failure.

mortared block banks, sufficient stands of older deciduous trees were present to supply substantial leaf litter and woody debris to the channel.

4.2.4.3 Biological Assessments

Screening level assessments (RBPI) for Onondaga Creek indicated moderate (OC1) to moderate-severe (OC2, OC3) impairment; the regional reference site on the Tioughnioga River was screened as having slight impairment. Further comparisons between the screening and rigorous assessments is discussed in Section 5.1.

This creek has been channelized along most of its length; for each of the three sites sampled in this study, characteristics of the channelization differed markedly and might temper conclusions. The upstream-most station, OC1, is in a section of the channel that is very wide and shallow and produced 39 taxa (in essence identical to the 40 from the Tioughnioga River, the regional reference site). Of these 39, however, 24 were genera of Chironomidae (midges), considered to be an overall pollution-tolerant taxon. This finding is reflected in its relatively high HBI of 6.1, contrasted to the 4.4 of the Tioughnioga site (OC4). Although *Cryptochironomus* (Chironomidae) accounts for the 11 percent dominant taxon (29 individuals), another 3 genera produced 23, 22, and 19 individuals. *Hydropsyche* and *Limnodrilus* (Oligochaeta: Tubificidae) were also dominant in these samples; they are both considered relatively pollution tolerant and often respond positively to organic nutrients. Station OC1

was calculated as being 63 percent comparable to the regional reference site and should be considered to have moderate impairment.

Station OC2, the middle station on Onondaga Creek and the first to receive CSO inputs, produced a sample that seemed to have a biological condition slightly improved over that seen at OC1. This is due primarily to a higher EPT-Chironomidae ratio, higher percentage of shredders in the sample, and higher QSI-Taxa (Table 4-3). However, as with OC1, the dominant taxa was comprised of Hydropsychidae and Chironomidae. This station was 70 percent comparable to the regional reference and should be considered to have moderate impairment.

The farthest downstream station, OC3, was rated as 41 percent comparable to the regional reference site. It produced a sample that was dominated primarily by *Nais* and *Ophidonais* (Oligochaeta: Naididae) (24 percent and 20 percent of total sample, respectively), and *Cricotopus* (Diptera: Chrionomidae) (12 percent of total sample). Also represented in a larger proportion than other taxa in this sample were *Dugesia* (Turbellaria) and Hydracarina sp. (Acari). All of these groups are considered to be tolerant to some levels of physical habitat degradation and toxicant input, and they often respond positively to increased nutrient loads. Station OC3 should be considered as severely impaired. Heavy urbanization effects make it difficult to isolate CSO effects in Onondaga Creek. However, excessive algal growth and high numbers of tolerant taxa suggest that substantial organic enrichment had occurred; the nutrient loading had likely originated with the CSOs.

5

Evaluation of Method Variation

The purpose of this chapter is to address issues related to method rigor and the effect of different levels of rigor on assessment results. A critical factor in selecting the level of application of RBPs is the availability of tiers. RBPI, II, and III represent three levels of intensity with RBPI being the most rapid and least rigorous (Plafkin et al. 1989). RBPI is based only on field observation of benthic invertebrates without any standardized sampling effort or index/metric calculations and interpretations. RBPII and RBPIII employ standardized sampling gear and effort, field and laboratory taxonomic identification, respectively, and subsampling. Decisions on which of these protocols to use are usually focused on some combination of these components in the context of protocol sensitivity and resource availability (Ferraro et al. 1989; Ferraro and Cole 1992). The analyses below are designed to evaluate the effects of subsample size and taxonomic level on metric performance and overall assessment results; these comparisons were conducted only for the New York case study. Though the results here might produce some conclusions on methods, it should be realized that these comparisons will not necessarily apply to other regions of the country.

5.1 Adequacy of Screening Level (Rapid Bioassessment Protocol)

RBPI screening level assessments are based on the relative abundance of organisms collected at a site. Collection of macroinvertebrates consists of turning over rocks (hand picking) and/or taking qualitative samples with a dipnet. These samples are supplemented by field examinations of periphyton, macrophytes, slimes, and fish which provide additional information for determining presence or absence of degradation. The variety of organisms (taxa richness), their relative tolerance levels, and factors observed for other biota, are then used to determine if the site is impaired. The adequacy of this approach relies on three basic factors: (1) that the assessment needed provides only the presence or absence of degradation, not detailed information as to the nature and cause of the degradation, (2) that the individual performing the assessment has a strong familiarity with aquatic invertebrate taxonomy generally at family-level, and (3) that the individual has knowledge of or access to information on relative pollution tolerance and functional feeding group associations of different aquatic biota.

The assessments produced by this screening level effort are presented in Table 5-1. These results did show sampling stations where there was impaired biological condition (Table 5-1). Most of the screening level assessments fell within the range of the higher level assessment (Table 5-2).

This screening level of assessment did underestimate impairment on one occasion, station CHR4, the initial regional reference site for Canastota and Harbor Brook that was dropped after further assessment. The screening level assessment notes the relative abundance and variety of organisms observed. The categories of abundance are:

- Rare <3
- Common 3-9
- Abundant >10
- Dominant >50 (estimate).

Initial assessment of station CHR4 showed a good variety of sensitive organisms (e.g., Plecoptera, Ephemeroptera-dominant). However, with such a rating system, the hyper-abundant Amphipola was given the same rating, i.e., dominant. Further evaluation of CHR4 using RBPIII level assessment revealed that Gammarus (Amphipoda: Gammaridae) comprised ~76 percent of the sample thus indicating impairment of the aquatic community. Overall, however, the RBPI is an adequate and cost-effective screening level assessment.

5.2 Metric Performance with Variable Methods

The different assessment levels of RBPs provide a means for agencies to tailor their biological monitoring programs to suit

Table 5-1 Narrative screening-level assessments (RBPI) of 10 study stations in New York State performed 20-23 September 1993. Use of narratives for impairment is based on the following categories of *increasing* biological degradation or impairment: minimal-slight-moderate-severe.

STATION	IMPAIRMENT	REASON(S) FOR ASSESSMENT
CC1	slight to moderate	1. Dominance of relatively tolerant Hydropsychidae (net-spinning caddisflies) and Elmidae (riffle beetles). 2. Heavy embeddedness of substrate, some upstream bank instability. 3. Narrow buffer zone, both sides. 4. Potential organic enrichment from agricultural operations.
CC2	moderate	1. Dominance of Hydropsychidae and Elmidae (both relatively tolerant); abundant Oligochaeta (aquatic earthworms). 2. Substrate almost completely sand and some small gravel. 3. Considerable upstream bank instability. 4. Removal of canopy on one side. 5. Abundant growths of blue-green and filamentous green algae on substrate. 6. Habitat degradation and organic enrichment.
CC3	moderate	1. Dominance of Hydropsychidae and Elmidae; Oligochaeta and Chironomidae (midges) common. 2. Substrate almost completely composed of sand and small gravel. 3. Severe bank instability. 4. Narrow buffer zones on both sides; agricultural fields within 5-7 meters on both sides. 5. Habitat degradation, organic enrichment, potential highway and agricultural runoff problems.
HB1	moderate	1. Dominance of Amphipoda (scud), Chironomidae and Hydropsychidae, all relatively tolerant. 2. Some embeddedness as evidence of upstream erosion. 3. Narrow vegetated buffer zone, both sides; little or no canopy cover. 4. Abundant growths of filamentous green and blue-green algae, and mosses. 5. Habitat degradation, organic enrichment, potential toxicants.
HB2	severe	1. Dominance of Amphipoda and Chironomidae, both considered relatively tolerant; Oligochaeta and Physidae abundant. 2. Copepoda, normally inhabiting standing waters, abundant. 3. Extreme habitat modification, channelized, stone walls, very low current velocity, deep, no riffles. 4. Habitat degradation, organic enrichment, potential toxicants.
HB3	severe	1. Dominance of Gastropoda (probably physidae), Chironomidae, and Hirundinea, all considered tolerant. 2. Extreme habitat modification, channelized, stone walls, low current velocity, deep, no riffles, silty/muck bottom with macrophytes. 3. Habitat degradation, organic enrichment, potential toxicants.
CHR4	minimal	1. Hyper-dominance of Amphipoda outweighed by considerable diversity of taxa recognized as relatively pollution-sensitive including Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies) (several families of the latter). 2. High-gradient, no upstream habitat degradation/modification. 3. Dominant growths of epilithic mosses and some filamentous green algae, potential for minor organic enrichment.
OC1	moderate	1. Dominance of Oligochaeta and Chironomidae, both relatively tolerant of both physical and chemical disturbances. 2. Ephemeroptera, Plecoptera, and Coleoptera, each with a mixture of tolerant and intolerant species, considered common. 3. Channelized, uniform habitat, embedded substrate, lack of riparian vegetation. 4. Potential organic enrichment.

Table 5-1 (continued).

STATION	IMPAIRMENT	REASON(S) FOR ASSESSMENT
OC2	moderate to severe	1. Dominance of Hydropsychidae, exhibits strongly positive response to organic enrichment and tolerance to some physical degradation. 2. Amphipoda, Oligochaeta, and Chironomidae considered abundant; all are tolerant. 3. Channelized with mortared stone walls, and considerable accumulation of gravel and cobble; minimal riparian vegetation. 4. Likely receiving considerable organic inputs.
OC3	moderate to severe	1. Dominance of Oligochaeta. 2. Planaria, Hirudinea, Amphipoda, Hydropsychidae, and Chironomidae considered abundant-all pollution-tolerant forms. 3. Channelized, very narrow riparian zone, heavy urban development on both sides, much coarse human trash and other debris. 4. Strong sewage odor. 5. Likely receiving heavy organic inputs combined with other urban runoff.
OC4	slight	1. Dominance of Trichoptera (several families) and Elmidae; some sub-taxa can be positively responsive to organic enrichment. 2. Hydracarina, Ephemeroptera (several families), and Chironomidae considered abundant; some taxa are sensitive, others are tolerant. 3. Good substrate diversity and riparian vegetation with canopy. 4. Some potential for asphalt runoff and a mixture of slight organic enrichment combined with low-level toxicants.

their needs. RBPI is used as an initial screening level assessment for many sites. If an impaired biological condition is noted, further assessment may be carried out with RBPII (family level taxonomy) or RBPIII (genus/species level taxonomy). The study was designed to compare results from RBPII with RBPIII. RBPII requires specimen identification no finer than to family level, whereas RBPIII uses "the lowest practical taxonomic level" (Plafkin et al. 1989), generally genus or species level. Therefore, to address the questions related to level of taxonomic identification, two datasets, one based on family-level taxonomy and one based on genus/species level, were needed. Results received from the laboratory were generally at the genus or species level (Appendix B). For a family-level dataset, taxa were combined under the family name and the number of individuals for each family was summed.

In order to evaluate sample size, it was necessary to calculate metrics and develop scoring criteria based on both the 100-organism and 300-organism subsamples. Data sets representing the latter were obtained by combining the data from 100- and 200-organism subsamples for each sampling station.

Metric values calculated based on 300-organism subsamples with genus/species-level taxonomy are presented in Table 4-3. The metric values to which these are compared are based on (1) family-level identification of 100-organism subsamples (Table 5-3) and (2) genus-level identification of 100-organism subsamples (Table 5-4).

5.2.1 Taxonomic Level Effects on Metric Performance

The level of taxonomy used for a biological assessment depends on the program objectives and resources. Biological assessment results may not vary substantially between family versus genus/species level taxonomy, however, interpretation of results may be problematic at the family (or higher) level. If broad-scale status analyses are desired for a large number of sites, RBPII assessment level may be adequate. If, for example, causal relationships need to be identified, RBPIII would be a better alternative potentially giving greater sensitivity.

Using the metric values calculated on 100-organism subsamples, comparisons of the effect of taxonomic level were made based on (1) performance of single metrics and (2) total bioassessment score. For both, correlation scatterplots were developed that illustrate the relationship between these measures at a single sampling station when differential taxonomic resolution is used. At the family level of identification, we would expect a smaller number of groupings with a larger number of individuals than is

Table 5-2 Comparison of biological assessments between RBPI and RBPIII

STATION	RBPI ASSESSMENT	RBPIII ASSESSMENT
CC1	slight to moderate	slight to moderate
CC2	moderate	slight to moderate
CC3	moderate	moderate
HB1	moderate	moderate
HB2	severe	severe
HB3	severe	*
CHR4	minimal	severe
OC1	moderate	moderate
OC2	moderate to severe	moderate to severe
OC3	moderate to severe	moderate to severe
OC4	slight	slight

*no further assessment was conducted on this site due to severe habitat alterations.

expected with genus/species-level taxonomy. Perfect (1:1) agreement between the metric values at a single station with different taxonomic levels will be reflected by a point lying on the diagonal. Conversely, the more a point is removed from the line, the greater is the disagreement between treatments. In cases where there seemed to be a non-trivial difference between the two treatments, a Spearman rank correlation was used for confirmation. The Spearman rank correlation provides a measure of how similar the rank order is between two ordered lists. For example, if the rank order is A>B>C for both treatments, the results would give a high R and low p-value for the Spearman's test.

Alternatively, if the order is A>B>C for one treatment and C>B>A for the other, we would see a low R and a high p-value. The interpretations between the two treatments could be very different. This test provides one indication of whether differences in treatments will cause differences in interpretation of results, that is, relative station condition.

Total Bioassessment Score. There was no difference in total aggregated metric score between the two taxonomic levels (Figure 5-1) when comparing station rank orders (Spearman rank correlation, R=0.94, p=0.0001).

Metric 1. Taxa Richness. This metric had a value range of 8 to 16 among stations when based on family-level identifications; the value range broadened to 8 to 31 when based on genus/species-level identification. When compared within each station (Figure 5-2), the expected relationship of higher number of taxa for finer taxonomic resolution was observed. For those stations which are in more degraded condition, there was generally a lower magnitude of increase of taxa when identifications were made to the genus/species level. This may illustrate potential partial redundancy with some other metrics (e.g., percent contribution of dominant taxon, HBI, Hydropsychidae/Total Trichoptera). That is, when examining a benthic community at a degraded site, there is often a dominance by few taxa, sometimes one or two. In those cases, the one or two dominant taxa are usually ones with higher tolerance values (as in the Hilsenhoff scheme), thus translating into a higher HBI value (see Metric 2).

Metric 2. Hilsenhoff Biotic Index. Most stations showed little effect of taxonomic levels. However, the HBI is strongly reliant on tolerance values used in its calculation. In some cases, tolerance values were not available for the different taxa at either genus or family level since they are primarily developed for species. In general, however, the largest changes in calculated values were seen for the stations that were in the worst condition overall, with higher HBI values resulting from a more specific taxonomy (Figure 5-3).

Metric 3. Scrapers-Filterer Collectors Ratio. These metric values exhibited large changes when calculated on more specific taxonomic levels. At the family level of identification, the range of values was 0 to 66.7 (Table 5-3), whereas at the genus/species level it was 0 to 52.3 (Table 5-4). Interpretation of this metric is sensitive to two factors: (1) rarity of one of the two functional feeding groups in a sample and (2) increased uncertainty associated with assigning feeding

Table 5-3 Family-level metric values calculated from 100-organism subsamples. Bioassessment scores (in parentheses) are derived by comparing metric values to scoring criteria.

METRIC	CC1	CC2	CC3	HB1	HB2	OC4	OC1	OC2	OC3
1. Taxa richness	14(4)	16(6)	11(4)	9(4)	7(4)	15(6)	10(4)	9(2)	10(4)
2. HBI	4.8(0)	5.5(0)	5.2(0)	4.6(2)	6.3(0)	4.7(2)	6.3(0)	5.5(0)	6.8(0)
3. Scr/(Scr+Fc) x 100	36.6(6)	31.9(6)	22.6(4)	0(0)[a]	0(0)[b]	51.6(4)	25.0(2)	8.6(0)	66.7(6)
4. EPT/(EPT+Chir) x 100	75.9(6)	59.2(6)	88.2(6)	69.7(6)	0(0)	86.4(6)	30.5(2)	60.9(4)	3.6(0)
5. % Contr. Dom. Taxon	30.5(4)	34.7(2)	59.6(0)	70.1(0)	62.4(0)	21.3(4)	49.5(0)	43.1(0)	40.2(0)
6. EPT index	7(4)	5(2)	3(2)	3(2)	0(0)	8(4)	4(2)	1(0)	1(0)
7. (Shred/Tot) x 100	6.8(6)	5.5(6)	1.8(2)	0(0)[c]	0(0)[c]	1.1(4)	0(0)	0.8(4)	0(0)
8. (Hydro/Trich) x 100	92.3(0)	97.9(0)	100(0)	95.2(0)	0(6)[d]	62.5(2)	95.5(0)	100(0)	100(0)
9. Pinkham-Pearson index	UR(6)	6.4(6)	2.7(2)	2(2)	0.1(0)	RR(6)	4.6(6)	4.7(6)	1.0(0)
10. QSI Tax x 100	UR(6)	73(6)	64(6)	16(0)	1(0)	RR(6)	42(6)	46(6)	18(2)
11. DIC-5	UR(6)	3(4)	5(6)	2(2)	1(0)	RR(6)	4.0(6)	4.0(6)	2(2)
12. QSI-FFG x 100	UR(6)	85.6(6)	73.7(6)	49.7(4)	27.6(2)	RR(6)	53.7(6)	66.6(6)	31.1(2)
Total (with paired) metrics	50	50	38	22	12	56	34	34	16
Biology (with paired) % comparison to reference	--	100	76	44	24	--	61	61	28
Total (without paired) metrics	26	28	18	14	10	32	10	10	10
Biology (without paired) % comparison to reference	--	108	69	54	20	--	31	31	31
Habitat Score	139	132	92	182	57	191	86	114	118
Habitat % comparison to reference	--	95	66	131	41	--	45	60	62

UR = Upstream Reference; RR = Regional Reference; CC1 also served as reference for Harbor Brook, see page 4-11 for further discussion.

[a] No scrapers

[b] No scrapers or filterer-collectors

[c] No shredders

[d] No Trichoptera

designations, which are usually assigned to species, to higher taxonomic levels. The is because (1) many invertebrate taxa are poorly known and (2) some taxa are known to shift feeding behavior upon entering subsequent developmental life stages.

Metric 4. EPT-Chironomidae Ratio. There is no effect on this metric since it is based on the number of individuals in these taxonomic groups (family and order, not genus/species).

Metric 5. Percent Contribution of Dominant Taxon. When taxonomic groups are split (as accomplished by more specific taxonomy), there are fewer individuals representative of each of the subgroups and an overall lower contribution to sample composition. In sites considered to be in better condition, values for this metric would thus be expected to substantially decrease with more specific levels of taxonomy. However, this expectation was not consistent with some of the results (Table 5-4, Figure 5-4). Station OC4, the regional reference

Table 5-4 Genus/species-level metric values calculated from 100-organism subsamples. Bioassessment scores (in parentheses) are derived by comparing metric values to scoring criteria.

METRIC	CC1	CC2	CC3	HB1	HB2	OC4	OC1	OC2	OC3
1. Taxa richness	24(4)	31(6)	17(2)	13(2)	11(2)	26(6)	28(6)	19(4)	16(4)
2. HBI	4.4(2)	5.2(2)	5.0(2)	5.9(2)	8.5(0)	4.4(2)	6.0(0)	5.6(0)	7.0(0)
3. Scr/(Scr+Fc) x 100	36.7(6)	21.7(4)	16.7(2)	0(0)	0(0)[b]	52.3(6)	3.1(0)	0(0)	0(0)
4. EPT/(EPT+Chir) x 100	75.9(6)	59.2(4)	88.2(6)	69.7(6)	0(0)	86.4(6)	30.5(2)	60.9(4)	3.6(0)
5. % Contr. Dom. Taxon	27.5(4)	13.1(6)	25.7(4)	70.1(0)	58.1(0)	20.2(0)	12.4(4)	17.9(0)	22(2)
6. EPT index	9(6)	8(4)	6(4)	5(2)	0(0)	13(6)	6(2)	4(2)	1(0)
7. (Shred/Tot) x 100	9.3(6)	9.1(6)	5.5(4)	5.8(4)	0.9(0)	4.3(0)	9.5(2)	13(4)	19.7(6)
8. (Hydro/Trich) x 100	92.3(0)	97.9(0)	100(0)	95.2(0)	(0)(6)[a]	62.5(2)	95.5(0)	100(0)	100(0)
9. Pinkham-Pearson index	UR(6)	7.8(6)	3.3(2)	2.0(2)	0.1(0)	RR(6)	5.1(4)	8.3(6)	0.7(0)
10. QSI Tax x 100	UR(6)	46(6)	55(6)	16(2)	1(0)	RR(6)	22(4)	34(6)	10(2)
11. DIC-5	UR(6)	2(4)	2(4)	2(4)	0(0)	RR(6)	0(0)	1(2)	0(0)
12. QSI-FFG x 100	UR(6)	82.2(6)	72.9(6)	49.7(4)	27.6(2)	RR(6)	60.3(6)	62.8(6)	30.6(4)
Total (with paired) metrics	58	54	42	28	10	52	30	34	18
Biology (with paired) % comparison to reference	48	93	76	--	17	--	58	65	35
Total (without paired) metrics	34	32	24	16	8	28	16	14	12
Biology (without paired) % comparison to reference	--	94	71	47	23	--	57	50	43
Habitat score	139	132	92	182	57	191	86	114	118
Habitat % comparison to reference	--	95	66	131	41	--	45	60	62

UR = Upstream Reference; RR = Regional Reference; CC1 also served as reference for Harbor Brook, see page 4-11 for further discussion

[a] No scrapers

[b] No scrapers or filterer-collectors

site on the West Branch Tioughnioga River, only changed from 21 percent (family-level) to 20 percent (genus/species-level). Conversely, the farthest downstream station on Canastota Creek, which exhibited moderate impairment, decreased from 60 to 27 percent.

Metric 6. EPT Index. Because this metric is a restricted form of taxonomic richness, a similar general response to level of identification is expected. Small increases in this value are seen with genus/species-level taxonomy (Tables 5-3, 5-4).

Metric 7. Shredders/Total No. Individuals. There is minimal effect on this metric except where families are not designated as shredders and genera or species are designated.

Metric 8. Hydropsychidae/Total Trichoptera. There is no effect on this metric since it is based on the number of individuals in these two taxonomic groups only.

Metric 9. Pinkham-Pearson Community Similarity Index. The effect of taxonomic level on this metric was minimal (Figure 5-5). Values ranged from 0.1 to 6.4 for family-level identifications and from 0.1 to 8.3 for genus/species-level identification. The middle station on Onondaga Creek (OC2) had a value shift from 4.7 to 8.3, the largest change by far.

Metric 10. Quantitative Similarity Index-Taxa. The effect of more specific taxonomy was minimal, as indicated by a high correlation of rank orders (Spearman rank correlation,

R=0.93, p=0.002) between the two treatments. The largest difference in values was observed at Station OC1 with a family-level value of 42 and a genus/species-level of 22.

Metric 11. Dominants-in-Common-5. A minimal range of possible values for this metric makes it difficult to interpret. An example of unpredictable changes in this metric is illustrated at Stations OC1 and OC3, where the DIC value fell from 4 to 0 and 2 to 0, respectively, when the calculation was done at the generic level. At both stations, there were dominant, family-level taxa in common but they were represented by different genera, thus accounting for the lower DICs. When subjected to the two treatments, there was a relatively low correlation of rank orders (Spearman R=0.35, p=0.44); therefore, taxonomic treatments could lead to different comparisons between stations for this metric.

Metric 12. Quantitative Similarity Index-Functional Feeding Group. There were only minor changes in values when calculated at family versus genus/species level. Any differences were probably due to differential availability of functional feeding group designations among the taxonomic levels. However, rank order correlations showed no difference with a Spearman rank correlation R of 1.0.

5.2.2 Subsampling Level Effects on Metric Performance

RBPs provide a mechanism for substantially reducing the level of effort through randomized subsampling. The comparisons presented here illustrate the behavior of identical metrics when calculated on differential subsampling intensities. Using metric values calculated at the taxonomic level of genus/species, the effect of subsample size on metric performance was evaluated. Comparisons of RBPIII with subsampling at the 100-organism (Table 5-4) and 300-organism (Table 4-3) levels were done through a combination of correlational scatterplots and confirmation of differences with Spearman rank correlations.

A previous unpublished study (Stribling and Gerardi 1993 [draft report]) has shown that two metrics are strongly biased by different organism counts, taxa richness and EPT index, showing a marked increase with higher numbers of individuals. However, two factors diminish the importance of these biases. First, the relationship is a predictable one; second, metrics used in RBP site assessments are evaluated based on their value relative to reference conditions rather than on absolute numbers. Thus, if data representing reference sites or conditions are collected in the same manner, these biases become essentially irrelevant. The following analyses provide further confirmation of these conclusions, including those concerning minimal effects on the other metrics.

Total Bioassessment Score. Overall bioassessment score is not affected by differential subsample sizes (Figure 5-6); rank order correlation is perfect (R=1.00).

Metric 1 Taxa Richness. This metric had a value range of 8 to 31 taxa at the 100-organism subsample and 16 to 41 at the 300-organism subsample (Figure 5-7). Number of taxa increases significantly as larger samples are analyzed, but correlation of rank orders is nearly perfect (Spearman R=0.95, p=0.000066). Therefore, a larger sample size would not affect comparisons between stations when using this metric.

Metric 2 Hilsenhoff Biotic Index. Subsampling level had no effect on the HBI values with a nearly 1:1 correlation (Spearman R=0.99, p=0.00) between the two treatments.

Metric 3 Scraper-Filterer Collector Ratio. Although somewhat more variable, rank orders show significant correlation for the subsample size (Spearman R=0.93, p=0.0003) (Figure 5-8). Therefore, subsample size had no effect on station comparisons using this metric. No scrapers were selected in the 100-organism subsample at HB1, which caused the metric to have a value of 0; one scraper was selected in the 300-organism subsample giving a value of 16.7.

Metric 4 EPT-Chironomidae Ratio. Subsample size had no effect on the results calculated from this metric (Spearman R=0.92, p=0.0005).

Metric 5 Percent Contribution of Dominant Taxon. Subsample size had no effect on the values calculated for this metric (Spearman R=1.0).

Metric 6 EPT Index. As seen for taxa richness (Metric 1), a difference was detected for this richness metric, but there was no difference in rank orders (Spearman R=0.98, p=0.000002) of the samples. The number of EPT taxa increases as larger samples are taken, especially at less degraded sites, due to the sensitivity of the species.

Metric 7 No. Shredders/Total Sample. Similarly to the Scraper-Filterer Collector Ratio, this functional feeding group metric appears more variable, but differences in rank orders are nonsignificant (Spearman R=0.97, p=0.00002) (Figure 5-9). Different subsample sizes have no effect on interpretations using this metric. By chance, we got a higher percentage of shredders in the 100-organism subsample (19.7 versus 15.8 for the 300-organism subsample).

Metric 8 Hydropsychidae/Total Trichoptera. This metric is not significantly affected by different subsample sizes (Spearman R=0.97, p=0.000014).

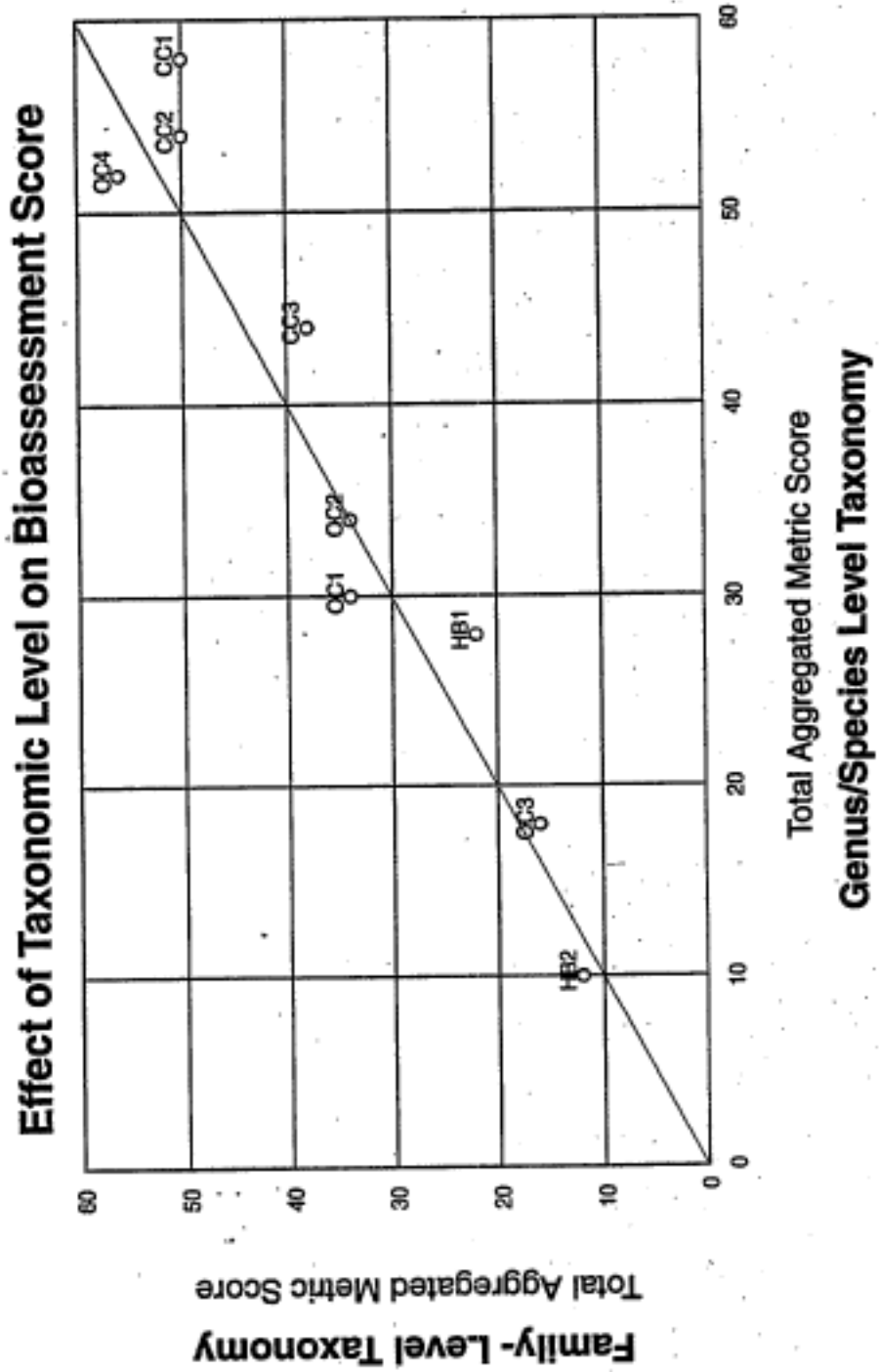

Figure 5-1. Correlational scatterplot (1:1) total bioassessment score, family vs. genus/species-level taxonomy.

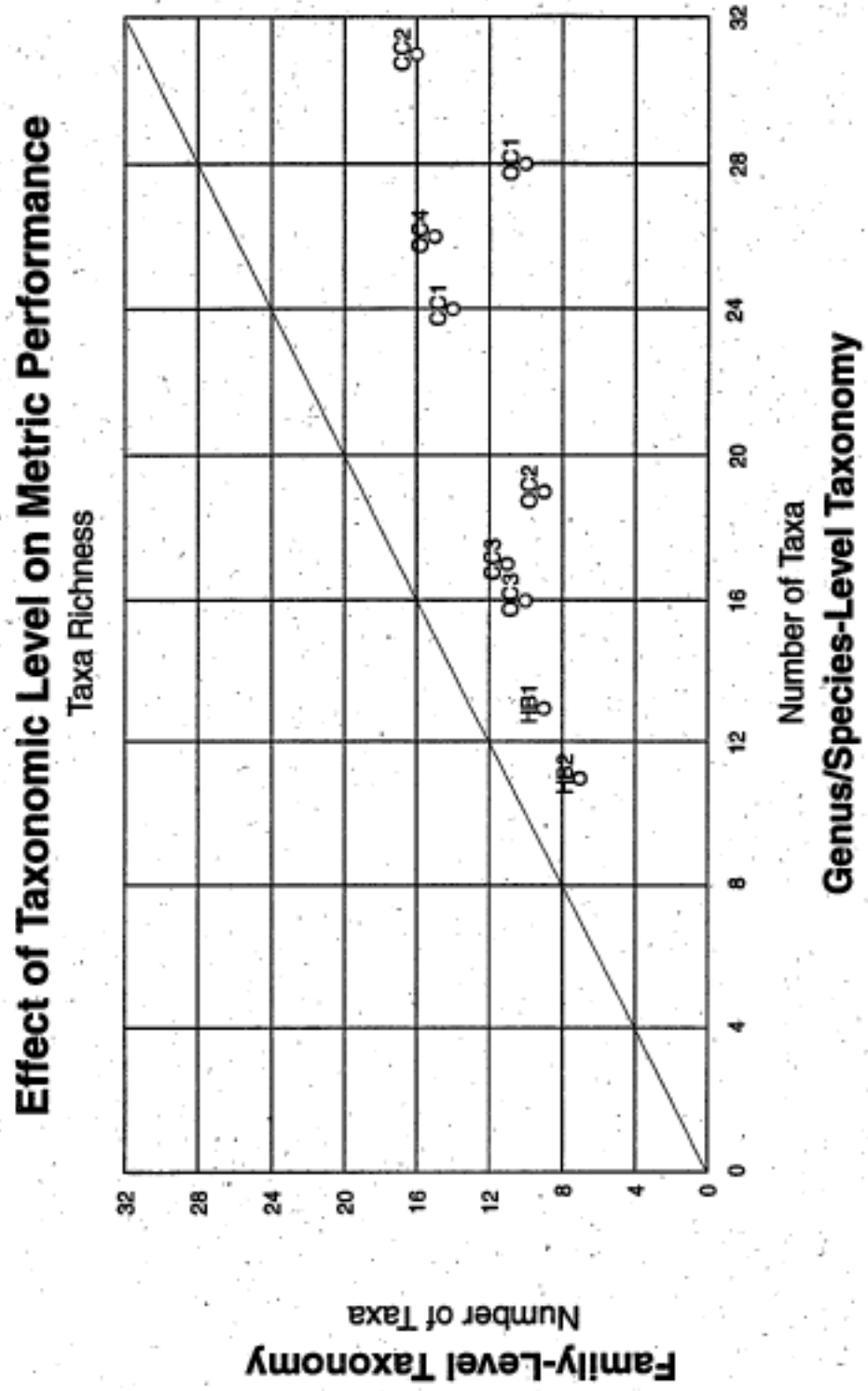

Figure 5-2. Correlational scatterplot (1:1) of taxa richness, family vs. genus/species-level taxonomy.

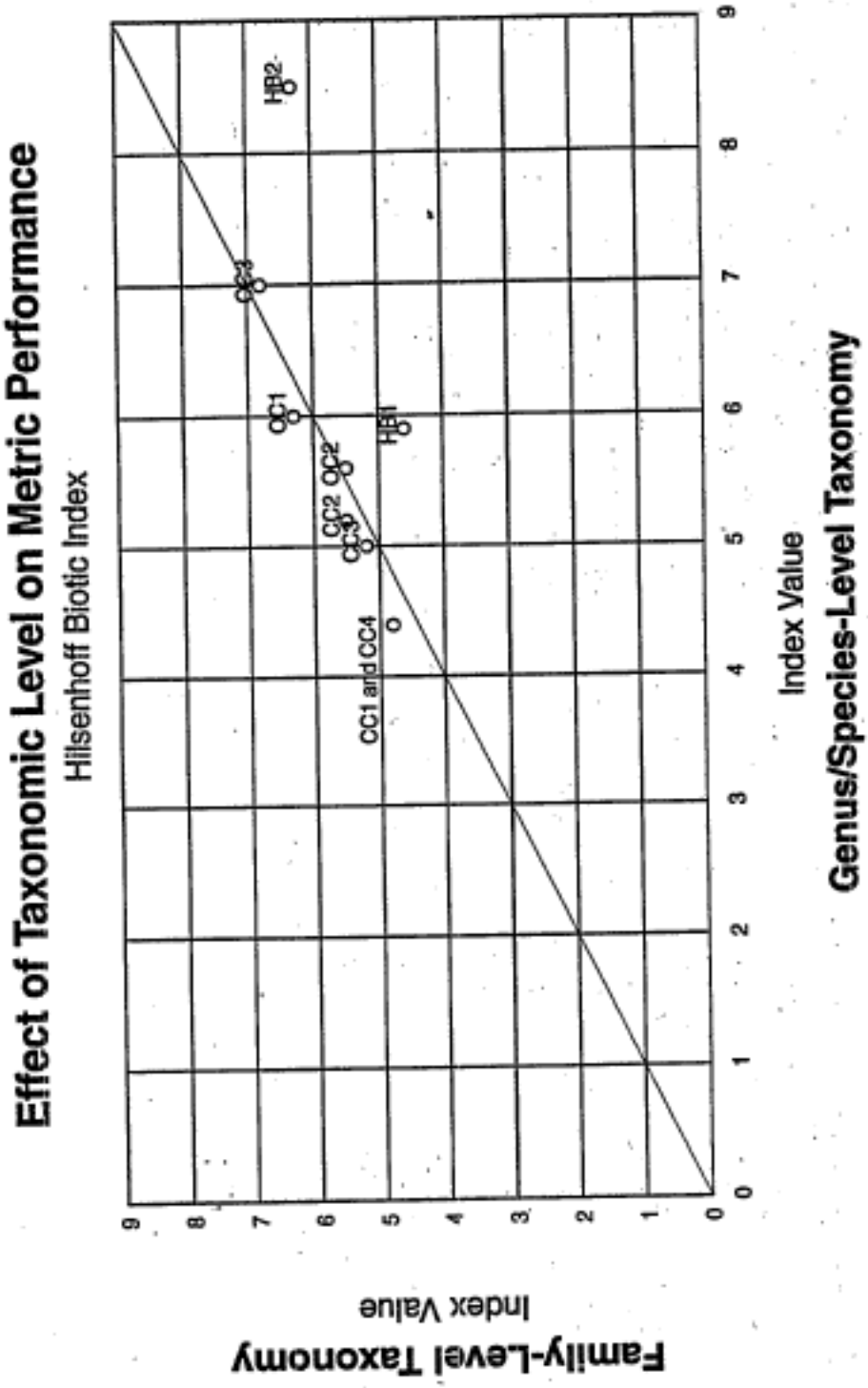

Figure 5-3. Correlational scatterplot (1:1) of the Hilsenhoff Biotic (HBI) values, family vs. genus/species-level taxonomy. Note: Stations CC1 and OC4 share the same coordinates.

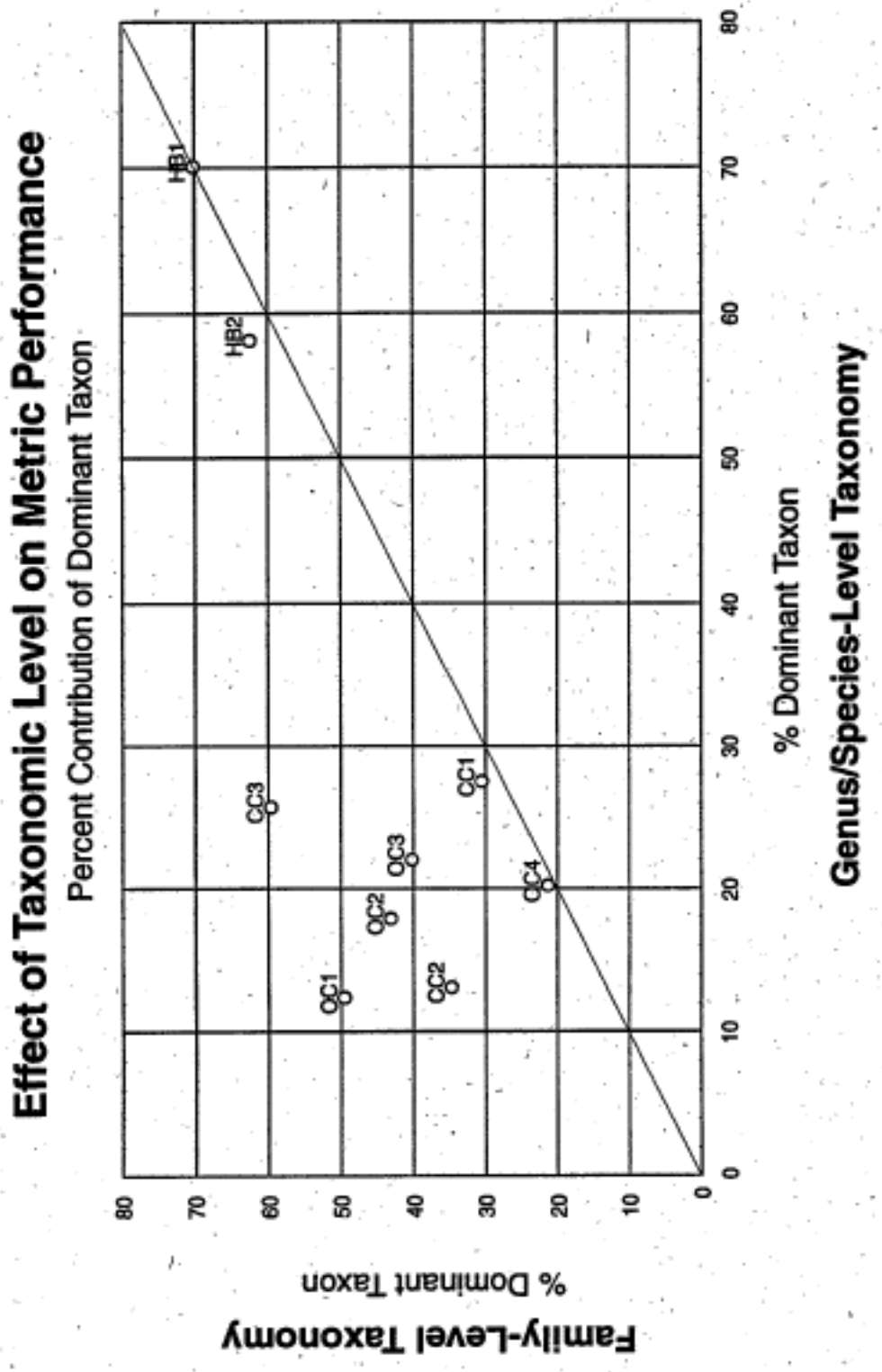

Figure 5-4. Correlational scatterplot (1:1) of percent contribution of dominant taxon, family vs. genus/species-level taxonomy.

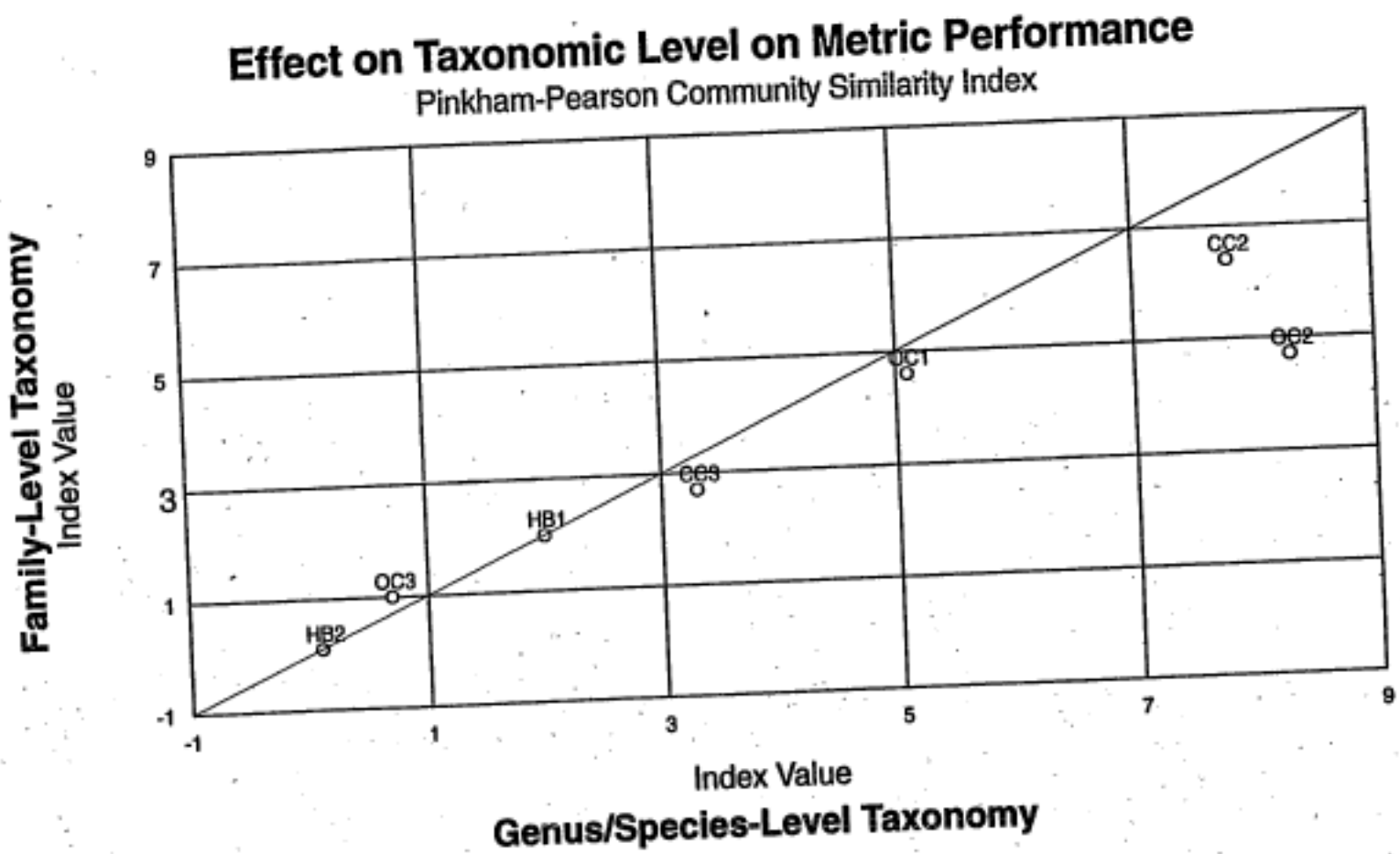

Figure 5-5. Correlational scatterplot (1:1) of Pinkham-Pearson Community Index, family vs. genus/species-level taxonomy.

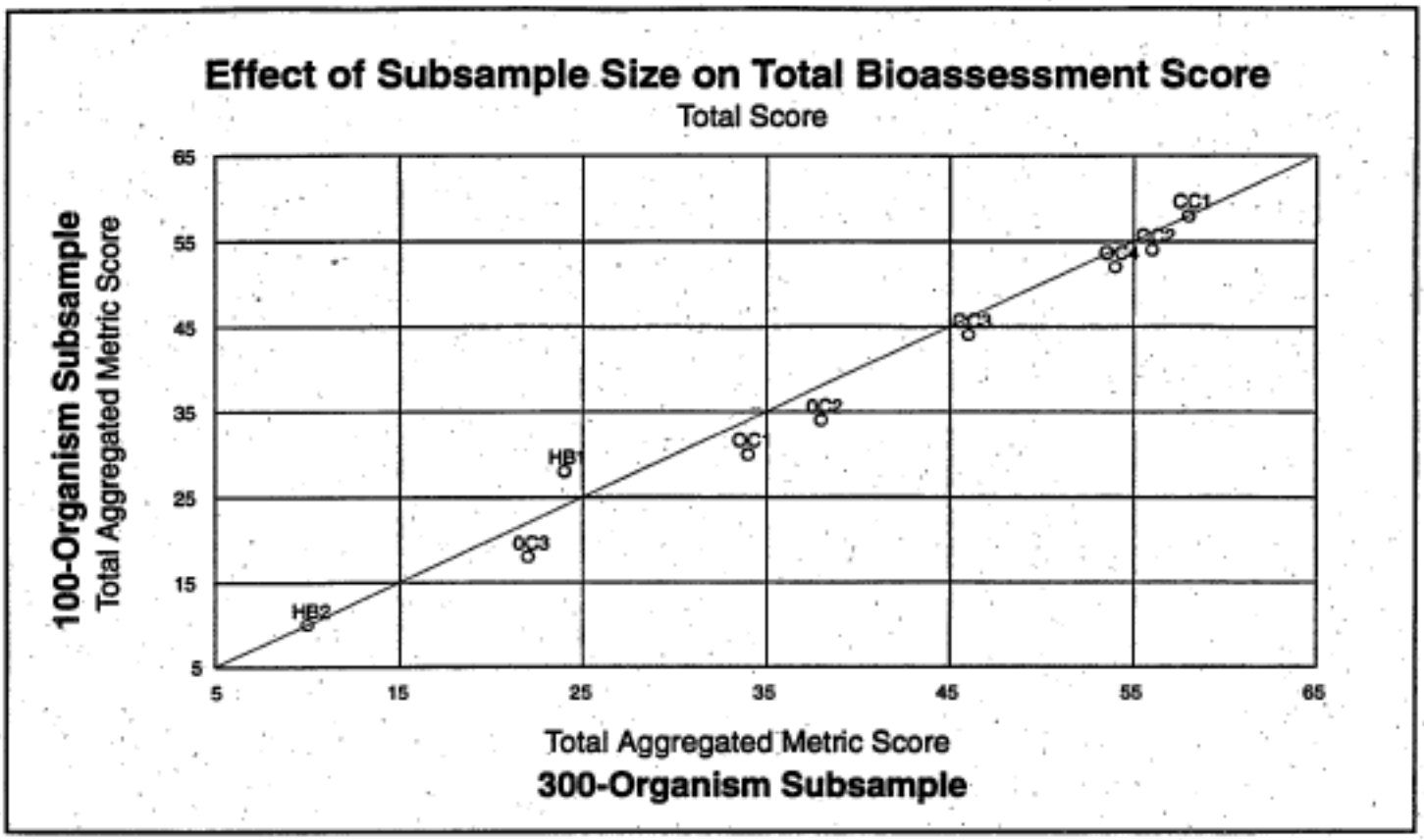

Figure 5-6. Correlational scatterplot (1:1) of bioassessment score, 100 vs. 300 organism subsample.

Metric 9 Pinkham-Pearson Community Similarity Index. This metric appears to be more variable with differential subsample size, but differences are nonsignificant in comparison of rank orders (Spearman R=0.92, p=0.0025).

Metric 10 Quantitative Similarity Index-Taxa. Different subsample sizes have little effect on this metric and differences in values are shown to be nonsignificant (Spearman R=0.93, p=0.0025).

Metric 11 Dominants-in-Common-5. This metric does not seem to be affected by subsample size, but similar to the analysis of differential taxonomic levels, correlated variation is difficult to determine due to a narrow range of possible values. Because it is common to have several ties in a small data set such as this, ordinal analyses such as Spearman rank correlation can have diminished meaning. Station CC1 was used as the upstream reference; the other stations for which this was used as a baseline had no changes in value (HB1 and HB2) or changed by only one (CC2 and CC3). For the OC4-compared stations, there were no changes in metric values with higher levels of subsampling.

Metric 12 Quantitative Similarity Index-Functional Feeding Group. This metric is not affected by subsample size; there is a perfect rank order correlation (Spearman R=0.96, p=0.0004).

5.3 Summary of Results

These comparisons have shown that there are some effects on metric behavior when subjected to different treatments. For taxonomic level, five metrics (taxa richness, HBI, scraper-filterer collector ratio, percent contribution of dominant taxon, and shredders-to-total ratio) were found to be substantially different; for another seven, there was either perfect 1:1 correlation or nearly perfect. For the different subsampling levels, only two metrics performed differently between higher and lower levels of organisms: taxa richness and EPT index. For both sets of treatments, total bioassessment scores were not affected, with essentially perfect agreement between them. Refer to Section 6.2 for further discussion.

The screening-level assessment (RBPI) proved to be a useful tool for identifying sites with biological impairment. One site was screened as minimally impaired and was further assessed, using RBPIII, as having severe impairment.

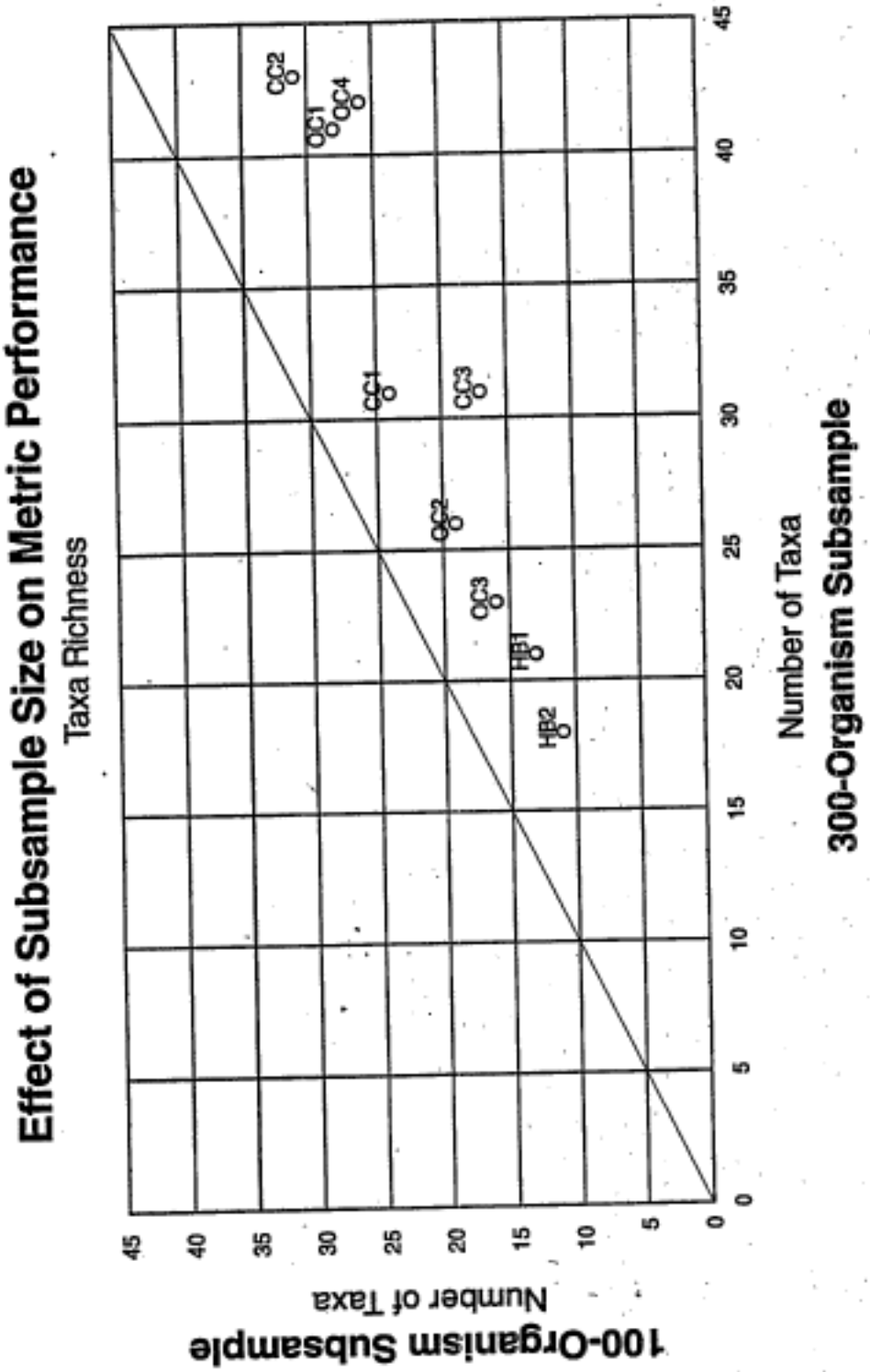

Figure 5-7. Correlational scatterplot (1:1) of taxa richness, 100 vs. 300 organism subsample.

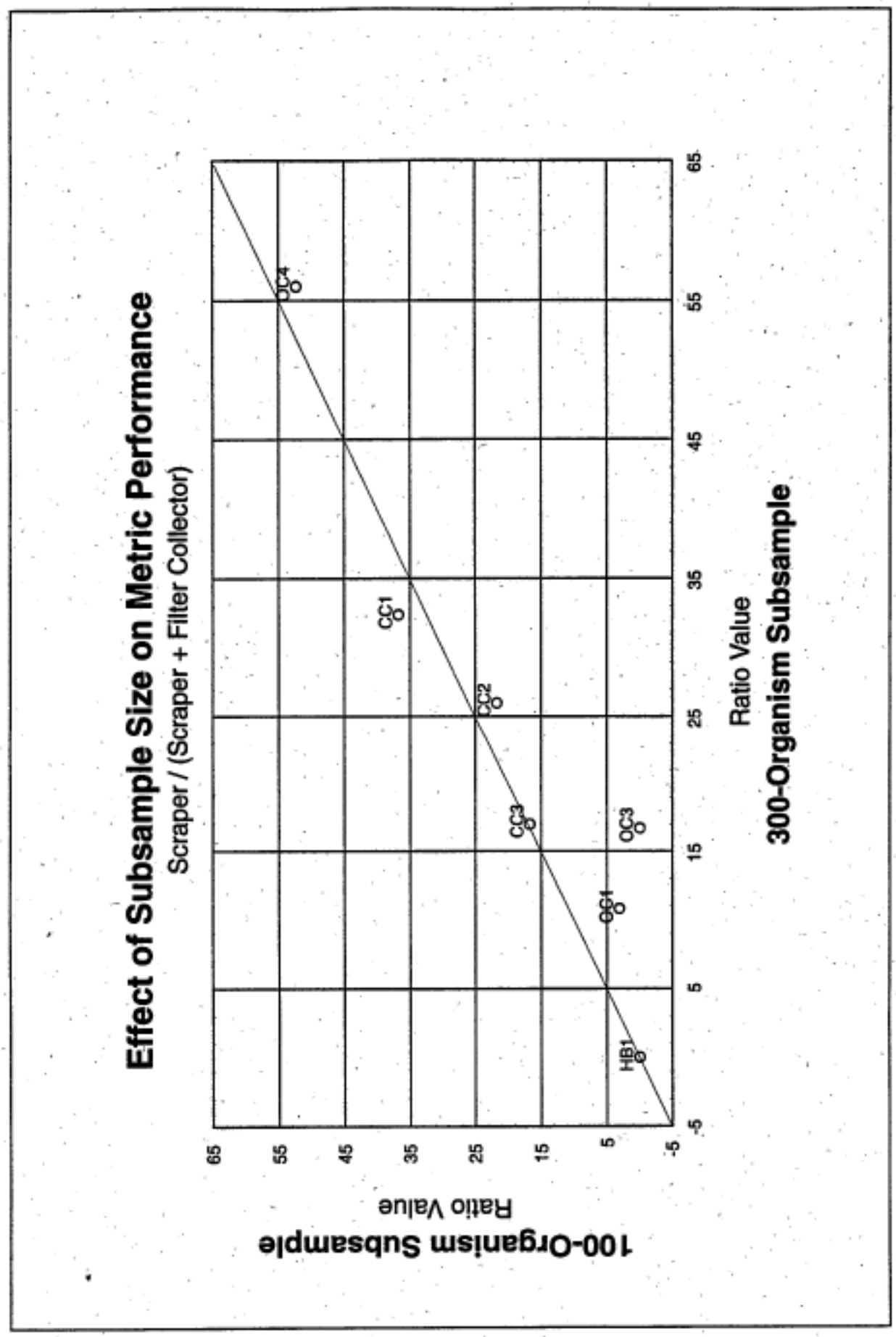

Figure 5-8. Correlational scatterplot (1:1) of Scraper/(Scraper + Filter Collector, 100 vs. 300 organism subsample.

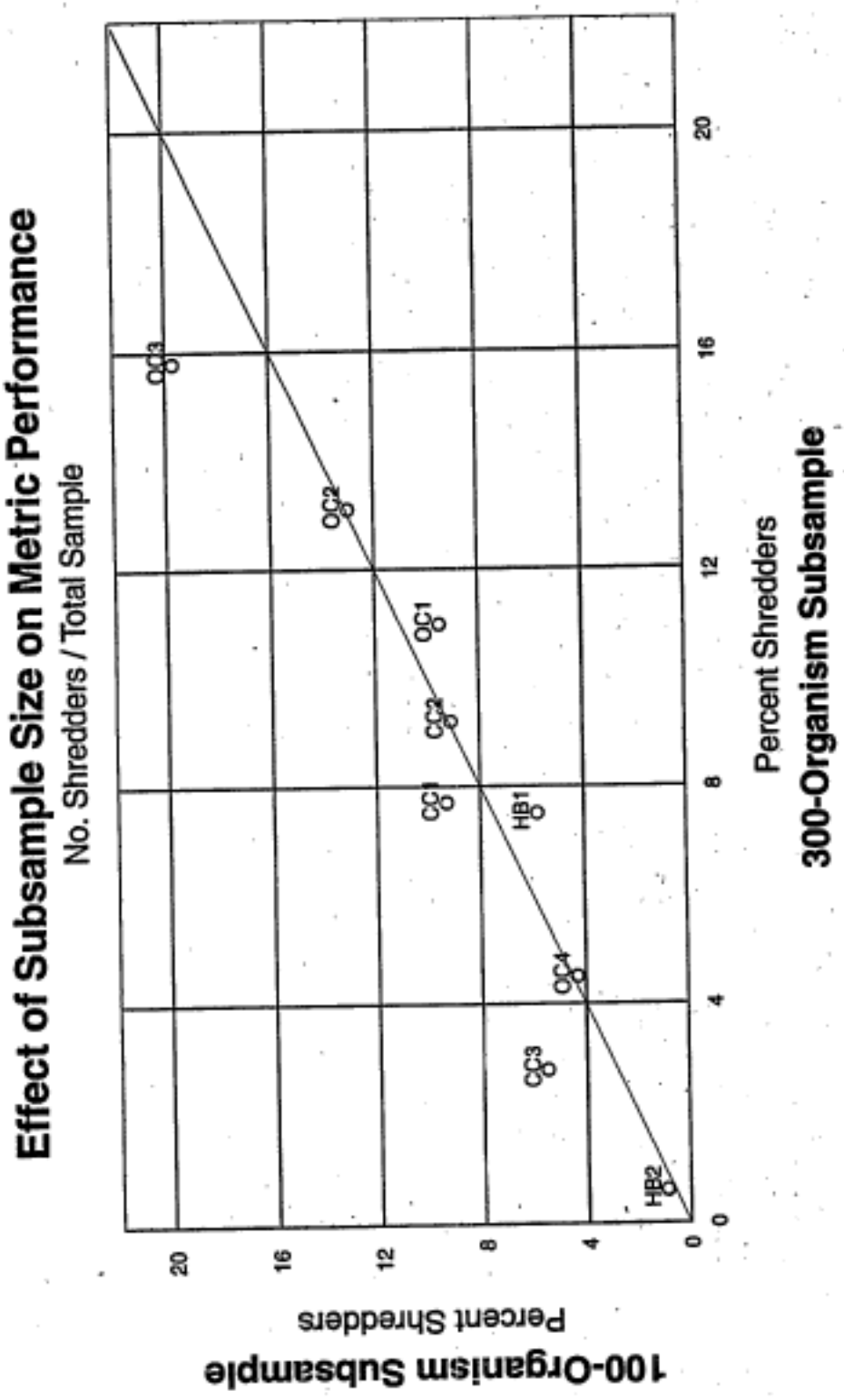

Figure 5-9. Correlational scatterplot (1:1) of No. Shredders/Total Sample, 100 vs. 300 organism subsample.

6

Conclusions/Recommendations

6.1 Historical Assessment Comparisons

Comparisons presented in this document are of three types:

- RBP results with different types of historical data: Hester-Dendy multiplate samplers (Ohio) and the traveling kick net (New York).
- RBP sampling with variation of taxonomic level (New York).
- RBP sampling with variation of subsample size (New York).

A comparison of results suggested a reasonably good fit between Ohio EPA findings and those of the present study. Subtle discrepancies between the data sets are most likely a result of the lack of regional calibration for the RBP analysis technique; that is, there is not a complete understanding of which benthic metrics are most appropriate for the upper Midwest when using kick nets. This might have weakened the interpretive power of the approach. Also, there is likely some effect of the different sampling methodologies (Hester-Dendy multiplate samplers and square-meter kick nets) on the assessments. It is difficult to determine if these more subtle differences are due to differences in methods or changes in biological condition over time. Bioassessment, as exemplified by the Ohio EPA ICI (for macroinvertebrates) and IBI (for fish) and the EPA RBP (for macroinvertebrates), is a valid and technically sound tool for evaluating impaired waters, particularly when calibrated on a regional level as is done for the ICI and IBI. This validation is supported by similar assessments being arrived at by approaches differing in detail (this study).

For the New York portion of the study, all assessments compared favorably with those most recently performed by the DEC (Bode et al. 1993). In 1990 sampling, Canastota Creek was found to be "moderately impacted" at a single station downstream of the town. At three stations along its length, we assessed it as "slightly to moderately impaired" and "moderately impaired."

A downstream station on Onondaga Creek was assessed as "severely impacted" in both 1989 and 1990 sampling efforts (Bode et al. 1993). Our assessments showed this creek to be "moderately impaired" in upstream reaches and "severely impaired" near the same station assessed by DEC. Harbor Brook was assessed similarly between DEC in 1989 (Bode et al. 1993) and here as "moderately" to "severely impaired."

Traditional comparisons of biological assessment methods occur through side-by-side sampling and analysis. These temporally separate data have provided some useful insights into the process of bioassessment comparisons. As mentioned above, differences in results might arise directly from sampling biases inherent in the sampling gear. This might be a problem when attempting to directly compare data from separate bioassessment samples (e.g., the number of species, the calculated value of an individual metric or the number of individual organisms collected). The problem of sampling error (bias) is reduced if comparisons are made at the level of the overall assessment score rather than individual metrics.

6.2 Statistical Comparisons

Comparisons were made between RBPII (family-level identifications) and RBPIII (lowest-practical-level identifications, usually genus/species), as well as subsample size (100-organism versus 300-organism). As long as the reference conditions are treated in the same manner as test station data (taxonomic and subsampling levels), comparisons between assessment results are valid. We found that although there might have been some differences in specific metric performance (i.e., metric values) with different treatments, those values relative to reference

conditions varied little. Further, there was perfect agreement among total bioassessment scores between the treatments.

6.2.1 Taxonomic Level Conclusions

When addressing the question of appropriate taxonomic level, different concerns do arise. Although similar site rankings based on condition might be found with different levels, there can be difficulty in interpretation of potential causative factors when using more gross-level identifications. This is especially true when dealing with metrics dependent on how individual species adapt to the environment rather than how they relate to other species. These metrics include the HBI and those related to functional feeding groups (scrapers, filterer collectors, shredders). The tolerance values on which the HBI is based are usually assigned to species (or genus) level and might not be available for family. Likewise, functional feeding group designations become more uncertain as they are assigned to more general (or higher) taxonomic levels. It is recommended that, in general, taxonomy be performed to the lowest practical level that will suit the objectives of the study, which will usually be the genus or species level for biological assessments beyond the screening level. The decision on taxonomic level might also be refined with regional calibration of bioassessment techniques.

6.2.2 Subsample Size Conclusions

The argument can be made that a lower number of organisms does not allow a reasonable estimate of biological diversity. However, as was shown with the taxa richness metric, as higher numbers of organisms are included in a sample, the higher the number of detected taxa will be. This is due to an increase in the probability of rare taxa being included within a larger subsample. In essence, rare taxa have little influence on biological assessments using a multimetric approach because even if rare taxa are collected, their contribution to a multimetric index is minimal. Conversely, if one's goal is to describe biological diversity at a site, even an analysis of the total sample (versus a subsample) is likely inadequate. It is possible to collect continuously larger samples from a broader diversity of microhabitats within a site and continue to get additional taxa. The critical factors are to have consistency in sampling effort and a properly randomized subsampling procedure. As with other sample treatments, subsampling is appropriate as long as samples from reference sites are treated in the same way; subsamples less than 100-organisms are not recommended. The recommendation is to base benthic macroinvertebrate biological assessments on 100-organism subsample when using RBPs in New York.

6.3 Usefulness of RBPs in Assessing CSO Biotic Effects

Attributing cause and effect to the specific CSO activity is complicated by other related problems associated with urbanization, e.g., habitat alteration and industrial discharges. However, the bioassessment procedures, with its integration of total scores, individual metrics (which are based on known ecology of the benthic community) and habitat description, provide reasonable technical support for identifying potential sources of biological impairment. An impairment due to CSO outfalls was noted in biological data collected by both Ohio EPA and the present study for a 15- to 20-mile reach of the Scioto River, a 4-mile reach of the Sandusky River, and a 10-mile reach of the Little Cuyahoga River. In the cases of the Scioto and Little Cuyahoga Rivers, upstream stations also located in urbanized areas had relatively healthy biological communities and were effective for comparisons of biological data. The unimpaired middle station of the Little Cuyahoga River exhibited recovery of the biota since the correction of upstream CSOs. The assessments were performed prior to our gaining information concerning the outfalls.

For the New York study, severe habitat degradation and alterations were evident at all Onondaga Creek sites and at the two downstream sites on Harbor Brook. There were many instances of major habitat differences between stations on the same stream or between a station and its regional reference site. However, even with these differences, impairment due to stressors commonly produced by CSOs was seen at the middle and lower stations on Canastota Creek and Onondaga Creek.

Results indicated that CSO outfalls had an adverse impact on the downstream macroinvertebrate assemblages. Impairment of the benthic biota, in both the Ohio and New York studies, was manifested by the metrics (1) taxa richness, (2) scraper/scraper + filterer collector, (3) EPT/EPT + Chironomidae, (4) percent contribution of dominant taxon, (5) Hydropsychidae/total Trichoptera, (6) Pinkham-Pearson Community Similarity Index, (7) QSI-taxa, and (8) DIC-5.

The bioassessments were instrumental in identifying impaired reaches of each river at periods that reflected residual and cumulative effects of CSO outfalls that were not necessarily actively discharging. Sampling was performed during normal flow conditions (i.e., not during the wet or dry season) although several of the Ohio sampling locations were being affected by increased flow levels. Results illustrate the utility of biological data for capturing the effects of intermittent discharge events without sample collection during stormflows.

The use of multiple metrics aids in achieving more accurate assessments than single-parameter assessments. This was seen in the case of HB1 and HB2, which had nearly identical metric values for taxa richness but very different overall biological assessments (HB1-moderate, HB2-severe). The multimetric approach uses the total assessment score for comparison to the reference in determining the biological integrity at a site and uses individual metrics for interpreting the assessment and gaining insight as to cause-and-effect relationships. The associated habitat assessment enabled a characterization of the physical habitat alteration, strengthening the ability to identify additional potential sources of impairment. For example, the nonimpaired biological condition assessment in the presence of degraded habitat on the Sandusky River (SA2) is a likely indication of some form of nutrient enrichment since, as discussed earlier (Section 3.2.3.4), the initial phases of nutrient enrichment cause an increase in the biota. If the nutrient enrichment is mild to moderate, the biological community balances between the effects of enhanced biota and the next phase of enrichment, oxygen depletion. In such instances, the biology would continue to score higher than the surrounding habitat would be expected to support.

6.4 The Place of Bioassessment In Watershed Protection

Another potential application for bioassessments is within the total maximum daily load (TMDL) process, which is one of the essential tools of the watershed protection approach. The watershed protection approach attempts to evaluate watersheds on a holistic, rather than piecemeal, basis. A TMDL is defined by USEPA guidance and regulations as being equivalent to the loading capacity of a waterbody and the sum of the individual wasteload allocations (WLAs) for point sources, load allocations (LAs) for nonpoint sources, and natural background sources, and a margin of safety to account for uncertainties about the relationship among stressors, controls, and the quality of the receiving water (USEPA 1994b).

TMDLs are required when states determine that technology-based controls will not result in a waterbody's meeting water quality standards, including its designated uses. The TMDL process can provide sufficient and necessary information for making decisions on the implementation of appropriate pollution reduction tools such as best management practices (BMPs), ecological restoration, or engineered active or passive treatment technologies (USEPA 1994c).

Although TMDLs until now have been primarily chemical-specific, biological assessment shows promise as a tool for going beyond chemical water quality to biological endpoints and the aquatic life uses of the waterbody. Biological assessments provide a direct evaluation of ecosystem condition by integrating physical habitat quality with biological condition. The evaluation is accomplished by comparison to empirically-defined, regionalized expectations of biological conditions (reference conditions). As was demonstrated in these case studies, bioassessments can often detect the biological impact of CSOs and other intermittent discharges in urbanized watersheds affected by multiple stressors. Because CSOs contribute to the pollution load entering a waterbody, they must be considered in TMDL development. Biological assessment used in the TMDL process can help:

- Identify waters that are ecologically impaired and might be in nonattainment of chemical water quality standards; this would help in the siting and installation of appropriate controls.
- Prioritize and target ecologically impaired waters.
- Aid in the development and implementation of TMDLs for nonchemical stressors within a watershed.
- Assess the effectiveness of installed pollution control tools in protecting aquatic resources.
- Where the metrics for a region have been sufficiently refined, the diagnostic capabilities of some metrics might allow some conclusions to be drawn with regard to specific causes of biological impairment in a waterbody.

Other current USEPA programs that can benefit from the use of biological assessments include 1994 CSO Control Policy (section 1.1) , stormwater and wet-weather monitoring, 305(b) reporting, and biological criteria. Many states have incorporated biological assessments into their 305(b) reports, and many are currently developing biological criteria for waterbodies in their ecoregions. As illustrated in this report, biological assessments are useful for determining impairments from episodic events such as those accompanied by wet weather and stormwater without the necessity of sampling during the actual event.

7

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Appendix A
Taxonomic List of Benthic Macroinvertebrates Collected in Ohio, September 1992

Appendix A. Taxonomic list of benthic macroinvertebrates collected in Ohio; September, 1992.

						HBI	Feeding	Station Locations										
Phylum	Class	Order	Family	Genus	Species	Tol	Group	CR1	CR2	CR3	SA4	SA1	SA2	SA3	S1	S2	S3	S4
Cnidaria	Hydrozoa					5	PRE											
Turbellaria						6	PRE	1					1			1		
Nematoda						5	CG	2								2		
Annelida	Oligochaeta	Lumbriculida	Lumbriculidae			8	CG		2	1								
		Tubificida	Tubificidae			10	COL	1	3	1	1		21	14				
				Branchiura		10	COL				2					2	4	1
			Naididae			7	COL	2	3	1				2				
		Pharyngodellida	Erpobdellidae			8	PRE	2	1									
		Branchiobdellida	Branchiobdellidae			6	COL		7									
Mollusca	Gastropoda	Mesogastropoda	Pleuroceridae			6	SCR									15		
				Elimia		6	SCR								1			
		Basommatophora	Ancylidae	Laevapex		6	SCR						1					
			Physidae			8	SCR											
	Bivalvia	Veneroida	Sphaeriidae	Sphaerium		8	FIL						10	1	8		1	
Arthropoda	Crustacea	Hydracarina				6	PRE	1	3	1						1	3	
		Cyclopoda	Cyclopoida			?	?											
			Calanoida			?	?	4										
		Cladocera	Daphnidae	Daphnia		?	?	6680	93	19						2		
				Ceriodaphnia		?	?											
			Sydidae	Sida		?	?											
			Chydoridae			?	?											
			Bosminidae			?	?			1								
		Decapoda	Cambaridae	Orconectes		6	SHR		1			1	3	5	1			
			Hyalellidae	Hyalella	H. azteca	8	COL	7										
			Caenidae	Caenis		7	COL			1	2		2	35		1	6	
			Ephemeridae	Ephemera		2	COL				1							
			Heptageniidae			3	SCR											
				Leucrocuta		1	SCR				23	3	34	35			4	1
				Stenacron		4	SCR		3		25	12	8	103	8		16	1
				Stenonema		3	SCR				27		10	8	2	1	10	22
			Oligoneuriidae	Isonychia		2	COL				35	8	4		5		6	24
			Tricorythidae	Tricorythodes sp.		4	COL								2	4	10	36
		Trichoptera	Hydropsychidae			5	FIL	5				20			5	5	3	11
				Cheumatopsyche		5	FIL	26			95	282	216	13	94	82	107	69
				Hydropsyche sp A		5	FIL				29	13	29	7	100	4	8	29
				Hydropsyche sp B		5	FIL	221	1		1	63	110	20			4	23
				Potamyia sp.		5	FIL										2	8
				Macrostemum sp.		3	FIL											2
			Hydroptilidae	Hydroptila sp.		6	PCR						1			1	1	7
			Philopotamidae	Chimarra sp.		3	FIL				11				1			
		Odonata	Coenagrionidae			8	PRE	1	1									
				Argia sp.		6	PRE							3				

Appendix A. Taxonomic list of benthic macroinvertebrates collected in Ohio; September, 1992.

Phylum	Class	Order	Family	Genus	Species	HBI Tol	Feeding Group	Station Locations CR1	CR2	CR3	SA4	SA1	SA2	SA3	S1	S2	S3	S4
		Hemiptera				5	PRE											
		Coleoptera	Elmidae			5	SCR					1						
				Stenelmis		5	SCR	4			71	35	71	62	55	7	3	64
				Ancyronyx sp.		2	COL							1				
			Hydrophilidae			5	PRE					1						
			Psephenidae	Psephenus		4	SCR				28	4	2	3	1		1	1
			Scirtidae	Cyphon sp.		7	COL								1			
		Megaloptera	Sialidae	Sialis		4	PRE				6	1						
		Lepidoptera	Pyralidae	Petrophila sp.		5	SHR				1							
		Diptera	Empididae	Hemerodromia		6	PRE	1				1				2	1	1
			Simuliidae	Simulium		6	FIL	4		5	13	22	2	1	4	12	9	74
			Chironomidae	Tanypodinae		6	FIL		1	1	1	2	9	20		16	21	
				Orthocladiinae		5	COL		2	6		3	4	7	11	42	21	23
				Tanytarsini		6	COL						1	2	1	4		1
				Chironomini		6	COL	4	4	14	6	7	19	18	34	68	97	3
		*not included in taxa count		Chironomidae pupae				1	1				4	12		13	15	10
		*not included in taxa count		Diptera pupae									1		1		1	
		*not included in taxa count		Diptera adults					1		2	1			2			
		*not included in taxa count		Hydropyschidae pupae								1			13	4	3	
		Number of Individuals						8968	133	60	418	492	592	382	354	293	358	436

L=larvae
A=adults
a=non-benthic taxon (not included in metric calculations)
b=terrestrial taxon (not included in metric calculations)

Appendix B
Taxonomic List of Benthic Macroinvertebrates Collected in New York, September 1993

Appendix B. Taxonomic list of benthic macroinvertebrates collected in New York; September, 1993.

Phylum	Class	Order	Family	Genus	Species	HBI Tol	Feeding Group	Station Locations CHR4-300	OC4-300	OC1-300	OC2-300	OC3-300	CC1-300	CC2-300	CC3-300	HB1-300	HB2-300
Turbellaria				Dugesia?		6	PRE		1		2	31					5
Nematoda						5	PRE							5			
Annelida	Oligochaeta	Lumbriculida	Lumbriculidae			8	MIN	18	1						3		
			Enchytraeidae			8	COL					8					1
		Tubificida				10	COL										
				Limnodrilus		10	COL			24	3	15	1	23	12		12
				Potamothrix		5	COL					7		3			
				Tubifex		9	COL										2
			Naididae			7	COL										
				Dero		10	COL										1
				Nais	N. sp.A	8	COL	8	3	1		88					2
					N. bretscheri	6	COL		1				5	2	1		
				Ophidonais	O. serpentina	6	COL				1	74					
				Paranais		6	COL					12					
				Pristina		8	COL							1			1
Mollusca						6	SCR										
		Basommatophora	Ancylidae			7	SCR										
				Laevapex		6	SCR		7	2		1			2		
			Physidae			9	SCR										
				Physa		8	COL										3
Arthropoda	Crustacea	Hydracarina				6	PRE										
					Sp. A	6	PRE		21	5	12	28	18	2			
					Sp. B	6	PRE		1		2		1				
		Cyclopoda	Cyclopoida			?	?										13
		Isopoda	Asellidae			6	COL										
				Caecidotea		8	SHR				13	11		1	2	3	
		Amphipoda				7	COL										
			Crangonyctidae			4	COL										
				Crangonyx		4	COL										6
			Gammaridae			4	COL										
				Gammarus		6	COL	644		1	16	13		5	3	246	66
	Insecta	Ephemeroptera	Baetidae			5	COL										
				Baetis		5	COL	95	8				5	6	1	5	
			Caenidae			7	COL										
				Caenis		7	COL		7	10							
			Ephemerellidae			2	COL							2			
				Ephemerella		2	COL						3				
				Serratella		2	COL		13				17	1		1	
			Heptageniidae			4	SCR		2								
				Heptagenia		3	SCR		3								
				Stenonema		3	SCR		54								
			Leptophlebiidae			3	COL										
				Paraleptophlebia		1	COL		1	1			11		1		
		Plecoptera				1	SHR										
			Chloroperlidae			0	PRE										
				Sweltsa		0	PRE	56								1	
			Perlidae			2	PRE										
				Acroneuria		2	PRE				1						

Appendix B. Taxonomic list of benthic macroinvertebrates collected in New York; September, 1993.

Phylum	Class	Order	Family	Genus	Species	HBI Tol	Feeding Group	Station Locations CHR4-300	OC4-300	OC1-300	OC2-300	OC3-300	CC1-300	CC2-300	CC3-300	HB1-300	HB2-300
				Phasganophora		2	PRE							2	1		
			Taeniopterygidae			2	SHR						14	1	1		
		Trichoptera	Brachycentridae			1	FIL										
				Micrasema		2	SHR		11				7				
			Helicopsychidae			3	SCR										
				Helicopsyche		3	SCR		3								
			Hydropsychidae			5	FIL	1	25	3	20		14	40	43	18	
				Cheumatópsyche		5	FIL		31	6	8		23	75	64	3	
				Hydropsyche sp A		5	FIL		47	8	53	5	135	90	83	38	
				Hydropsyche sp B		5	FIL		10	27	50		2	20	3		
				Parapsyche		0	FIL	4									
			Hydroptilidae			4	PCR										
				Hydroptilidae pupae										1			
				Hydroptila sp.		6	PCR			1							
				Leucotrichia		6	SCR		1								
			Lepidostomatidae			1	SHR										
				Lepidostoma		1	SHR	1									
			Leptoceridae			4	COL										
				Oecetis		8	PRE		3		1						
			Philopotamidae			3	FIL										
				Chimarra sp.		3	FIL		29								1c
			Polycentropodidae			7	PRE										
				Polycentropus		6	PRE									1	
			Glossosomatidae			1	SCR										
				Protoptila		1	SCR		2								
			Rhyacophilidae			0	PRE										
				Rhyacophila		1	PRE		1				4	1			
		Hemiptera				5	PRE					2					
			Corixidae			5	PCR										1a
		Coleoptera															
			Dytiscidae			5	PRE									1	
			Carabidae										1b				
			Elmidae			5	SCR										
				Dubiraphia		6	SCR			6				7	2		
				Optioservus (L)		4	SCR		101				87	81	35		
				Promoresia		2	SCR		1				5	1			
				Stenelmis (L)		5	COL		4	8	14	3	3	43	26		
				Optioservus (A)		5	COL	1	11		5		21	33	8		
				Stenelmis (A)		7	COL		4	1	16		1	3	2		
			Hydrophilidae			5	PRE										
				Berosus		5	COL		1								
			Psephenidae			4	SCR										
				Ectopria		4	SCR			1							
				Psephenus		4	SCR		14								
		Megaloptera				2	PRE										
			Sialidae			4	PRE										
				Sialis		4	PRE	1		1							
		Diptera															

Appendix B. Taxonomic list of benthic macroinvertebrates collected in New York; September, 1993.

Phylum	Class	Order	Family	Genus	Species	HBI Tol	Feeding Group	Station Locations CHR4-300	OC4-300	OC1-300	OC2-300	OC3-300	CC1-300	CC2-300	CC3-300	HB1-300	HB2-300
			Athericidae			2	PRE										
				Atherix		4	PRE						7	1	3		
			Empididae			6	PRE										
				Chelifera		6	PRE						2	1			
				Hemerodromia		6	PRE		3				12	6	2		
			Muscidae			8	PRE										
				Limnophora?		8	PRE	6								7	
			Simuliidae			6	FIL										
				Simulium		6	FIL		1				16	2			
				Prosimulium		2	FIL	1								8	
			Tipulidae			3	SHR										
				Antocha		4	COL		2		1		10	20	1		
				Dicranota		3	PRE									1	
				Pedicia		6	PRE	1									
				Tipula		4	SHR							1			
			Chironomidae			7	COL										
				Cladotanytarsus *		7	FIL			2							
				Chironomus		10	COL			2				7		1	194
				Cryptochironomus		8	PRE		1	29		1					
				Cryptotendipes		6	COL			1							
				Demicryptochironomus		8	COL			1							
				Dicrotendipes		6	COL			9							8
				Micropsectra *		5	COL	1					1				
				Microtendipes		6	FIL		3	22	3			7			
				Nilothauma		2	COL			7							
				Paratanytarsus *		6	FIL			3							
				Paratendipes		5	COL								1		
				Phaenopsectra		7	SCR							1			
				Polypedilum		5	SHR		1	4		2		1	2		
				Rheotanytarsus *		6	FIL				1						
				Stempellinella *		4	COL			3							
				Tanytarsus *		6	FIL		2	3	1		2	22	2	2	1
				Tribelos		5	COL			1	1			1	1	2	
				Diamesa		4	COL	1									
				Prodiamesa		3	COL							1			4
				Undet. Orthocladinae		4	COL								1		
				Cardiocladius		6	PRE		1		19	1				1	
				Cricotopus		6	SHR		8	23	28	44	16	55	4	26	2
				Eukiefferiella		6	COL	9		2	31	2	16	19	5	15	
				Nanocladius		3	COL		1	2		1					
				Orthocladius		6	COL			1		1	3	4			
				Parakiefferiella		4	COL			19			1				
				Parametriocnemus		4	COL	1		1			9	1	1	4	
				Psectrocladius		5	SHR			2							
				Rheocricotopus		6	COL				2	3		19	1		
				Ablabesmyia		7	PRE			5							
				Conchapelopia		6	PRE		1	8	2			3	1		2
				Larsia		6	PRE			1							

Appendix B. Taxonomic list of benthic macroinvertebrates collected in New York; September, 1993.

Phylum	Class	Order	Family	Genus	Species	HBI Tol	Feeding Group	Station Locations CHR4-300	OC4-300	OC1-300	OC2-300	OC3-300	CC1-300	CC2-300	CC3-300	HB1-300	HB2-300
				Procladius		9	PRE			1							
				Chironomidae pupae				1	3	6	6	8	8	16	2	1	
				Diptera adults										1	1		
		Number of individuals						850	449	264	312	361	480	638	322	365	323
			Lepidoptera - terrestrial										1				

Samples from New York
Processed by Dr. Bozena Kukdinska
Monticello Ecological Research Station

L=larvae
A=adults
a=non-benthic taxon (not included in metric calculations)
b=terrestrial taxon (not included in metric calculations)
c=Station HB2-300 has one Chimarra sp. noted as part of this sample. This organism was believed to be left in the kick net from the previous station; OC4; and was thus, not included in the calculations for station HB2.

Appendix C
Quality Control Elements

Activity	Routine, Method, or SOP and Responsibility
habitat assessment	as per Barbour and Stribling 1991; parameters and rating procedure described in section 2.1, this document; observations performed prior to benthic sampling in order to avoid bias; original field data sheets archived in Tetra Tech, Owings Mills, MD, office; responsibility - **Dr. J.B. Stribling**, Tetra Tech, Inc., 10045 Red Run Blvd., Suite 110, Owings Mills, MD 21117
benthic sampling	as per Plafkin et al. 1989; also described in section 2.2, this document; double-composite $1m^2$ kicknet samples, mesh size, standard no. 30 mesh (openings 600μ), larger substrate particles (cobble and small boulder) scrubbed by hand to dislodge attached organisms; 1 from fast water riffle composited with 1 from slow water riffle in sieve-bottomed bucket (openings 600μ); organisms adhering to or entwined in net removed with forceps and placed into sieve bucket; responsibility (for Ohio case study) - **Dr. J.B. Stribling, S. W. Lipham**, Tetra Tech, **Dr. G.A. Burton, Ms. Katherine Jacher**, Biological Sciences Department, Wright State University, Dayton, OH 45435, **Mr. Chris Faulkner**, U.S. EPA/AWPD/Monitoring Branch (WH-553), 401 M Street, Washington, DC 20460; (for New York case study) - **Dr. J.B. Stribling, Ms. C. Gerardi**, Tetra Tech, **Ms. Marjorie C. Coombs**, U.S. EPA, Office of Science and Technology, Standards and Applied Sciences Division, 401 M Street, SW #4305, Washington, DC 20460
subsampling	described in section 2.2, this document; emptied from sieve bucket into gridded sorting tray (with numbered grids), manipulated into even spread within tray; if too much detrital or algal content, sample split into two trays (when split between two trays, identical grids are picked simultaneously between the two); using random numbers table, individual grids selected for picking, all organisms removed with fine forceps and placed directly into prelabelled sample container with approximately 70% ethanol; counted organisms placed in container; successive grids selected until AT LEAST 300 organisms were obtained (Ohio), 200 or 100 organisms (New York); if subsample total was reached prior to completing a grid, the remaining organisms were removed form that grid; for mobile organisms, visual estimates were made of the number of individuals moving into and out of the grid being picked and an approximation of that estimate was taken (Ohio), new subsampling screen greatly reduced mobility of organisms for the New York study; responsibility - **Dr. J.B. Stribling**
taxonomy	taxonomic literature used in performing identifications is presented in section 2.2, this document; responsibility - **Dr. M.C. Swift and B. Kulinska**, Monticello Ecological Research Station, University of Minnesota, P.O. Box 500, Monticello, MN 55362; cladocerans were identified by **Dr. Stanley Dodson**, Department of Zoology, Birge Hall, University of Wisconsin, Madison, WI 53706 (Ohio study only)
voucher specimens (samples)	in storage, responsibility - **Dr. J.B. Stribling**
abundance totals in metric calculations	special considerations in the use of abundance totals for calculation of the metrics is presented in section 2.2 of this document; responsibility - **Dr. J.B. Stribling, Ms. C. Gerardi**, Tetra Tech
metric calculations	metric calculations were performed by hand according to the individual metric descriptions presented in section 2.3 of this document; approximately 21% of the metrics were recalculated by hand as a QC check; another approximately 10% were recalculated by computer as further check; responsibility - **Ms. C. Gerardi, Dr. J.B. Stribling**
report preparation	authorship, organization, graphics production; responsibility - **Dr. J.B. Stribling, Dr. Michael T. Barbour**, Tetra Tech

Problems (Ohio Study)	Action(s) taken
high water, unable to sample Cuyahoga River stations	aborted sampling activity on 9/10/92 following completion of Scioto (9/8/92) and Sandusky (9/9/92) sampling; opted to return in 2 weeks, tentatively set return for 9/24/92; on returning 9/24/92 and Cuyahoga still 3 feet above normal and unable to sample, via pay telephone to Ohio EPA (J. DeShon) located workable stations on the Little Cuyahoga River
ecoregional reference station for Little Cuyahoga River flooded, 9/24/92, unable to sample Breakneck Creek at Kent	decided to rely on site-specific upstream reference (station CR1 at Mogadore)
high water at Ohio EPA-recommended sampling station prevented sampling (Little Cuyahoga River at Mogadore)	sampled approximately 0.1 mile farther upstream
depressed abundance of organisms in kicknet samples at Little Cuyahoga stations CR2 and CR3	total samples picked, but still falling below 300-organism goal
needed rapid turnaround time on taxonomic analysis of samples	primarily generic-level identifications performed
Problems (New York Study)	**Action(s) taken**
deep water, muck bottom - unable to sample beyond RBPI screening assessment at HB3	ended assessment at RBPI level, site (HB3) not used in biological assessment
hyperabundance of amphipods at regional reference site (CHR4) for Harbor Brook & Canastota Creek	upstream site on Canastota Creek (CC1) used for reference comparison
conductivity meter began to give erratic reading	stopped taking conductivity readings