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Biological Criteria: Research and Regulation

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Opening Remarks

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This symposium is in part the result of a series of developments within EPA which have progressively led to a more expanded and comprehensive role by the Agency in the protection and restoration of our nation's surface water resources.

These developments particularly received impetus from the mandate of the 1987 Clean Water Act and from a report written by our Administrator, William Reilly. He wrote "Meeting Environmental Challenges" earlier this year which summarized EPA's progress and innovations in environmental protection. As head of our agency, Bill Reilly urged our renewed commitment to ecology and natural resources conservation. The Agencies' mission goes beyond protection of human environmental interests. EPA is equally responsible for protecting fish and wildlife habitats and other ecological systems. In his report and in many other reports and directives, Bill Reilly has set Agency policy and given EPA a clear mandate to truly protect the environment in the broadest sense of the phrase.

Contributing to Bill Reilly's strong statement was a report by the EPA Science Advisory Board which stressed much the same breadth of concern. That 1987 report, "Unfinished Business", also advocated that we assume an expanded environmental responsibility and it identified many vital environmental risks requiring attention.

The basic, and perhaps obvious, conclusion therefore is that many environmental problems still exist even after 20 years of progress. EPA must now view its responsibilities for broad environmental protection as a proactive policy. It should do so as an expansion of our earlier agenda of pinpointing environmental strategies to react to preexisting problems. Thus, EPA recognizes that we should provide more guidance to state and federal authorities as to what is necessary to protect as well as to restor the environment. With relative success in the latter, we can now emphasize the former.

Finally, EPA must lead the nation in understanding that national environmental policy must include new initiatives focusing more on opportunities for improvement through nonregulatory and nonlegislative means. There is definitely now an emphasis on outreach; on training and education; as well as on identifying economic incentives to encourage the public to help protect the environment through responsible consumerism such as energy and water conservation, safe product disposal, and recycling.

More specific to our meeting here, the Science Advisory Board also recommended that the EPA attach as much importance to reducing ecological risk as it does to reducing human health risk. This focus is especially relevant to this conference because natural reproductive ecosystems are essential to human health and to sustaining long-term economic growth. These natural systems are, of course, also intrinsically valuable for their own sake.

The Board has also recommended that EPA improve the data gathering, handling procedures, sampling methodologies, and assessment methods to identify and evaluate environmental risks. They urged us to develop more and better scientific and technical measures to improve our understanding of primary ecological risk problems and to identify solutions.

In his subsequent report, Administrator Reilly focuses on four areas of risk the Board identified as critical. These include: habitat alteration and destruction which has always been a prime concern of EPA, particularly with respect to wetlands; species extinction and overall loss of biological diversity, also very important to our Office; and ozone depletion and global climate change both of which affect many EPA initiatives.

We feel that work on ecological criteria is essential to several of the objectives of the Office of Water. These include not only the regulation of discharges, but also the control of nonpoint source pollution

and storm water runoff and the determination of overall surface water quality problems and management priorities. In pursing this direction, we are concerned about biological community quality, integrity, and diversity of the collection of species and organisms. Thus, we aim to develop biological criteria that can be used independently, but to complement not replace existing physical and chemical water quality criteria and standards.

Several states have already begun to implement such ecological criteria as will be documented by the presentations at this conference. We are depending on you, the scientists, citizens, and state agency managers here to help us meet the challenge presented to the Office of Water by the Administrator. We want to consult with top federal, state and private organizations to help us design our biological criteria.

Thank you all for being here. We look forward very much to your participation and contributions, and I hope this is just a part of the beginning as we continue through the development and the implementation of biological criteria.

Establishing Biological Criteria: Functional Views of Biotic Community Organization

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As we enter the 90s, public awareness of environmental issues is at an all-time high, because the planet's environmental "health" is at an all time low. Since water is probably the most critical limiting resource, the status of surface waters is often at the forefront of concern. Terminology such as "drinkable," "swimmable," fishable," and "biological integrity" in the 1972 Water Quality Act, and its subsequent amendments, indicated the broad nature of this concern.

Over the past few years, the Nation has addressed the relatively easy, short-term problems of point source contamination with reasonable success. In many cases, chemical analyses based on the single grab sample technique were sufficient to identify a problem and verify the remediation. However, it soon became clear that the real long-term problems stemmed from nonpoint sources. Degradation over ever larger spatial and longer temporal scales was the result of agriculture (row and field crops, grazing), timber harvest (clear cutting), surface mining, landscape pesticide applications, general urbanization, and regional watershed fallout such as acid rain.

Monitoring strategies such as EPA's MAP (Environmental Monitoring Assessment Program) and the U.S. Geological Survey NAWQA Program (National Water Quality Assessment Program) are attempting to assess the severity of the problem. Yet, as before, significant questions still remain regarding how to evaluate the condition of our surface waters, and what role biological criteria should play in its assessment.

The Gap in Technology Transfer

The fundamental problem is that the process of establishing biological criteria for water quality assessment has not been coupled to the major advances in lotic ecology of the last 25 years. Some examples are: the River Continuum Concept (Vannote et al. 1980; Minshall et al. 1983, 1985; Cummins et al. 1984; Cummins, 1988), including the related paradigms of the Intermediate Disturbance and the Serial Discontinuity Hypotheses (Ward and Stanford, 1983); Nutrient Spiralling (Elwood et al. 1983); Riparian Control (Cummins, 1988); Watershed Budgets (Fisher and Likens, 1973; Cummins et al. 1983); and Functional Groups (Cummins and Klug, 1979; Merritt and Cummins, 1984; Cummins and Wilzbach, 1985). Furthermore, few of the researchers who have played major roles in developing the above paradigms in running water ecology have been involved in the development or implementation of the assessment criteria.

Symptomatic of this schism is the almost complete dependence of the biocriteria development on taxonomic procedures, while basic researchers have moved into questions or process and function. Thus, an identification and quantification approach formed the basis for biological water quality assessment and remains fundamental today. The pattern can be seen in EPA workshops: indicator species in the late 1960s, diversity indices in the late 1970s, and biocriteria in the 1980s.

The reliance on taxonomically dependent numerical indices, most frequently applied to invertebrates, has severely limited the development of bioassessment indices. The main problem is that the taxonomic definition of North American freshwater invertebrates is an ongoing, long-term process, with all groups remaining incompletely described.

Probably the most extreme example among the macroinvertebrates is the dipteran insect family, the Chionomidae. This family is, almost without exception, the most diverse species complex in any running water system (Merritt and Cummins, 1984),

and yet it typically is the least known. In a Pennsylvania stream that has been studied for more than 25 years, over 180 species of Chionomidae have been found on an annual basis (Coffman et al. 1971; Coffman 1973, 1974). Many of these species are undescribed and keys for their separation await monographic treatment. It is clear that the assignment of a taxonomic value of 1 to the Chionomidae in an assessment of a running water system will usually subsume a species complex in excess of 50 (on an annual basis).

Given the embryonic state of taxonomic resolution of freshwater organisms, significant support for taxonomic research efforts on the part of agencies that need the information, such as EPA, might be expected. Unfortunately, such support is not forthcoming. Thus, derived indices that purport to detect biological redundancy in a given system usually find redundancy in the taxonomy. Because of the unavailability of sufficient taxonomic keys to separate running water organisms, process- or function-related measurements should be prime criteria for incorporation into bioassessment and biomonitoring protocols. Agencies such as EPA would greatly benefit from research devoted to moving some of the time-tested methods in the area of process and function in basic running water ecology into the field of application.

Reference Sites

The concept of reference sites within a given ecological region (Omernik, 1987) has been a welcome addition to strategies used in the evaluation of fresh waters (Hughes et al. 1986), particularly by state Environmental Protection Agencies. Evaluation of the biological condition of a given freshwater system relative to an appropriate reference ("control") site should be fundamental. However, the site to be evaluated must be placed not only in the appropriate spatial context (e.g., ecoregion, basin, watershed), but also within the relevant temporal (historical) perspective (Sedell and Frogatt, 1984; Cummins et al. 1984; Cummins, 1988).

Furthermore, the selection of reference sites needs careful attention. Any comparison must be made between systems of similar scale because there are fundamental differences between running water ecosystems that transcend ecoregions. For example, paramount in the River Continuum Concept is the idea that relative position of a stream in its watershed, designated by stream order, stream size, or drainage area, confers some similarities in ecosystem structure and function independent of the local setting of geology, soil, and stream-side riparian vegetation (Vannote et al. 1980; Minshall et al. 1983, 1985). Riparian vegetation can also exert influences that override such parameters as altitude, stream gradient, of bottom sediment type.

First-, second-, and many third-order streams in the Cascade Mountains of the Pacific Northwest have dominant populations of invertebrates, termed shredders, that feed on litter derived from the riparian zone. Such headwater streams of various gradients and lithology share this functional component and differ from streams of higher order (e.i. < 3 or 4) that are in the same region. However, if the riparian vegetation is predominantly deciduous, especially red alder (*Alnus rubra*), the normal recovery tree following a disturbance (such as clear cutting), the shredder populations are active in the streams largely in the fall–winter period. If the riparian vegetation is primarily coniferous, the shredder populations are most active in the spring–summer period (Cummins, et al. 1989). Thus, stream size is linked to the presence of shredders through the direct influence of the riparian zone, but the timing of the growth period of the shredder populations is keyed to the type of riparian plants and the timing of the litter inputs (Cummins, 1974; Cummins et al. 1989). The override is clear because both alder and conifer-dominated headwater streams can be found in all of the major ecoregions of the Pacific Northwest.

Given the need for carefully studied and well-documented reference sites, it is quite surprising that most of the best running water research sites have not been used for comparison. Much of the information on these potential reference sites is well documented in the literature. The tendency of workers involved in bioassessment to ignore this national data resource, which exists for virtually every major ecoregion, is further evidence of the gap between basic research and assessment. Many of the potential reference sites are associated with ongoing national research initiatives such as the National Science Foundation's Long-Term Ecological Research Program (OTER), the U.S. Geological Survey's National Water Quality Assessment Program (NAWQA), and projects at many of the 200 inland biological field stations. There can be no doubt that incorporation of these sites, and collaboration with their resident researchers, would help fill EPA's need for an extensive and carefully documented matrix of reference sites.

Function/Process Measurements as Biocriteria

Although many examples of procedures designed to incorporate functional and process-related data as biocriteria for bioassessment and biomonitoring

could be discussed, only two are given here. Both are based on the belief that the macroinvertebrates are often the most suitable lotic ecosystem components for such assessments. The small, rapidly reproducing microorganisms (algae, bacteria, etc.) that are quantitatively most significant at the level of nutrient cycling require specialized techniques for analysis. However, some methods that monitor the general activity levels of these organisms, such as the measurement of P/R (the ratio of gross primary production to total community respiration) certainly could be employed as biological criteria of ecosystem condition (Minshall et al. 1983).

At the other end of the scale, fish, often the biological components of most direct human interest, can also serve as effective subjects to establish biocriteria for the assessment process (Karr, 1981, 1987, 1991). However, analysis of fish populations requires special equipment due to their low densities and species complexity relative to microorganisms and invertebrates, and their great mobility.

Thus, the invertebrates constitute both a biological and operational link between the microorganisms and fish. That is, most invertebrates feed on microorganisms and in turn serve as food for fish, and their size, abundance, species diversity, ease of capture, and annual life cycle enhance their suitability for observation and interpretation relevant to ecosystem function and processes (Cummins and Klug, 1979).

The example of a functional analysis to be discussed here was first described 18 years ago (Cummins, 1973) and has been modified in some details since then (Cummins and Klug, 1979; Merritt and Cummins, 1984; Cummins and Wilzbach, 1985). The macroinvertebrate Functional Feeding Group (Cummins and Wilbach, 1985) method is based on the association between a limited set of feeding adaptations found in freshwater invertebrates and their basic nutritional resource categories. These food resources are categorized as detritus (coarse CPOM, or fine FPOM, particulate organic matter and the associated microbiota), periphyton (attached algae and associated entrained material), live macrophytes, and prey (Cummins, 1974).

The level of morphological and behavioral adaptation of the invertebrates that allows them to exploit these resource categories can be obligate or facultative (Cummins and Klug, 1979). The obligate specialist forms are more readily displaced and the facultative generalists are more tolerant under conditions of disturbance. The presence and abundance of the various functional feeding groups, and the dominance of obligate or facultative representatives, is a direct reflection of the availability of the required food resources and the condition of the related environmental parameters.

The invertebrate functional groups are:

- Shredders feeding on CPOM (primarily litter of terrestrial origin from the riparian zone) or live macrophytes;
- Collectors feeding on FPOM either by filtering from the water column (filtering collections) or by "mining" the sediments or browsing surface deposits (gathering collectors);
- Scrapers feeding on periphyton;
- Piercers feeding on macroalgae by piercing individual cells; and
- Predators feeding on prey (Cummins and Wilzbach, 1985).

The analysis is made on a hierarchical basis of increasing levels of resolution. The first (lowest) level of resolution allows separation of live invertebrate collections in the field at an efficiency of 80 to 85 percent. The second level of resolution increases the efficiency another 5 to 10 percent. When comparisons are to be made between sites on a regional basis the level of resolution must be set so that all workers involved in the assessment can accomplish the task. Levels of greater resolution allow groups having the appropriate expertise to produce more detailed analyses. An example of the method from Cummins and Wilzbach (1985) is given in Figure 1. Assignments into functional groups of most of the North American genera of aquatic insects can be found in the ecological tables in Merritt and Cummins (1984).

The figure shows two levels of resolution are indicated; the first level allows general separation, the second level allows correction of the assignments of some of the more common exceptions. The third level of resolution would rely on the taxonomic resolution (essentially at the generic level) given in Merritt and Cummins (1984).

The Leaf Pack Bioassay method is an example of a procedure that would be suitable for establishing biocriteria to be used in freshwater ecosystem assessment and monitoring. The technique was first described 17 years ago (Peterson and Cummins, 1974) and has undergone some modification since (Merritt et al. 1979; Hanson et al. 1985). The objective of the method is to use the rate of leaf litter processing, defined as the total weight loss from simulated litter accumulations from all causes, as a general measure of ecosystem structure and function. The method is suitable primarily for low order

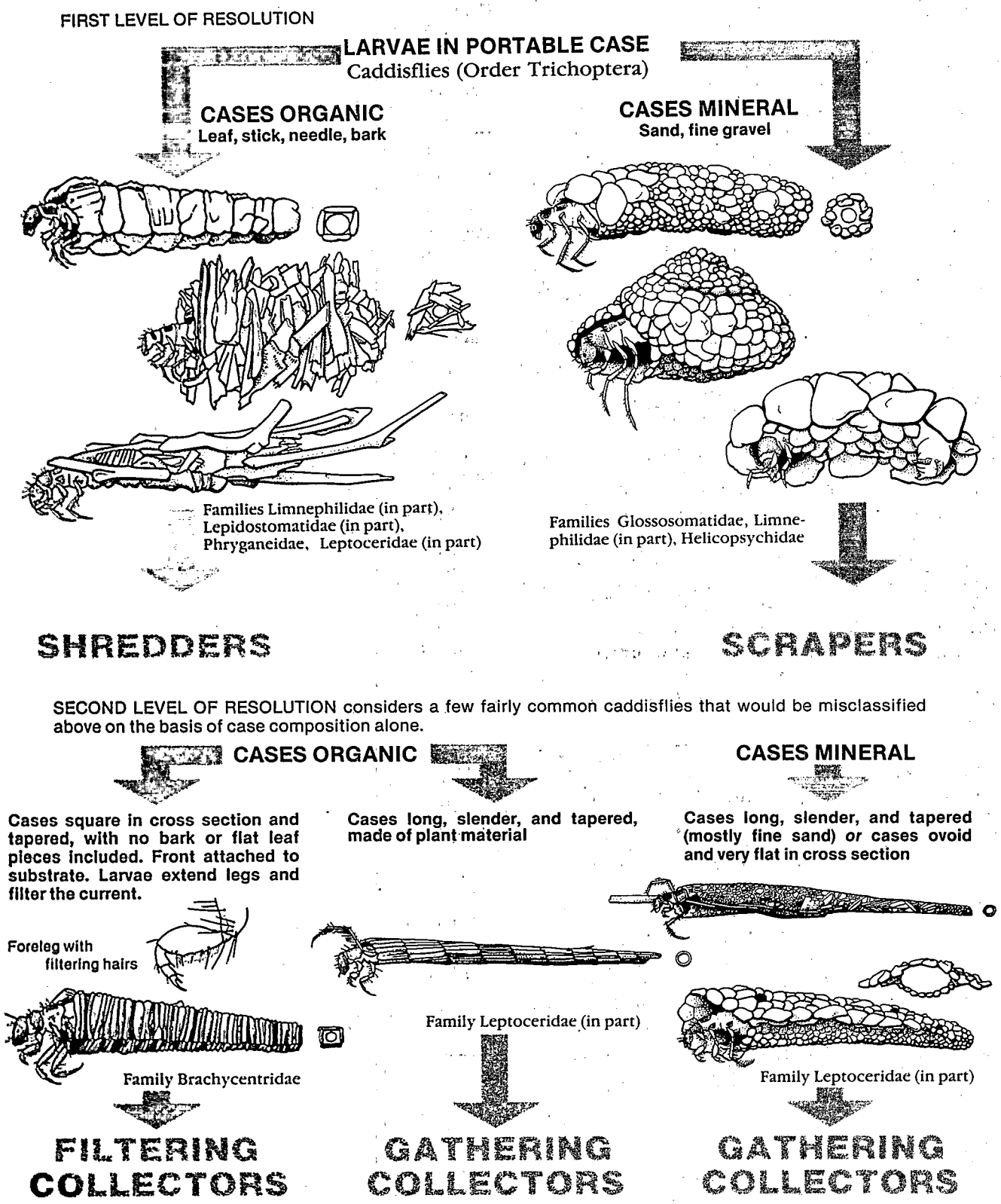

Figure 1.—An example of functional group separations in the case-bearing caddisflies (Cummins and Wilzbach, 1985).

(1-3 or 4), headwater streams where litter inputs and invertebrate shredder activity will be maximized per unit of habitat.

Processing rates have been determined for a wide variety of riparian plant species in a range of stream orders (Webster and Benfield, 1986). In general, the method involves the preparation of packs of leaves of a given species, or species combination, that are fastened together with plastic Tees of the type used to fasten buttons or price labels to clothing. The packs of preweighted leaves (usually 5 grams dry weight) are attached to elastic bands, slipped around common masonry bricks, and placed on the stream bottom with the pack facing into the current to simulate a natural accumulation of litter at the leading edge of an obstruction in the water. After 24 to 28 hours, the initial sets of packs are removed to determine leaching and handling weight loss; additional sets of packs are removed at intervals, determined by temperature (usually every 150 to 300 degree days) to follow the rate of processing (per degree day).

The invertebrates are removed and the recovered packs are reweighed. The invertebrate functional groups can be noted, especially shredders. Other analyses such as microbial population composition, densities, and biochemical changes from initial conditions can also be performed. The leaf pack bioassay method has been shown to accurately simulate natural rates of processing of unconfined leaves in exposed, aerobic stream sites, which was not true for litter held in mesh bags (Cummins et al. 1980).

As indicated previously, the examples given are only two among many that are well documented in the literature and have significant potential for use in establishing biocriteria in freshwater ecosystems. The point is that very few such methods have been incorporated into bioassessment and biomonitoring and, given the present policy of agencies such as EPA, it is unlikely that they will be. For example, the original papers describing the two methods used as examples above have both been designated "citation classics." Thus, it is clear that such techniques are well known (i.e., adequately documented) but, with few exceptions (Plafkin et al. 1989), they have not made the technological transfer to the problem-solving arena within which agencies such as EPA operate.

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Biocriteria in Regulations: the EPA Headquarters NPDES Permitting View

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ABSTRACT

Sections 303 and 304 of the Clean Water Act now require States to develop biological criteria as part of their water quality standards. Biological criteria are presently used in the NPDES permitting program as a tool to identify waters that are not achieving their designated use, and therefore may be impacted by point source discharges. In the future, there are two areas in which biological criteria can be used in an NPDES permitting context. The first way is to verify that NPDES permit limits are indeed resulting in achievement of State water quality standards. A second approach that would require considerable development is to establish NPDES effluent limits that directly assure compliance with biological criteria. Once biological criteria become part of State water quality standards, NPDES regulations require that permit limits assure compliance with these standards. To accomplish this, a permitting authority must develop a protocol that can demonstrate the relationship of the biological criteria to effluent characteristics. This goal has already been accomplished for toxicity. However, EPA takes the position that biological criteria must not supersede chemical-specific numerical standards or toxicity tests now used to achieve compliance with the narrative standards.

Introduction

Sections 303 and 304 of the Clean Water Act now require States to develop biological criteria as part of their water quality standards. As States begin to adopt biological criteria, NPDES permitting authorities are obligated by federal regulations to establish effluent limitations to ensure that these criteria are maintained. This paper describes the ways in which NPDES permits now assure compliance with water quality standards, and explores ways in which biological criteria could be used in concert with NPDES permits to achieve full implementation of water quality standards.

NPDES Program Overview

National Point Discharge Elimination System permits are the instruments by which EPA and States control the types and amounts of pollutants discharged into ambient receiving waters. The permits contain effluent limits that set the maximum level of discharge for each pollutant. The limits require the use of best-treatment technology to maintain State water quality standards. Many NPDES permits are issued with technology-based limits that are sufficient to protect the quality of the receiving water. For those permits where technology-based limits are insufficient, more stringent water quality based limits are required. These are set at the maxi-

mum level of a pollutant that, after mixing with the receiving water, will not violate a water quality standard.

EPA and States are now issuing a number of NPDES permits that contain new requirements to control the discharge of toxic substances that may endanger aquatic life or human health. These permits are issued to comply with EPA's national policy (Federal Register, 1984) stating that

> *. . . to control pollutants beyond Best Available Technology Economically Achievable (BAT), secondary treatment, and other Clean Water Act technology-based requirements in order to meet water quality standards, the EPA will use an integrated strategy consisting of both biological and chemical methods to address toxic and nonconventional pollutants from industrial and municipal sources. Where State standards contain numerical criteria for toxic pollutants, NPDES permits will contain limits as necessary to assure compliance with these standards. In addition to enforcing specific numerical criteria, EPA and the States will use biological techniques and available data on chemical effects to assess toxicity impacts and human health hazards based on the standard of 'no toxic materials in toxic amounts.'*

Existing Uses of Biological Criteria

The most effective use of biological criteria in the NPDES permitting program is as a screening and identification tool. Biological criteria provide the ability to identify waters that are not achieving their designated use, and therefore may be impacted by point source discharges.

The best example of the use of biological criteria in this fashion is in Arkansas, a State authorized to write NPDES permits. The State has developed a program to identify facilities with the potential for causing ambient toxicity through the use of whole-effluent toxicity (WET) testing. This program was initially established to apply to "major" NPDES facilities—i.e, those facilities where initial information suggested that toxics might be discharged. Arkansas has also used biological criteria and rapid bioassessments to identify other facilities that need WET evaluation. At several of these sites, the State found that a point source facility was discharging an effluent with sufficient toxicity to cause a biological effect. Two of these were Superfund sites in the remedial action stage.

In using biological criteria, EPA takes the position that biological criteria must not supersede chemical-specific numerical standards or toxicity tests used to achieve compliance with the narrative standards. Biological criteria and biosurveys should be fully integrated with toxicity testing and chemical-specific assessment methods in State water quality programs. Whenever any one of the three types of assessments demonstrates that the standard is not attained, appropriate action should be taken by the regulatory authority. However, since each method has unique as well as overlapping characteristics, sensitivities, and program applications, no single approach for detecting impacts should be considered inherently superior to any other approach. The inability to detect receiving water impacts using a biosurvey alone is insufficient evidence to waive or relax a permit limit established using either of the other methods. The most protective results from each assessment conducted should be used to establish the necessary NPDES permit limits. This concept is fully discussed in the 1990 *Technical Support Document for Water Quality-based Toxics Control* (U.S. Environ. Prot. Agency, 1990).

Potential Future Uses of Biological Criteria

In the future, there are two areas in which biological criteria can be used in an NPDES permitting context. The first way is to verify that NPDES permit limits are indeed resulting in achievement of State water quality standards. The biosurvey data used for assessment against the criteria could be collected by the regulatory authority or by the NPDES permittee. EPA and States have the authority under section 308 of the Clean Water Act to require information necessary to establish permit limitations. In some instances this authority could be used to require facilities to conduct the biosurvey.

A second way in which biological criteria could be used to assess water quality is to establish NPDES effluent limits that directly assure compliance with biological criteria. This approach requires considerable development before it can be generally implemented. Once biological criteria become part of State water quality standards, NPDES regulations require that permit limits assure compliance with the standards. To accomplish this, a permitting authority needs to demonstrate the relationship of biological criteria to effluent characteristics.

This goal has already been accomplished for toxicity. EPA conducted eight stream surveys to cor-

relate effluent toxicity tests to actual ambient biological impairment. The positive results of this research enabled EPA to use effluent toxicity controls in NPDES permits as a control mechanism. In addition, EPA developed a toxicity identification evaluation (TIE) process to identify the causes of toxicity in effluents and to suggest possible remediation measures. An analogous procedure is needed to fully implement biological criteria in an NPDES context.

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Biocriteria in the Water Quality Management Process

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Recent EPA management directives have stressed the need to attach as much importance to reducing ecological risk as to reducing human health risk. EPA and the States are beginning to address non-traditional problems such as nonpoint source pollution, habitat degradation, the effects of land use practices, and stormwater discharges. Many of these problems may not involve chemical pollution as the principal stressor. Biological criteria (biocriteria) will be necessary to identify these types of problems and to develop the tools needed to devise mitigation strategies. Biocriteria fit well into the existing regulatory approach. Substantial progress is being made to institute monitoring programs and resolve issues of consistency in application. EPA has adopted a policy of independent application to govern the interpretation of information from biosurveys, chemical analyses, and toxicity tests.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Biological Criteria: A Regional Perspective

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The Water Management and Environmental Services Divisions in Region III have initiated various activities in preparation for the integration of biological criteria into the existing state water quality programs. The Divisions have drafted a strategy which identifies both short- and long-term goals for incorporating biological criteria and standardized bioassessment protocols into the state programs and defines the responsibilities of each Division to implement the strategy. The Region's short-term goals include the adoption of narrative biological criteria and the use of EPA's Rapid Bioassessment Protocols by the state programs, and further definition and refinement of ecoregions in support of numeric biocriteria. The Region's long-term goals include the adoption of numeric biological criteria by the states, establishment of comprehensive data sets for ecoregion reference sites, and improvement of long-term water quality assessments through the use of environmental indicators.

The Water Management Division (WMD) has the lead in providing program guidance and assistance to the states for the incorporation of both numeric and narrative biological criteria into their water quality standards regulations. WMD has informed all Region III states that in order to satisfy EPA water program requirements, they are expected to adopt narrative biological criteria for all surface waters into their state water quality standards during the FY 1991-1993 triennium. In addition, WMD has begun working with individual States to determine what revisions are necessary to the existing state water quality standards to meet the program requirement for biological criteria.

The Environmental Services Division (ESD) has the lead in providing technical assistance to the states in the areas of bioassessment methodologies and their applications. During 1990, ESD, in cooperation with EPA Headquarters, held four Rapid Bioassessment Protocol workshops for the state aquatic biology staffs. These workshops outlined and demonstrated standard assessment procedures that are utilized to ensure consistent and valid data collection in support of biological criteria. Each session consisted of: 1) description and discussion of major concepts, 2) field trip to demonstrate habitat assessment and sampling techniques and 3) data analysis and discussion of assessment results. Approximately 30 representatives from state, interstate and federal agencies were in attendance at each workshop. The Region received positive response, and the state agencies are proceeding to adopt the rapid bioassessment protocols.

In addition, ESD coordinated a workshop on a specific habitat type (i.e., nontidal coastal streams) at which data collection procedures utilized by the Region III coastal states were described, standardization of methods was outlined, and the development of biocriteria for that system was initiated. At the workshop, the attendees decided to organize four subgroups whose purpose would be to develop standard operating procedures for the following areas: 1) field collection, 2) laboratory processing and data analysis, 3) habitat assessment and coastal stream characterization and 4) reference site selection. These work groups reviewed the ongoing standard operating procedures used by each individual state and developed common standard operating procedures suitable for all the states. The final draft of the procedures will be presented at the Region III Biology Workshop in March 1991. ESD will continue to provide training and technical support to the states regarding the assessment procedures and their application.

In addition, ESD is developing a cooperative program that will assist the states in accumulating baseline reference site data to support the biological criteria program. This program will refine ecoregions and subregions that cross state bound-

aries and will provide coordination of regional and interstate data collection activities for the development of biological criteria. In 1991 the Region, in cooperation with the EPA Office of Research and Development-Corvallis laboratory, will hold a Reference Site Workshop with the state agencies to identify reference watersheds and to select reference sampling sites for a pilot ecoregion (i.e., the Central Appalachian Ridge and Valley). Following the workshop, the reference sites will be field evaluated in cooperation with the states. Once the reference sites are finalized, comprehensive biological sampling and habitat assessments will be periodically performed to provide the necessary baseline data.

EPA Region III strongly supports the expansion of the use of biological parameters in the state programs. The Region's future role concerning the biocriteria implementation will be to provide technical assistance and reference site biological data. All of the Region III states have strong biological assessment programs and are capable of meeting the challenge for the expanded use of biological data.

A State Perspective on Biological Criteria in Regulation

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ABSTRACT

The State of Ohio has been a leader in developing numerical biological criteria and exploring their use in aquatic resource assessment. More recently Ohio has adopted state water quality standards which inextricably link aquatic life beneficial use designations and numerically expressed biological criteria. With this foundation Ohio has shown that biological criteria incorporated into state regulations can have a powerful impact on a wide range of water resource issues. This paper will highlight the role biological criteria have played in (1) section 401 water quality certifications for stream channel modifications; (2) the nonpoint source assessment process and the section 319 management plan for Ohio; (3) stream use attainability issues; and (4) the NPDES permit system. Although Ohio has realized extensive benefits through the biological criteria program, the development and application process has been controversial. Ironically, the focus of biocriteria on water resource quality, as opposed to just water quality, can, in some situations, pose a real dilemma for managers charged with both narrowly defined regulatory responsibilities and the obligation to carry out reasonable actions in the public's interest. For example, our propensity to "solve" water quality problems on a cost-effective, regional basis has led to dewatering and habitat alteration of small headwater streams in Ohio and elsewhere. Is this furthering the goal of biological integrity? In other situations, small communities have spent large sums of money to upgrade wastewater treatment to meet water quality standards on waterways that in subsequent years may be subjected to routine "channel maintenance" (i.e., habitat destruction and loss of biological integrity) associated with the petition ditch laws of many midwestern states. These are just two examples that illustrate the need for major re-drafting of much of our water resource legislation in this country if we are to realize the biological integrity goal of the Clean Water Act. For the environment, as well as the public's sake, we must begin to better manage the whole of water resource quality. The present programmatic emphasis on developing biological criteria affords us the opportunity to forcefully re-examine and extend the focus of federal, state and local regulations from just water quality to water resource quality.

Introduction

Ohio has developed a fairly extensive biological survey program and biological criteria stream performance standards, based on various attributes of the resident fish and macroinvertebrate communities. Three separate numerical biological criteria were incorporated into the Ohio administrative code in a water quality standard rulemaking in February 1990.

Before describing Ohio's regulatory experience with biological criteria though, it is valuable to focus on the fairly common perception of water pollution control regulation as an impossible goal and constantly moving target.

The same scenario plays out time and time again. First technology standards are issued, then revised downward to reflect best available technology, new water quality standards are adopted, followed by new or revised water quality based permitting procedures—all of which results in new

or more stringent permit limitations. When faced with typically hostile reactions from the permittee over this moving target and the need for investment in pollution abatement, a common reply from the regulatory official is that in keeping with the philosophy behind the Clean Water Act regulators are seeking incremental steps in pollution abatement and—step by step—just racheting permit limits toward zero discharge.

This cycle has recurred several times in Ohio, and probably in most other states. In fact, this racheting syndrome has become so pervasive in our regulation that it is fair to compare the fate of the permit holder to that of Sisyphus, a figure from Greek Mythology. Sisyphus was the king of Corinth and was noted for his trickery and wicked nature. He was condemned to forever push a huge stone up a mountain in Hades only to have it roll down again just as he almost reached the peak. The stone represents the permit, the top of the slope compliance.

A myth represents how people perceive the world around them and their place in it. The regulated community perceives itself in the role of Sisyphus. Scientists and technicians of the permitting process don't necessarily believe or share that perception. There are very complete and defensible reasons for writing new and more stringent permit limitations. However, technical explanations of the new permit limitations will do little to change the perception of the present situation by the regulated community, and increasingly, some members of the informed public.

Thus, it is important to shape positive perceptions of the permitting process and its goals in the minds of the regulated community and the public, because, ultimately, they shape the lawmaker's opinion of our regulations. The mandate to regulate was given by the public through the nation's elected officials, and is subject to review and change. The challenge of rational and understandable regulation development can possibly be met through use of biological criteria in the regulatory process.

Five different examples of how biological criteria have been applied in Ohio should make this clear. These examples are: (1) water quality standard use designations; (2) section 319 – the nonpoint source program; (3) section 401—water quality certifications; (4) section 305(B) – water quality inventory report; and (5) NPDES discharge permits.

These examples contain a common characteristic; in each, biocriteria provide a direct measure of water quality, protecting the chemical, physical, and biological integrity of our nation's waters. Such a direct and objective assessment of the biological integrity goal fosters a positive perception of regulation.

Stream Use Designations

Stream use designation is the longest running regulatory application of biological criteria. It began as a system of tiered aquatic life use designations launched in narrative language, and has evolved to incorporate numerical biological criteria. The linking of biological criteria to stream use designations was particularly important to Ohio because the State Water Quality Standard Rules in 1978 classified over 60 highly polluted stream segments as less than fishable/swimmable uses. The task of reevaluating the potential aquatic life use designations for these and other streams was based upon biological and water quality survey results and the biological criteria.

Although some appeals are still pending, to date the upgrade of all these stream segments to fishable/swimmable uses has withstood legal challenges. Part of the credit for this must be given to the strong theoretical and technical foundation provided by the biological criteria. It was clear to Ohio's environmental Board of Review and the appeal courts that the Ohio Environmental Protection Agency had done its homework. More important, water resource quality goals were clearly articulated and measured, and the goals were in fact attainable given the installation of reasonably advanced treatment technologies.

The Nonpoint Source Program

The state of Ohio has developed a nonpoint source program consisting of two parts, an assessment and a management program. The development of the assessment program was coordinated by the Ohio EPA, while the development of the management program was coordinated by the Ohio Department of Natural Resources. Both components of the program have been approved by U.S. EPA.

Biological survey data and Ohio's biological criteria for streams and rivers played a key role in the assessment process. A total of 48 stream segments were targeted for nonpoint source implementation projects based upon aquatic life use impairment that was documented in biosurvey results. This was done by comparing the measured Index of Biotic Integrity (IBI) values to the IBI criteria values for the appropriate ecoregion specified in the Ohio water quality standards. For the purpose of the assessment, only IBI values that departed from ecoregion standards by more than 10 percent were considered significant.

Streams and rivers with severe habitat problems caused by channel modification also were setaside in this analysis because the impairment problems were considered to be long term and intractable through land application of best management practices. Next, a cumulative score for the severity of pollution over the length of the stream segment was calculated by graphing the measured IBI values versus river mile and connecting the points. The area below the biological criteria for IBI was determined and used as a ranking score.

The end result was a list of 48 stream segments with reasonably good habitat characteristics that should respond in a relatively short time period to accelerated implementation of best management practices. The management program has given priority to funding section 319 projects in these watersheds. In summary, monies were targeted toward stream segments that were selected based upon a direct and objective measurement of extent and severity of biological use impairment.

Finally, Ohio also established a special setaside of 10 percent of section 319 project funds to be directed toward the protection of threatened waters. These were defined as waterbodies that were known through the biological survey program to be of high quality. These setaside funds help preserve high quality waters in the face of mounting nonpoint source pollution threats caused by rapid changes in land use patterns.

Section 401 Water Quality Certifications

The placement of fill in navigable waters of the United States is regulated through section 404 permits issued by the Army Corps of Engineers. Pursuant to section 401, all permits for such activities must include a water quality certification from the appropriate state authority. The purpose of this program is to ensure the permitted activities will comply with state water quality standards.

For purposes of example, assume a proposed 404 permit is drafted to allow the placement of fill in a stream channel and the relocation of several thousand yards of stream bottom. Because Ohio has adopted narrative and numerical biological criteria associated with aquatic life use designations, section 401 issues can be reviewed based on the traditional chemical water quality criteria, plus the expected impact of the proposed channel work upon the direct biological measurement of use attainment. The state has been very successful in achieving major adjustments in the scope and design of stream channel work, and when such accommodations are not forthcoming, the Ohio EPA has denied the section 401 Water Quality Certification.

The Section 305(B) Report

A biennial water quality report is required of each state by the U.S. Environmental Protection Agency. Whenever available, Ohio uses biocriteria as the final arbitrator for determining the status of stream use attainment or impairment. It is estimated that 50 percent of impaired waters would be misclassified as attaining Clean Water Act goals if biocriteria were not available. Although this is unfortunate, the protocol allows identification of problems so reasonable corrective measures can be formulated.

The NPDES Permit Program

A direct use of biological criteria for permit limitations or for specific design of wastewater treatment in the point source program is not possible, and given the present regulatory structure, such a use should not really be expected. However, biosurvey results and comparisons of stream performance as measured by the biological criteria have been helpful in the permit reissuance process in Ohio. Some of the typical applications and their advantages include:

1. Documentation of aquatic life use impairment under present pollutant loading. These data provide strong evidence that investment in pollution abatement is justified. However, evidence of aquatic life use impairment should not be a prerequisite for the appropriate implementation of limits based upon a reasonable expectation that water quality standards may be violated.

2. Documentation of aquatic life impact, either near by or far from the discharge point, can help determine appropriate regulatory response to whole effluent toxicity testing programs that are inconclusive regarding the potential for a hazard to resident aquatic life. Based on this data, the permittee might be required to begin a toxicity identification evaluation or toxicity reduction evaluation.

3. Documenting aquatic life impact can detect previously unknown or "under regulated" sources. Nearly every biological and water quality survey conducted by the Ohio EPA

has detected a problem previously unknown to the paper world of permits and compliance. Such impacts include the discovery of contaminated nonprocess waters, unreported episodes of chemical spillage, generally poor housekeeping at the facility, or nonchemicals at the point of discharge. In urban areas bioassessment can also better define the need for attention to combined sewer overflows and stormwater controls.

The concept of a reasonable potential to violate water quality standards is open to various interpretations. When biological criteria are violated downstream from a point source, regulators need to react quickly and with environmentally conservative methods and assumptions to control the source. However, when biological criteria are attained downstream from a point source, regulators need to react in a slightly different manner if those same environmentally conservative methods predict possible water quality standards violations. That reaction should consider the documented biological criteria attainment as feedback that triggers a reassessment of the modeling techniques that predicted the water quality standard violations. These situations require stepping up from simplistic mass balance models to more complex applications such as fate and transport, dynamic, or probabilistic type modeling. The outcome may still be the same—a prediction that water quality standards have been violated—or have not. The point is that biological criteria were used to help define what a reasonable showing is.

In summary, there is no doubt that the public perception of water pollution regulation is becoming increasingly important. Biocriteria can play a valuable role in creating positive, reasonable, and environmentally protective approaches to water pollution regulation.

Utilization of Biological Information in North Carolina's Water Quality Regulatory Program

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ABSTRACT

The utility and effectiveness of biological assessments in water quality programs is intrinsically linked to the criteria and standards within a state's regulations. Realizing the scope and limitations of this information, as well as seeking innovative and expanded uses, can enhance program capabilities. Environmental surveys assess existing in-stream conditions and identify source and extent of impairments to biological integrity. This information is transferred to the regulatory process by having aquatic life standards (narrative or numerical) within state regulations. North Carolina Water Quality Regulations contain narrative standards for aquatic life protection. Biological information is used in assigning appropriate designations for supplemental classifications including Nutrient Sensitive Waters, High Quality Water, Outstanding Resource Waters, and Trout Waters. Development of biological assessment capabilities and regulatory use of information gathered should not be independent processes. Classifications and standards can be written to use biological information, and choice of biological information gathered and presented should be influenced by its regulatory use. Qualified ecologists with strong interpretive capabilities are the most necessary element of good programs. Integrating science and regulations is an ongoing challenge for state and federal programs.

Introduction

In-stream biota has been utilized to assess water quality in North Carolina since the mid-1970s. Initially, work was conducted in free-flowing streams to identify impaired waters of the State using colonization of macroinvertebrates onto artificial substrata. However, beginning in 1982, criteria in three major ecoregions were developed to establish relative degrees of impairment in flowing streams. Impairments in lakes and large rivers due to cultural eutrophication led to development of expertise in limnology and phytoplankton ecology within the North Carolina program. Assessments of these waters and changes in regulations provided specific means of protecting waters from impairment due to enrichment.

Work has begun in developing an environmental integrity index similar to Karr's IBI and based on fisheries community habitat and community structure (Karr, 1981). Development of this tool would provide information from another major group of biota, and should facilitate assessment of impacts resulting from sedimentation and other nonpoint source pollutants.

Refining biological criteria to account for all of the variables resulting from natural and human causes is a goal of many programs but has yet to be completely achieved. Attempts to develop these criteria have resulted in many indices, some useful, but all with limitations. The presence of gross pollutants is fairly easy to identify with any assessment method. Identifying subtle impairments requires

species/genus-level analyses involving the ecology of specific organisms and groups of organisms.

Careful selection of baseline sites and subsequent acquisition of baseline information is imperative when dealing with the inherent variability of stream size, as well as seasonal and regional differences among samples. Selection of defensible, reproducible, and efficient sampling methodology and analyses by competent, well-trained ecologists provide the basis for acquiring assessments that meet the immediate needs of the program, as well as long-term needs such as development of biocriteria. Biological information must be integrated with all phases of water quality assessment, including chemical, physical, and toxicological information. The use of all information must be reflected and defined in the state regulations to maximize its utility.

Biological Information and Standardized Methods

Information derived through biological monitoring is useful only if it is collected and analyzed using standard scientific methods. These methods must be adequately tested for reproducibility independent of the collector, provide a good census of stream community, and produce information that can be related to water quality. Method selection also requires consideration of program needs and available resources. Statistical indices and other analytical tools are in constant review and development. Therefore the data base generated should be consistent, additive, and flexible to be utilized for immediate and long-term needs.

The North Carolina Program

These requirements faced macroinvertebrate ecologists in North Carolina when they were employed to assess impacts to the aquatic community in free-flowing streams. Initial sampling was conducted with the U.S. Environmental Protection Agency-accepted methodology, using artificial substrata. Results were quantitative and amenable to statistical analyses, but several problems were identified with this methodology. The substrata required multiple visits, were often missing because of high flow or vandalism, were habitat specific, and were costly in time, money, and technician attrition.

A standard qualitative sampling technique was developed for wadable streams in North Carolina (Lenat, 1988). The method samples all available habitats; samples are processed in the field; one visit to the site is required; and a good census of the macroinvertebrate community is taken. Most importantly, this community changes consistently with variations in water quality. Several years were required to address within-method variability, including level of effort, collector, and site selection. Natural variability, including regional differences, watershed size, and seasonality continue to be analyzed with a data set of over 1,500 collections.

Phytoplankton ecologists also selected sampling and analytical methods best suited to program needs, which are capable of initiating and maintaining a quality data base. Logistical considerations mandated preservation of samples. An inverted microscope technique with species-level taxonomy and measured biovolumes produced estimates of density and biovolume for each sample, as well as each species within the sample. Resulting information (over 5,000 samples) has been used to identify nutrient-sensitive surface waters in North Carolina requiring additional management strategies.

Program review suggested the need for program expansion. Multiple community components and a variety of metrics enhance the capability of evaluating biological integrity in surface waters. Work has begun on method development of a North Carolina biotic index relative to fisheries community structure and habitat. Approximately 22 baseline stations will be used to test the method and metrics used for analysis. Acquisition of qualified ecologists and selection of standard methodology are the first steps toward providing biological information useful to a regulatory program. Development of criteria is dependent on the consistency with which these tools measure water quality impairment.

Statutory Authority

Biological information is an assessment tool. Impacts to biological integrity should reflect some measurable deviation from baseline or reference conditions. Establishing and maintaining a network of baseline stations capable of accounting for seasonal and regional variability is ideal, but often not feasible for each survey. In those instances upstream or paired watershed reference collections may be used.

The measurement of biological impairment as a part of the regulatory process is driven by the significance of that information relative to defined use. North Carolina adjusted its water quality regulations to take the degree of biological impairment into account (N. Carolina Dep. Environ. Health Nat. Resour. 1990). Code revisions included the anti-

degradation statement, supplemental classifications of High Quality Waters and Outstanding Resource Waters, and inclusion of protection for aquatic life propagation and maintenance in all best-use definitions. Changes also included addition of whole effluent toxicity standards and an addendum to the chlorophyll *a* standard that provides authority to prohibit or limit any discharge of waste into surface waters that would result in violations caused by nutrient enrichment. A brief overview of these adjustments follows.

■ **Antidegradation Policy.** The policy includes a statement of purpose in maintaining, protecting, and enhancing water quality within North Carolina. Existing uses shall be protected by properly classifying waters and having standards sufficient to protect these uses. The policy also includes a definition for High Quality Waters (HQW) and specific protection for waters with quality higher than the standards to prevent degradation below the quality necessary to maintain existing and anticipated uses, including uses not specified by the assigned classification. HQW are identified by the Division on a case-by-case basis. Use attainability analyses are conducted to identify waters with excellent water quality as determined by chemical and biological information. Outstanding Resource Waters (ORW) are a special subset of High Quality Waters with unique and special characteristics. Classification and protection of ORW will be discussed with other supplemental classifications. Specific point and nonpoint protection measures include:

- All new National Pollutant Discharge Elimination Systems (NPDES) wastewater discharges will have effluent limitations of BOD5 = 5 mg/L, NH_3–N = 2 mg/L and DO = 6 mg/L. More stringent limits may be set, if necessary to ensure that the discharge will not result in a drop of more than .5 mg/L of DO in the receiving waters below background levels. The total volume of treated wastewater for all discharges combined will not exceed 50 percent of the total in-stream flow under 7Q10 conditions. In general only the discharge of domestic or nonprocess wastewater will be permitted into HQW. Whole effluent toxicity is allocated to protect for chronic toxicity at an effluent concentration equal to twice that which is acceptable under design conditions.

- All expanded NPDES wastewater discharges will be required to meet the treatment described, except for those existing discharges that expand with no increase in permitted pollutant loading.

- Development activities that drain to and are within one mile of HQW are required to control runoff from a 1-inch design storm. Two options are provided related to density of development. The "low density option" for development limits single-family residences to one acre or larger lots and other developments to 12 percent. This option does not require stormwater collection systems but built-upon areas must be at least 30 feet from surface waters. The "high density option" requires stormwater control systems utilizing wet detention ponds. These systems must be installed, operated, and maintained to control runoff generated from 1-inch of rainfall from all developed areas. Systems must be sized to control runoff from all pervious surfaces draining to them. More stringent requirements may be required on a case-by-case basis with either option.

■ **Fresh Surface Waters Classifications and Standards.** Changes were made in "best usage" requirements for all fresh surface waters allowing use of ecological surveys in establishing protection strategies. Conditions related to best usage include suitability for aquatic life propagation and maintenance and restrictions on sources of water pollution that preclude any of these uses on either a short-term or long-term basis. The chlorophyll *a* standard now prohibits or limits discharge of waste into surface waters if so doing would exacerbate or result in growths of microscopic or macroscopic vegetation that would violate standards or impair best usage.

■ **Tidal Salt Water Classifications and Standards.** Standards for these waters use language similar to that for fresh waters. In addition, added protection is included for shellfishing waters, requiring resource and water quality necessary to provide shellfishing for market purposes. No sewage is allowed in shellfishing waters, which are by definition included as High Quality Waters. All waters in the 20 coastal counties have requirements for nonpoint source controls similar to those required for protection of HQW.

■ **Standards for Toxic Substances and Temperature.** Aquatic life standards limit the concentration of toxic substances to less than that which would

sessment of fisheries integrity through community structure analyses.

Summary, and the Future

North Carolina currently incorporates ecological information throughout its water quality program. Fiscal realities dictate that expanded use within the program will probably be through increased efficiency in data analysis, criteria development, and basin management. All NPDES permits within a given river basin are scheduled to be issued within the same year. Plans are well underway to adjust the ambient chemical and physical monitoring network to produce information necessary for model needs. Intensive surveys including time of travel studies, long-term BOD, and sediment oxygen demands have also changed dramatically in scope to address basin management. Benthic macroinvertebrate surveys, limnological work, and other ecological tools will shift spatial emphasis to provide assessment throughout basins prior to management plans and permit decisions. Conducting this work while meeting the other needs of the program, continued development of analytical tools, and collection of baseline information to address natural variability provide a significant challenge.

The single largest need for state programs involves data management. Accessing digitized data layers in a usable format is critical to efficient preparation of study plans that effectively analyze results to include cumulative impacts and can retrieve resultant information in formats appropriate to meet state and federal needs. Accessible geographic data layers must become a reality for maximum efficiency in state programs.

Biological information is obviously of greater utility to a regulatory agency if numerical criteria are used directly or indirectly in regulations. Benefits exist for both approaches. Administratively, numerical biological criteria simplify interpretation and enforcement. However, natural variability in aquatic environments has frustrated most attempts to standardize regulation with a numerical index. Well-constructed narrative criteria are not necessarily a perfect solution, but can provide immediate protection to surface waters. Numerical biological criteria, however used, will need to be modified with new information; caution should be taken that administrative simplicity does not circumvent the ability of qualified ecologists to assess changing situations and recommend effective action to protect water quality.

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Use of Habitat Assessment in Evaluating the Biological Integrity of Stream Communities

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ABSTRACT

An evaluation of habitat quality is critical to any assessment of ecological integrity (Plafkin et al. 1989). For streams, a holistic approach to assessing habitat quality would include an evaluation of variety and quality of substrate, channel morphology, bank structure, and riparian vegetation. Biological potential is limited by the quality of the habitat. Three general relationships between habitat quality and biological condition can be expected: (1) a direct response of the biological community to variation in the habitat quality in the absence of water quality problems; (2) a degradation of the biological community greater than habitat quality would predict, when combined with toxicant or organic pollution loadings to the stream; and (3) an artificial elevation of the biological condition beyond that predicted by habitat quality due to organic enrichment. Studies from different areas of the United States have shown that a knowledge of the habitat quality has enhanced an assessment of biological impairment caused by water quality problems. The establishment of parameters delimiting the relationship between habitat quality and biological integrity improves the ability to set environmental goals and evaluate program results. This relationship between habitat quality and biological integrity may vary among physiographic regions or ecoregions, but is determined by reference databases. Once confidence limits have been established, the reference database can be monitored to adjust for changes in habitat quality or the condition of the biological communities.

Habitat Quality and Biological Condition

Habitat assessment supports understanding of the relationship between habitat quality and biological conditions. Such assessment identifies obvious constraints on the attainable potential of the site, assists in the selection of appropriate sampling stations, and provides basic information for interpreting biosurvey results.

Assessment of biological potential is recommended as a correlate of community analysis. Variability of environmental conditions directly affects patterns of life, population, and micro- and macrogeographic distribution of organisms (Price, 1975; Smith, 1974; Cooper, 1984). Physical habitat quality is a major factor influencing the biological condition of aquatic communities. Bioassessment procedures such as the Rapid Bioassessment Protocols (RBPs) (Plafkin et al. 1989) stress the importance of this variable as a major determinant of the biological potential of a particular habitat. This potential relates to the structure and composition of the biota and must be recognized before habitat evaluations can be made.

To estimate biological potential, reference conditions are used to identify parameters of the assessment. An understanding of the expected characteristics or conditions is inherent in the judgment of impairment or degradation.

For most surface waters, baseline data were not collected prior to an impact; thus, impairment must be inferred from differences between the impact site and established references (U.S. Environ. Prot. Agency, 1990). This approach is also critical to the assessment because stream characteristics will vary dramatically across different regions (Plafkin et al. 1989). Furthermore, wide variability among streams and rivers across the country resulting from climatic, landform, and other geographic differences prevents the development of nationwide reference conditions (U.S. Environ. Prot. Agency, 1990). A range of parameters representing "best attainable" condition in terms of habitat quality and aquatic communities will be developed for ecosystems with similar physical and chemical dimensions, individual watersheds, or individual streams. A decision as to which of these reference points will be accepted as the attainable standard for the ecoregion is crucial to the bioassessment process.

Assuming that water quality remains constant, the predictable relationship between habitat quality and biological condition can be a sigmoid curve, as illustrated in Figure 1. On the x-axis, habitat is shown to vary from poor to optimal, relative to the reference conditions. Therefore, the quality of the habitat can range from zero to 100 percent of the reference, and can be categorized as nonsupporting, partially supporting, supporting, or comparable, referring to the support of well-balanced biological communities.

The curve is divided into three parts. The first, or upper right hand corner, reflects a situation with good habitat quality and good biological condition. Some variability in habitat quality is possible without affecting the condition of the biological communities. As the habitat quality decreases within some range of "good to excellent," the biological condition will remain high, and subtle differences will be difficult to detect. However, in the second, or midsectional part of the curve, the decrease in biological condition is proportional to a decrease in habitat quality. This situation occurs when habitat quality decreases, and the biological community responds with a concomitant decrease. In the lower left hand section of the curve, habitat quality is poor, and further degradation may result in relatively little difference in biological condition. Communities in this region of the curve are pollution tolerant, opportunistic, thrive in areas of re-

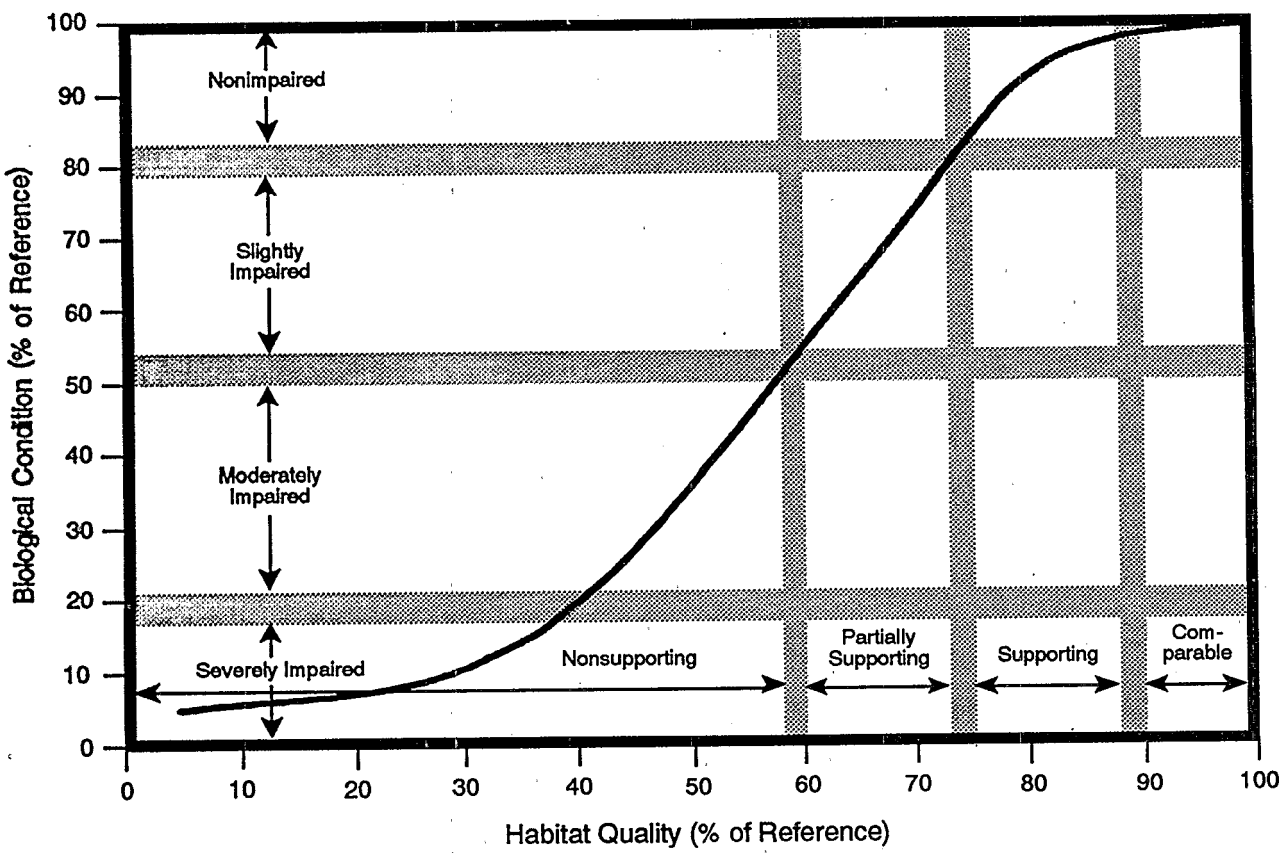

Figure 1.—The relationship between habitat and biological condition.

duced competition, and are able to withstand highly variable conditions.

Following the establishment of reference conditions for determination of water quality, habitat assessment that accounts for the various habitat parameters influencing the structure and function of communities should be conducted to enhance the interpretation of biological data (Plafkin et al. 1989). The actual orientation of the relationship line between habitat quality and biological condition is not fixed and may differ in the degree of linearity, slope, and y-intercept, depending on the physiographic region of the country. The collection of substantial reference data would allow for the development of this empirical line along with statistical parameters. From this information, the expected biological relationship can be determined from a known range of habitat quality conditions (Fig. 2). In this manner, estimates of water quality effects beyond those expected from habitat constraints are possible.

As depicted in Figure 2, three general outcomes are possible when comparing ambient stream stations to a reference: (1) no biological effects, or effects due to habitat degradation; (2) effects due to water quality; or (3) an artificial elevation of the perceived condition of the community beyond the expected relationship because of mild enrichment effects. A fourth outcome is where it is not possible to separate the combined effects of habitat and water quality degradation.

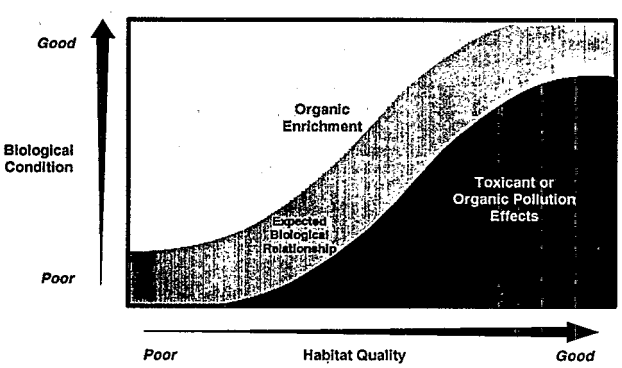

Figure 2.—Combined influence of habitat and water quality on biological condition.

The accurate determination of these possible outcomes is supported by a reference database adequate to defining the expected relationship between habitat quality and biological integrity. The theoretical regression line between habitat quality and biological condition should be substantiated with a larger database than is currently available. To date, habitat assessment results are not available in the historical database, with the possible exception of the U.S. Forest Service and the Ohio Environmental Protection Agency. Data analysis should be conducted to produce guidance on data variability expectations and the slope of the line to be used for predictions.

Establishing the reduction of habitat quality may be all that is needed to judge impairment. The quantification of habitat quality may be as important as measuring instream communities in documenting nonpoint source impact. Guidance for this type of definitive assessment needs to be developed. The following discussion provides guidance for establishing a minimum level habitat assessment.

Habitat Parameters

The habitat parameters designed to assess habitat quality are separated into three main categories: primary, secondary, and tertiary parameters. Primary parameters are those that characterize the stream "microscale" or specific niche habitats and have the greatest direct influence on the structure of the indigenous communities (Plafkin et al. 1989). The secondary parameters measure the "macroscale" habitat such as channel morphology characteristics. Tertiary parameters evaluate riparian and bank structure, features often ignored in biosurveys. These three categories are weighted according to their influence on the biota, with primary parameters having more weight than secondary or tertiary characteristics.

Although the streams across the country exhibit a wide range in variability, generalizations can be made about the types and similarities. The gradient of the streams is perhaps the most influential factor in categorizing a water body, because it is related to topography and landform, geological formations, and elevation, which in turn influence vegetation types. Four generic stream categories related to gradient can be identified: mountain, piedmont, valley/plains, and coastal. From these four categories, two sets of habitat parameters can be developed to conduct a holistic habitat assessment; these are roughly equivalent to evaluation of high gradient (riffle/run prevalence) and low gradient streams (glide/pool prevalence).

These two categorical approaches are intended to provide guidance in assessing habitat quality of two very different stream/river types based on gradient. Further subsets are possible, depending on regional specifications. However, the evaluation of habitat quality takes into consideration reference conditions that will automatically adjust for some regional differences. A mountain trout stream should not be used as a benchmark for a lowland

plains stream. Habitat parameters, which have been selected to support the assessment approach for the two general stream type categories, include primary, secondary, and tertiary characteristics. These are:

RIFFLE/RUN PREVALENCE

PRIMARY — Substrate, Instream Cover, and Canopy
1. Substrate variety/instream cover
2. Embeddedness
3. Flow or velocity/depth
4. Canopy cover (shading)

SECONDARY — Channel Morphology
5. Channel alteration
6. Bottom scouring and deposition
7. Pool/riffle, run/bend ratio
8. Lower bank channel capacity

TERTIARY — Riparian and Bank Structure
9. Upper bank stability
10. Bank vegetative stability (grazing/disruptive pressure)
11. Streamside cover
12. Riparian vegetative zone width

GLIDE/POOL PREVALENCE

PRIMARY — Substrate, Instream Cover, and Canopy
1. Substrate variety/instream cover
2. Bottom substrate characterization
3. Pool variability
4. Canopy cover (shading)

SECONDARY — Channel Morphology
5. Channel alteration
6. Deposition
7. Channel sinuosity
8. Lower bank channel capacity

TERTIARY — Riparian and Bank Structure
9. Upper bank stability
10. Bank vegetative stability (grazing/disruptive pressure)
11. Streamside cover
12. Riparian vegetative zone width

The categorical approach is intended to provide a refined framework for increased accuracy in, and applicability of, habitat assessment. The main differences between these two habitat assessment matrices are found in the primary parameter grouping. These parameters relate directly to the specific niche characteristics and will need to be altered depending on the stream type being evaluated. The secondary parameters differ only slightly in the specific parameter characteristics between the two matrices, and the tertiary parameters are identical. Some modification of the decision criteria might be useful to refine for regional purposes.

The Matrices

The original habitat assessment matrix presented by Plafkin et al. (1989) is based on Ball (1982) and Platts et al. (1983). Although these still make up the primary foundation for the RBP habitat assessment matrix, additional sources provide information for refinement of the habitat assessment approach.

Habitat parameters are categorized as primary, secondary, or tertiary, relating to the degree of influence exerted on the biological community. Through field observation and measurement, scores are assigned to each parameter ranging from 0 (poor) to 20 (excellent) for primary, 0 to 15 for secondary, and 0 to 10 for tertiary parameters. A more complete explanation of the scoring procedures for performing a habitat assessment is provided in Plafkin et al. (1989). A description of the parameters for the two categorical approaches is presented in the following section.

Riffle/Run Prevalence

The first matrix (Fig. 3) is similar to the original described in Plafkin et al. (1989), but has been modified to be more appropriate for wadable streams and rivers having a prevalence of riffles and runs. These primary habitat parameters are weighted the highest to reflect their degree of importance to the biological community. The primary parameters relate to substrate and instream characteristics and are:

■ **1. Bottom substrate/instream cover.** This characteristic refers to the availability of habitat for the support of aquatic organisms and cover for nesting, oviposition sites, or avoidance behavior. A variety of substrate materials and habitat types is desirable (U.S. Environ. Prot. Agency, 1983; Ball, 1982; Hamilton and Bergersen, 1984). The presence of broad variability in particle size of a rock/gravel substrate is considered to be the optimal habitat for benthic macroinvertebrate communities. However, instream materials such as logs and snags, tree roots, submerged and emergent vegetation, and

HABITAT ASSESSMENT FIELD DATA SHEET
RIFFLE/RUN PREVALENCE

Habitat Parameter	Category: Optimal	Sub-Optimal	Marginal	Poor
1. Bottom substrate/ instream cover (a)	Greater than 50% mix of rubble, gravel, submerged logs, undercut banks, or other stable habitat. 16–20	30–50% mix of rubble, gravel, or other stable habitat. Adequate habitat. 11–15	10–30% mix of rubble, gravel, or other stable habitat. Habitat availability less than desirable. 6–10	Less than 10% rubble, gravel, or other stable habitat. Lack of habitat is obvious. 0–5
2. Embeddedness (b)	Gravel, cobble, and boulder particles are between 0–25% surrounded by fine sediment. 16–20	Gravel, cobble, and boulder particles are between 25–50% surrounded by fine sediment. 11–15	Gravel, cobble, and boulder particles are between 50–75% surrounded by fine sediment. 6–10	Gravel, cobble, and boulder particles are over 75% surrounded by fine sediment. 0–5
3. ≤0.15 cms (5 cfs)→ Flow at rep. low	Cold >0.05 cms (2 cfs) Warm >0.15 cms (5 cfs) 16–20	0.03–0.05 cms (1–2 cfs) 0.05–0.15 cms (2–5 cfs) 11–15	0.01–0.03 cms (.5–1 cfs) 0.03–0.05 cms (1–cfs) 6–10	<0.01 cms (.5 cfs) <0.03 cms (1 cfs) 0–5
OR >0.15 cms (5 cfs)→ velocity/depth	Slow (<0.3 m/s), deep (>0.5 m): slow, shallow (<0.5 m); fast (>0.3 m/s), deep; fast, shallow habitats all present. 16–20	Only 3 of the 4 habitat categories present (missing riffles or runs receive lower score than missing pools). 11–15	Only 2 of the 4 habitat categories present (missing riffles or runs receive lower score). 6–10	Dominated by 1 velocity/depth category (usually pools). 0–5
4. Canopy cover (shading) (c) (d) (g)	A mixture of conditions where some areas of water surface fully exposed to sunlight, and other receiving various degrees of filtered light. 16–20	Covered by sparse canopy; entire water surface receiving filtered light. 11–15	Completely covered by dense canopy; water surface completely shaded OR nearly full sunlight reaching water surface. Shading limited to <3 hours per day. 6–10	Lack of canopy, full sunlight reaching water surface. 0–5
5. Channel alteration (a)	Little or no enlargement of islands or point bars, and/or no channelization. 12–15	Some new increase in bar formation, mostly from coarse gravel; and/ or some channelization present. 8–11	Moderate deposition of new gravel, coarse sand on old and new bars; and/or embankments on both banks. 4–7	Heavy deposits of fine material, increased bar development; and/or extensive channelization. 0–3
6. Bottom scouring and deposition (a)	Less than 5% of the bottom affected by scouring and/or deposition. 12–15	5–30% affected. Scour at constrictions and where grades steepen. Some deposition in pools. 8–11	30–50% affected. Deposits and/or scour at obstructions, constrictions, and bends. Filling of pools prevalent. 4–7	More than 50% of the bottom changing frequently. Pools almost absent due to deposition. Only large rocks in riffle exposed. 0–3
7. Pool/riffle, run/bend ratio (a) (distance between riffles divided by stream width)	Ratio: 5–7. Variety of habitat. Repeat pattern of sequence relatively frequent. 12–15	7–15. Infrequent repeat pattern. Variety of macrohabitat less than optimal. 8–11	15–25. Occasional riffle or bend. Bottom contours provide some habitat. 4–7	>25. Essentially a straight stream. Generally all flat water or shallow riffle. Poor habitat. 0–3
8. Lower bank channel capacity (b)	Overbank (lower) flows rare. Lower bank W/D ratio <7. (Channel width divided by depth or height of lower bank.) 12–15	Overbank (lower) flows occasional. W/D ratio 8–15. 8–11	Overbank (lower) flows common. W/D ratio 15–25. 4–7	Peak flows not contained or contained through channelization. W/D ratio >25. 0–3

Figure 3.—Habitat assessment field data sheets for riffle/run prevalent situations.

Habitat Parameter	Category: Optimal	Sub-Optimal	Marginal	Poor
9. Upper bank stability (a)	Upper bank stable. No evidence of erosion or bank failure. Side slopes generally <30°. Little potential for future problems. 9–10	Moderately stable. Infrequent, small areas of erosion mostly healed over. Side slopes up to 40° on one bank. Slight potential in extreme floods. 6–8	Moderately unstable. Moderate frequency and size of erosional areas. Side slopes up to 60° on some banks. High erosion potential during extreme high flow. 3–5	Unstable. Many eroded areas. "Raw" areas frequent along straight sections and bends. Side slopes >60° common. 0–2
10. Bank vegetative protection (d)	Over 90% of the streambank surfaces covered by vegetation. 9–10	70–89% of the streambank surfaces covered by vegetation. 6–8	50–79% of the streambank surfaces covered by vegetation. 3–5	Less than 50% of the streambank surfaces covered by vegetation. 0–2
OR Grazing or other disruptive pressure (b)	Vegetative disruption minimal or not evident. Almost all potential plant biomass at present stage of development remains. 9–10	Disruption evident but not affecting community vigor. Vegetative use is moderate, and at least one-half of the potential plant biomass remains. 6–8	Disruption obvious; some patches of bare soil or closely cropped vegetation present. Less than one-half of the potential plant biomass remains. 3–5	Disruption of streambank vegetation is very high. Vegetation has been removed to 2 inches or less in average stubble height. 0–2
11. Streamside cover (b)	Dominant vegetation is shrub. 9–10	Dominant vegetation is of tree form. 6–8	Dominant vegetation is grass or forbes. 3–5	Over 50% of the streambank has no vegetation and dominant material is soil, rock, bridge materials, culverts, or mine tailings. 0–2
12. Riparian vegetative zone width (least buffered side) (e) (f) (g)	>18 meters. 9–10	Between 12 and 18 meters. 6–8	Between 6 and 12 meters. 3–5	<6 meters. 0–2
Column Totals	____	____	____	____

Score _____

(a) From Ball 1982.
(b) From Platts et al. 1983.
(c) From EPA 1983.
(d) From Hamilton and Bergersen 1984.
(e) From Lafferty 1987.
(f) From Schueler 1987.
(g) From Bartholow 1989.

Figure 3.—Habitat assessment field data sheets for riffle/run prevalent situations (continued).

undercut banks provide exceptional habitat for a diversity of organisms, particularly fish. This parameter is evaluated by visual observation.

■ **2. Embeddedness.** This rating is a consideration of how much of the surface area of the larger substrate particles are surrounded by fine sediment (Platts et al. 1983). This parameter should allow evaluation of the substrate as a habitat for benthic macroinvertebrates and for fish spawning and egg incubation. Higher levels of embeddedness are thought to correlate with lower biotic productivity. Two aspects of embeddedness are of concern: (a) the degree to which the primary substrate, i.e., cobble, rubble, is buried in the finer sediments, and (b) the covering of the cobble with a layer of silt or organic floc. Both aspects of embeddedness will eliminate niche space and reduce attachment viability. Heavy silting (resulting in embeddedness) is known to cause a reduction in insect species diversity and productivity (Minshall, 1984). The degree of embeddedness can vary, depending on whether the riffle, run, or pool is being rated. Emphasis should be placed on the sampling area, which will normally be the riffle or run.

■ **3. Stream flow and/or stream velocity.** The size of the stream or river will influence the structure

and function of the aquatic communities. This habitat parameter rates the quality of the stream size with respect to the amount of water in small streams and the variety of velocity/depth regimes in larger streams and rivers. A particular waterbody being rated must be assigned one of these two conditions before a rating can be ascertained. The flow parameter (water quantity) indicates the ability of a stream to produce and maintain a stable environment in the substrate (Ball, 1982). This parameter is most critical to the support of aquatic communities when the representative low flow is ≤ 0.15 cubic meters per second (cms) (=5 cubic feet a second [cfs]). The evaluation is based on flow rather than velocity, since in small streams flow is the predominating constraint. In these small streams, flow should be estimated in a straight stretch of run area where banks are parallel and bottom contour is relatively flat.

In larger streams and rivers, i.e., those > 0.15 cms, velocity and depth are more important to the maintenance of aquatic communities (Osborne and Herricks, 1983; Oswood and Barber, 1982). The quality of the aquatic habitat can therefore be evaluated in terms of a velocity and depth relationship. Both factors are used to establish this parameter, with values patterned after Oswood and Barber (1982). Four general categories of velocity and depth are optimal for benthic and fish communities: (1) slow (< 0.3 m/s), shallow (< 0.5 m); (2) slow (< 0.3 m/s), deep (> 0.5 m); (3) fast (> 0.3 m/s), deep (> 0.5 m); and (4) fast (> 0.3 m/s), shallow (< 0.5 m) (Oswood and Barber, 1982). Habitat quality is reduced in the absence of one or more of these four categories. Characteristics of water current make up the major determining factors of substrate quality and, by implication, the structure and composition of benthic communities (Minshall, 1984).

■ **4. Canopy cover (shading).** The shading aspect provided by canopy cover is important in consideration of water temperature and its effect on biological processes in general, and as a mediating factor in the solar energy available for photosynthetic activity and primary production (U.S. Environ. Prot. Agency, 1983; Bartholow, 1989; Platts et al. 1983). Diversity of shade conditions is considered optimal, with different areas of the sampling station receiving direct sunlight, complete shade, and filtered light.

The secondary parameters are weighted less heavily than the primary, with a maximum score of 15 points. These characteristics relate directly to channel morphology or macrohabitat features. Local geological features, including soil character and human activities (Platts et al. 1983) influence these parameters. The sediment movement along the channel, as influenced by the tractive forces of flowing water and the sinuosity of the channel, also affects habitat conditions. The secondary characteristics are as follows:

■ **5. Channel alteration.** Sediment deposits transported from upstream and forming bars are an indication of watershed erosion or other more acute disturbances. This characteristic potentially allows crude estimation of stream system stability (Platts et al. 1983) and relates primarily to above-water deposits. Channelization involves reduction in sinuosity and results in increased velocity and subsequent intensification of erosional effects (U.S. Environ. Prot. Agency, 1983; Plafkin et al. 1989; Newbury, 1984; Schueler, 1987). Channel alteration may be caused by dredging activities, cementing or rip-rapping of banks, or natural watershed erosion. Channel alteration also results in deposition, which may occur on the inside of bends, below channel constrictions, and where stream gradient flattens out (Plafkin et al. 1989).

■ **6. Bottom scouring and deposition.** This parameter specifically targets disruption of instream habitat as a result of the channel altering factors discussed. With increases in velocity, there is more likelihood for scouring and streambed erosion. Also, scouring tends to occur during periods of increased discharge (floods), and these same areas of scour are refilled by material from further upstream (Hynes, 1970). The potential for scouring is increased by channelization. Characteristics to observe are scoured substrate and the degree of siltation in pools and riffles.

Deposition of sediment from large-scale watershed erosion can smother the instream habitat. The degree to which pools and run areas are filled with silt is an indication of the severity of this parameter. Deposition will also affect embeddedness (a primary habitat parameter). However, deposition is rated on a macrohabitat scale, and the focus is on the filling of runs and pools.

Evaluation of this parameter is by estimation of percentage of substrate affected by scouring or deposition; this should be evident from the observed degree of substrate stability in the reach of the stream being evaluated. The deposition and scouring parameter is rated by estimating the percentage of an evaluated reach that is scoured or silted (i.e., 50-m silted in a 100-m stream length equals 50 percent).

HABITAT ASSESSMENT FIELD DATA SHEET
GLIDE/POOL PREVALENCE

Habitat Parameter	Category: Optimal	Sub-Optimal	Marginal	Poor
1. Bottom substrate/ instream cover (a)	Greater than 50% mix of rubble, gravel, submerged logs, undercut banks, or other stable habitat. 16–20	30–50% mix of rubble, gravel, or other stable habitat. Adequate habitat. 11–15	10–30% mix of rubble, gravel, or other stable habitat. Habitat availability less than desirable. 6–10	Less than 10% rubble, gravel, or other stable habitat. Lack of habitat is obvious. 0–5
2. Pool substrate characterization (c)	Mixture of substrate materials with gravel and firm sand prevalent; root mats and submerged vegetation common. 16–20	Mixture of soft sand, mud, or clay; mud may be dominant; some root mats and submerged vegetation present. 11–15	All mud or clay or channelized with sand bottom; little or no root mat, no submerged vegetation. 6–10	Hard-pan clay or bedrock; no root mat or vegetation. 0–5
3. Pool variability (b) (c)	Even mix of deep/ shallow/large/small pools present. 16–20	Majority of pools large and deep; very few shallow. 11–15	Shallow pools much more prevalent than deep pools. 6–10	Majority of pools small and shallow or pools absent. 0–5
4. Canopy cover (shading) (c) (d) (g)	A mixture of conditions where some areas of water surface fully exposed to sunlight, and other receiving various degrees of filtered light. 16–20	Covered by sparse canopy; entire water surface receiving filtered light. 11–15	Completely covered by dense canopy; water surface completely shaded OR nearly full sunlight reaching water surface. Shading limited to <3 hours per day. 6–10	Lack of canopy, full sunlight reaching water surface. 0–5
5. Channel alteration (a)	Little or no enlargement of islands or point bars, and/or no channelization. 12–15	Some new increase in bar formation, mostly from coarse gravel; and/ or some channelization present. 8–11	Moderate deposition of new gravel, coarse sand on old and new bars; and/or embankments on both banks. 4–7	Heavy deposits of fine material, increased bar development; and/or extensive channelization. 0–3
6. Deposition (c)	Less than 5% of bottom affected; minor accumulation of coarse sand and pebbles at snags and submerged vegetation. 12–15	5–30% affected; moderate accumulation of sand at snags and submerged vegetation. 8–11	5–30% affected; major deposition of sand at snags and submerged vegetation; pools shallow, heavily silted. 4–7	Channelized; mud, silt, and/or sand in braided or nonbraided channels; pools almost absent due to deposition. 0–3
7. Channel sinuosity (b)	Instream channel length 3 to 4 times straight line distance. 12–15	Instream channel length 2 to 3 times straight line distance. 8–11	Instream channel length 1 to 2 times straight line distance. 4–7	Channel straight; channelized waterway. 0–3
8. Lower bank channel capacity (b)	Overbank (lower) flows rare. Lower bank W/D ratio <7. 12–15	Overbank (lower) flows occasional. W/D ratio 8–15. 8–11	Overbank (lower) flows common. W/D ratio 15–25. 4–7	Peak flows not contained or contained through channelization. W/D ratio >25. 0–3
9. Upper bank stability (a)	Upper bank stable. No evidence of erosion or bank failure. Side slopes generally <30°. Little potential for future problems. 9–10	Moderately stable. Infrequent, small areas of erosion mostly healed over. Side slopes up to 40° on one bank. Slight potential in extreme floods. 6–8	Moderately unstable. Moderate frequency and size of erosional areas. Side slopes up to 60° on some banks. High erosion potential during extreme high flow. 3–5	Unstable. Many eroded areas. "Raw" areas frequent along straight sections and bends. Side slopes >60° common. 0–2

Figure 4.—Habitat assessment field data sheets for glide/pool prevalent situations.

Habitat Parameter	Category: Optimal	Sub-Optimal	Marginal	Poor
10. Bank vegetative protection (d)	Over 90% of the streambank surfaces covered by vegetation. 9–10	70–89% of the streambank surfaces covered by vegetation. 6–8	50–79% of the streambank surfaces covered by vegetation. 3–5	Less than 50% of the streambank surfaces covered by vegetation. 0–2
OR Grazing or other disruptive pressure (b)	Vegetative disruption minimal or not evident. Almost all potential plant biomass at present stage of development remains. 9–10	Disruption evident but not affecting community vigor. Vegetative use is moderate, and at least one-half of the potential plant biomass remains. 6–8	Disruption obvious; some patches of bare soil or closely cropped vegetation present. Less than one-half of the potential plant biomass remains. 3–5	Disruption of streambank vegetation is very high. Vegetation has been removed to 2 inches or less in average stubble height. 0–2
11. Streamside cover (b)	Dominant vegetation is shrub. 9–10	Dominant vegetation is of tree form. 6–8	Dominant vegetation is grass or forbes. 3–5	Over 50% of the streambank has no vegetation and dominant material is soil, rock, bridge materials, culverts, or mine tailings. 0–2
12. Riparian vegetative zone width (least buffered side) (e) (f) (g)	>18 meters. 9–10	Between 12 and 18 meters. 6–8	Between 6 and 12 meters. 3–5	<6 meters. 0–2
Column Totals	____	____	____	____

Score _____

(a) From Ball 1982.
(b) From Platts et al. 1983.
(c) From EPA 1983.
(d) From Hamilton and Bergersen 1984.
(e) From Lafferty 1987.
(f) From Schueler 1987.
(g) From Bartholow 1989.

Figure 4.—Habitat assessment field data sheets for glide/pool prevalent situations.

A similar nonpoint source evaluation was conducted on Rock Creek, Idaho, in September 1988, using the RBPs. As in Texas, no habitat limitations were detected (Fig. 6). However, the biological community at one station (S-3) was classified as moderately impaired when compared to the reference (S-6). This level of biological condition is attributed to water quality effects.

The assessment of a point source influence wastewater treatment plant (WWTP) to Little Mill Creek, Kansas, indicated a highly degraded benthic community immediately downstream of the plant; but a recovery of the condition of the community was noted at Station 3 located approximately 1 mile downstream of the facility. In this study (Fig. 7), the habitat quality was highly comparable among all stations because of a riparian protection program implemented in Johnson County, Kansas.

The point source discharge being assessed on the North Nashua River, Massachusetts, was a small paper mill and a wastewater treatment plant. An additional complication at this site was the presence of urban runoff. A combination of habitat and water quality effects was noted from the bioassessment conducted in June 1989. Station 3 was influenced dramatically by a severe habitat degradation due to construction activities. Station 2, located less than a half mile downstream of the paper mill and treatment plant, was judged to be moderately impaired and having a supporting habitat quality. A recovery, both in terms of habitat quality and biological condition, was observed at Station 4, located approximately 6 miles downstream of the point source discharges (Fig. 8).

In these case studies, a knowledge of the variability to be expected in the relationship between

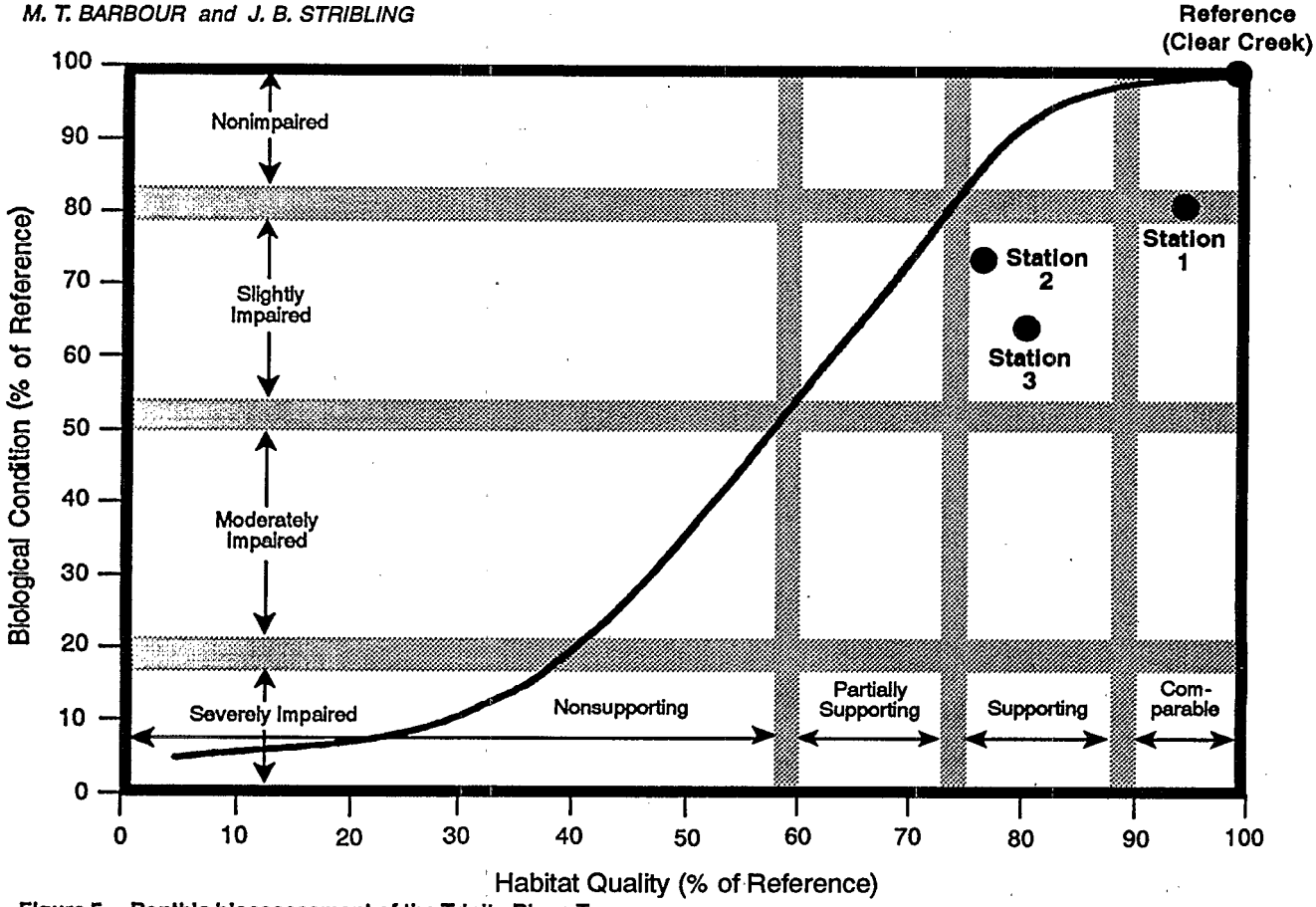

Figure 5.—Benthic bioassessment of the Trinity River, Texas.

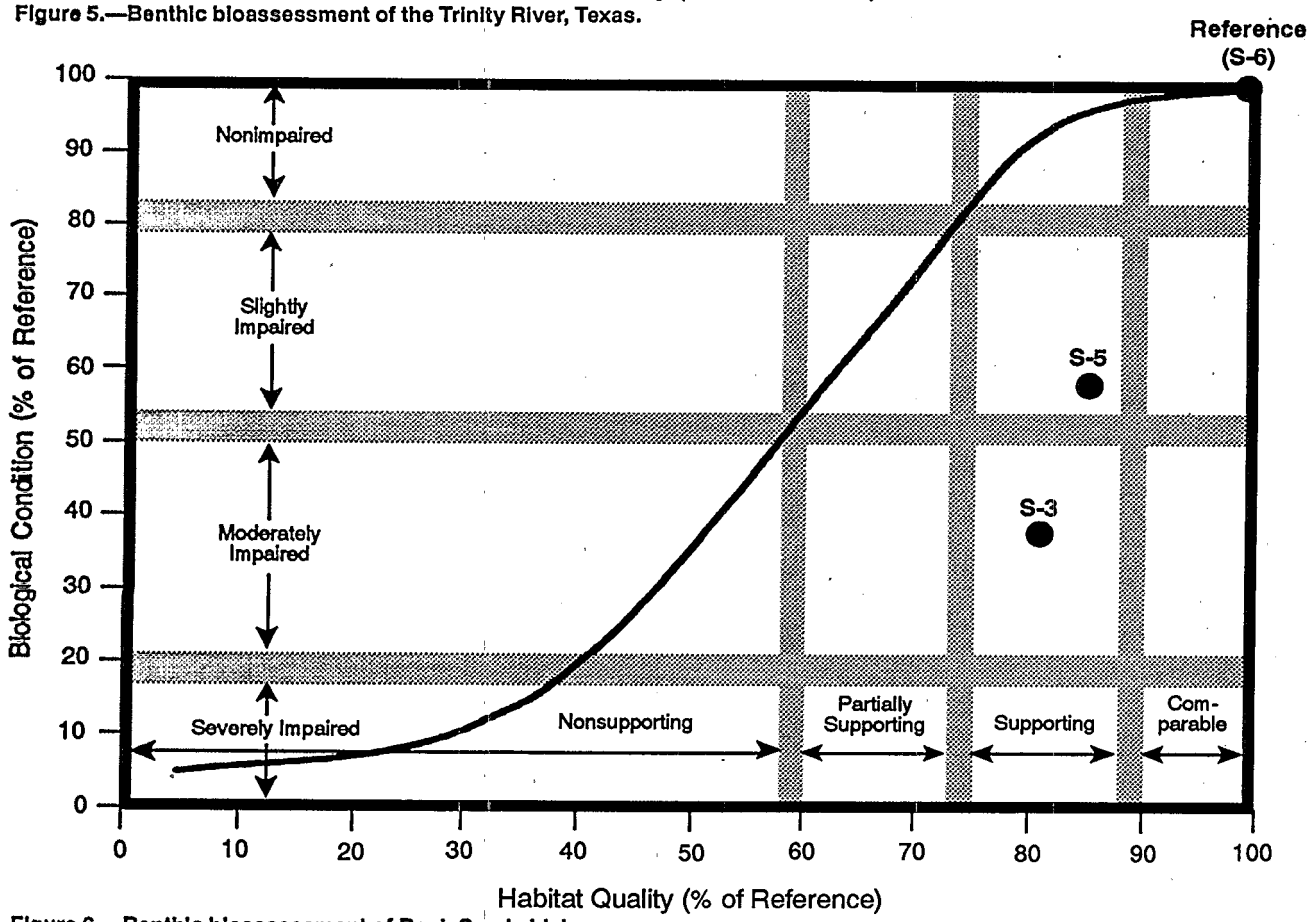

Figure 6.—Benthic bioassessment of Rock Creek, Idaho.

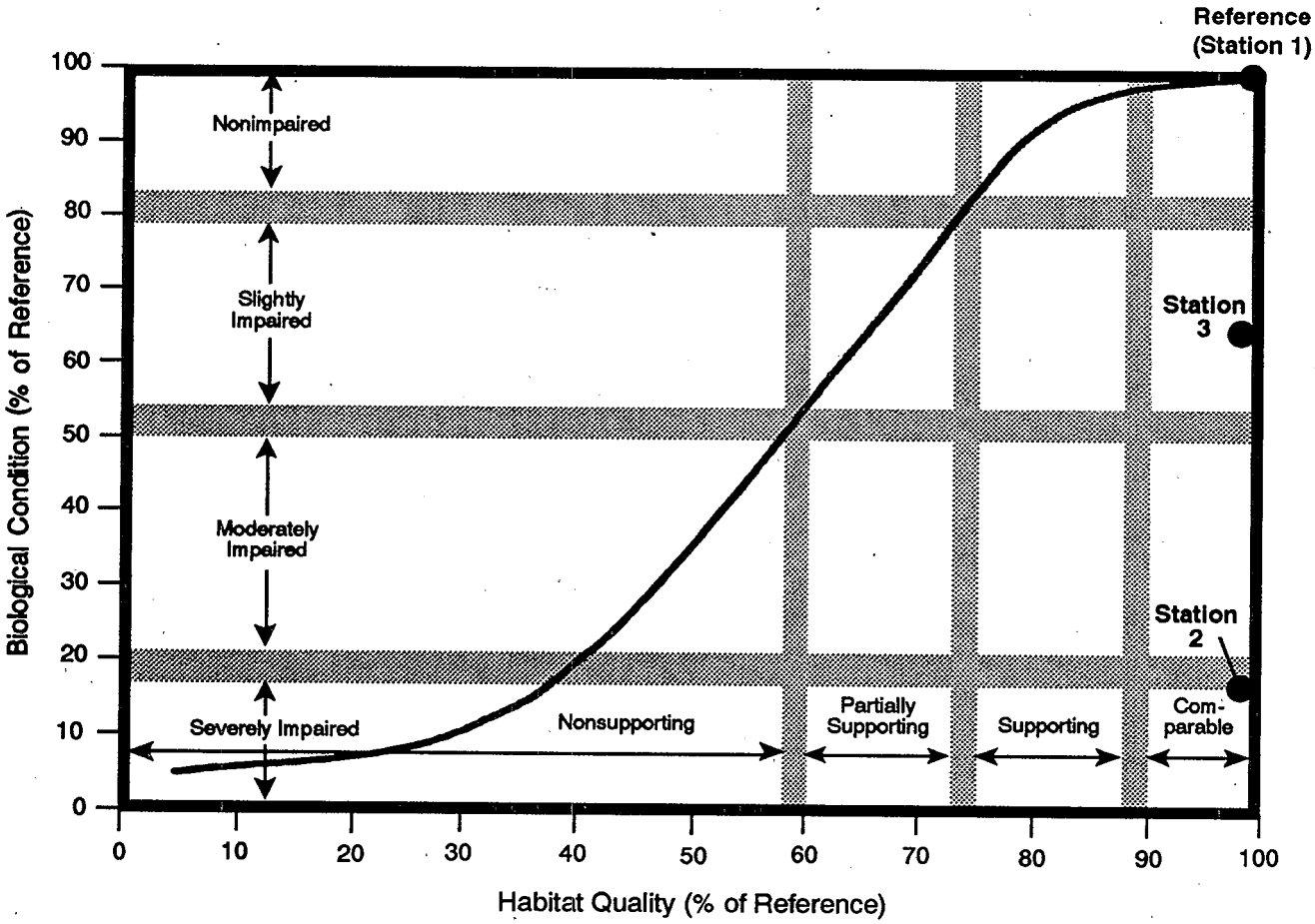

Figure 7.—Benthic bioassessment of Little Mill Creek, Kansas.

habitat quality and biological integrity would enhance interpretation of the results. An understanding of the impact of habitat degradation is critical to the assessment of the potential of a biological system. In situations where habitat has deteriorated, mitigation or improvement of the habitat through stream restoration activities should be evaluated. The implementation of water quality improvements can be independent of the habitat quality, but judgment of the improvement in biological integrity cannot.

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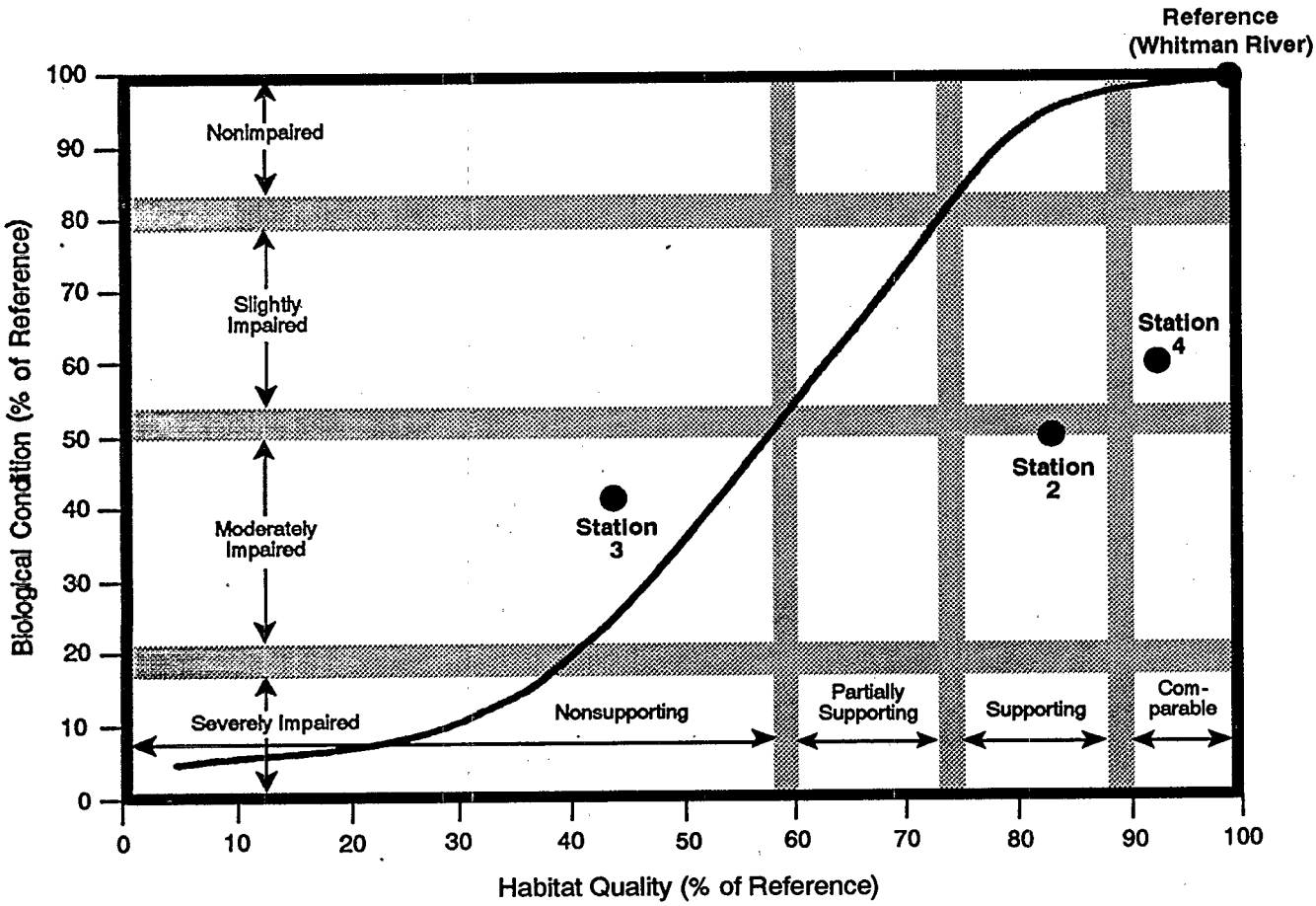

Figure 8.—Benthic bioassessment of North Nashua River, Massachusetts.

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Variability in Lakes and Reservoirs

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Variability occurs at all time and space scales and the extent and grain of the observation system determines many of the properties of the apparent variation. Grain and extent also are relevant when considering the choice of taxonomic breadth and level for biological analyses as well as when choosing the organisms with the most appropriate life span for the issue of concern. An analysis of variability, such as with coefficients of variation, can be used to help choose criteria or perhaps be criteria, in themselves, for surface waters. At the North Temperate Lake Long-term Ecological Research Site in Wisconsin, observed differences in interyear variation within lakes is related to the lake's position in the landscape rather than to its proximity to the other lake. Temporal coherence in the interyear variation among lakes is related more to their similarity in exposure to climate, i.e. surface area to mean depth ratios, than to their physical proximity. Adjacent lakes have incoherent patterns of interyear variability especially of biological properties. Knowledge of these patterns of variability can help in the design of measurements systems for evaluating surface water quality. Community analysis of fishes can be used as a water quality criterion and can be tracked through time to detect major changes in lake ecosystems.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

in the world, with average net primary production reaching 2,500 $g/m^2/yr$ (Whittaker and Likens, 1973), and it is the perpetual destruction and creation of individual wetlands within a general region that maintains long-term productivity of these systems.

In addition to high organic production in wetlands, these systems may remove inorganic, organic, and toxic substances from flowing waters, thus improving water quality. The high rate of productivity can lead to high rates of mineral uptake by vegetation and subsequent burial in sediments when the plants senesce (Sather and Smith, 1984). Denitrification, methane production, and chemical precipitation remove certain chemicals from wetland waters because of aerobic and anaerobic conditions. Reduction of stream velocity as water enters wetlands causes sediments to fall out of the water column. Because of shallow water conditions, a substantial water–sediment exchange occurs.

Unfortunately, productivity and water quality of our national wetland resources have been severely impacted through impacts on natural hydrology and alternative land use practices. Development of dams for flood control and hydropower, levees for flood protection, wetland drainage for urban, industrial, and agricultural developments, and dredging for marinas or ports have all modified wetlands across the continent (Fredrickson and Reid, 1990).

Although direct wetland conversion has been slowed in most regions, degradation continues through the alteration of flooding regimes. Changes in timing, depth, duration, or frequency of flooding causes alteration in the hydrologic cycle of wetlands. The four general categories of hydrologic alterations include: (1) stabilization, (2) shift in flood timing, (3) increased flooding, and (4) decreased flooding (Klimas, 1988). Prolonged inundation of substrates that were periodically exposed corresponds to stabilization and may involve modification of temporary, seasonal, annual, or multiyear flooding patterns.

Shifts in flood timing occur when natural flood periodicity and chronology change. Increased flooding is certainly a result of additions in flooding depth or duration, but may result from changes in timing or frequency as well. Although flood control reservoirs, levees, and drainage tiles generally decrease flooding, severe floods may still occur (Belt, 1975; Klimas, 1988; Reid et al. 1989). Any program that intends to evaluate the chemical, physical, or biological integrity of the nation's wetlands must be able to detect impacts caused by hydrologic alteration, as well as water quality modifications.

Evaluation at the System Level

As assessment criteria are developed for biological integrity of wetland waters (U.S. Environ. Prot. Agency, 1990), several habitat components need consideration. The broadest consideration is wetland type at the system level (Cowardin et al. 1979). The "system level" is defined as a complex of wetlands and deepwater habitats that share the influence of similar hydrologic, geomorphologic, chemical, or biological factors. These broad categories include:

1. **Marine:** Open ocean overlying the continental shelf and its associated high-energy coastal line.

2. **Estuarine:** Tidal wetlands that are usually semi-enclosed by land but have open, partially obstructed, or sporadic access to the ocean and in which ocean water is at least occasionally diluted by freshwater runoff from the land.

3. **Riverine:** Wetlands and deepwater habitats contained within a channel, with the exceptions of wetlands dominated by trees, shrubs, persistent emergents, emergent mosses, or lichens, or habitats with water containing ocean-derived salts in excess of 0.5 percent.

4. **Lacustrine:** Wetlands and deepwater habitats situated in a topographic depression or a dammed river channel; lacking trees, shrubs, persistent emergents, emergent mosses, or lichens with greater than 30 percent areal coverage; and with a total area exceeding 8 ha.

5. **Palustrine:** Nontidal wetlands dominated by trees, shrubs, persistent emergents, emergent mosses of lichens, and tidal wetlands where salinity resulting from ocean-derived salts is less than 5 percent (Cowardin et al. 1979).

Regional and Watershed Influences

Other habitat components that need consideration include regional and watershed influences. Certain wetlands may be confined within a single basin,

such as prairie potholes or southern playas, whereas other wetlands, such as lowland hardwood swamps, occur across an elevational gradient or mosaic. Differences in animal response occur across hydrophyte zones or plant types. One of the earliest recognized habitat relationships for aquatic invertebrates was that with aquatic plants. Hydrophyte leaf shape, structure, and surface are related to invertebrate abundance (Wieser, 1951; Rosine, 1955). Several investigators (Krecker, 1939; Andrews and Hasler, 1943; Krull, 1970) have found higher density of insects associated with aquatic plants containing highly dissected leaves. Community composition is dependent on plant condition and food habits of the invertebrates (Reid, 1985). Seasonal senescence of emergent vegetation encourages colonization by detritivore communities (Danell and Sjoberg, 1979). Annual periphyton shifts (Millie, 1979) undoubtedly influence grazer community composition. Water depth or soil moisture, water flow (stream or tidal), and type of water input (rain, headwater or backwater flooding) may all influence biological assessment. The timing of sampling should consider diurnal, tidal, or seasonal patterns.

Habitat Structure and Biological Response

Several patterns of biological response and habitat structure may be interrelated. A conceptual model has been presented for prairie wetlands, in which a dense emergent zone (*Typha* and *Scirpus* dominated) is compared to a deeper submergent zone (Nelson and Kadlec, 1984). In that example, differences existed in chemical and biological components across both spatial and temporal frameworks. For example, production:respiration (P:R), dissolved oxygen, and invertebrate production differed between these two plant zones, both in spring and summer periods.

Perhaps the potential of biological criteria assessment for wetlands can be illustrated by recent investigations in lowland hardwood swamp habitat in southeastern Missouri. Limnological investigations suggested that alteration of natural flooding regime may result in nutrient export from the wetland system (Wylie and Jones, 1986). Investigations of tree, invertebrate, and fish distributions suggested that flooding regime altered species responses and community assemblage structure (Batema et al. 1985: Finger and Stewart, 1988; Heitmeyer et al. 1989). Biological assessment identified community shifts in relation to human-induced hydrologic alterations.

If the United States is to truly "enhance and restore" wetlands, mechanisms must be found to identify degraded habitats and protect or replicate the natural hydrologic regimes for complexes of quality wetlands. Sites of historic wetlands that have been altered by other land practices should also be identified for potential restoration.

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Reference Ecosystems of the Upper Mississippi River—Past, Present and Future

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The Upper Mississippi River System (UMRS) includes the commercially navigable reaches of six large midwestern rivers. The Long Term Resource Monitoring Program for the System was established in 1986 to provide information needed by decisionmakers to maintain a balance between the System's multiple uses. Program objectives require that we know enough about the past and present ecological status of the system to be able to predict where it will be at selected points in the future. We used ideas and information from the field of large river ecology and publications on river history, populations and habitats to establish an ecological perspective of the UMRS. Five spatial scales are addressed in the perspective: watershed, stream network, floodplain reach, navigation pool, and aquatic area. The perspective describes the major abiotic and biotic factors, and natural and human-induced disturbances that operate at each scale. Each disturbance is in part defined by the time period over which it re-occurs. The perspective is being used to establish areas for research emphasis, identify required products, and develop research strategies. For instance, historical vegetation and land-use data at the floodplain reach scale are being digitized and mapped to visualize the ecological structure of the system during pre-settlement and pre-dam periods. Predictions of future UMRS ecological status will require an understanding of the relative contribution of each disturbance to the whole.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Biocriteria for Lacustrine Systems: A Case History from the Laurentian Great Lakes

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Comprehensive, long-term research and monitoring of the Laurentian Great Lakes has been underway only since the 1970's, although limnological and fisheries surveys over the past 100 years have provided important benchmarks on changes in water quality and biota. Historically, water quality monitoring has been the primary indicator of the need for water pollution control efforts, and continues to be an indicator in evaluating the effectiveness of implemented pollution control programs. Phytoplankton, zooplankton, benthos, fish, and fish-eating wildlife have been variously used as indicators with emphasis on biological function, community structure, and toxicity testing. Most recently, the Great Lakes community has been attempting to shift away from traditional "one-chemical-at-a-time" water quality objectives, criteria, and standards to integrative ecosystem objectives. Broadly based ecosystem objectives are being established for nearshore and offshore aquatic communities, wildlife communities dependent upon aquatic food chains, habitat, human health, and stewardship. Quantitative indicators are being developed to determine whether the ecosystem objectives are being met. This approach was first developed on Lake Superior using the lake trout (*Salvelinus namaycush)* as the indicator species. The criteria used in selecting the lake trout as the indicator of ecosystem quality are pertinent to selecting suitable indicators and reference sites elsewhere.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

The Development of Biocriteria in Marine and Estuarine Waters in Delaware

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ABSTRACT

Delaware is focusing its initial effort on developing biological criteria for marine and estuarine waters in the three inland bays within the State; Rehoboth, Indian River, and Little Assawoman bays. These areas have been selected because of high development pressure, consistently high salinities, and relatively stable biota. Benthic organisms are used as the indicator of environmental quality. The Department is evaluating the utility of this information for managing development activities in the Inland Bays. By rating or scoring the quality of the benthic community, the Department can direct development away from high quality areas and encourage development in low quality areas. This approach is similar to the Clean Water Act Section 404 Advance Identification Program used by the Environmental Protection Agency and the Corps of Engineers to identify and direct dredged and fill activities away from high quality wetlands. Two of the most important tasks in the development of biological criteria are (1) the selection of a biological collection method that is sensitive to pollution and (2) defining the condition and variability of reference sites. Data collected in the Lower Chesapeake Bay and in Rehoboth Bay shows that the selected method is sensitive to pollution, and shows greater sensitivity than other methods (e.g., biomass or diversity). The State is currently evaluating minor changes to the collection method before undertaking extensive sampling throughout the bays.

Introduction

Every two years the States must report on the status of their waters in attaining the fishable/swimmable goals of the Clean Water Act. The reporting requirements are met by determining, for each waterbody, whether State water quality standards are currently being attained. As in most States, Delaware does this by comparing water quality monitoring data with numeric water quality criteria (Del. Dep. Nat. Resour. Environ. Control, 1990a). Recently, this task has become more complex with the added emphasis on toxic pollutants in sections 303(c)(2)(B) and 304(l). of the Clean Water Act. The ultimate purpose of these assessments is to answer the simple question: "Is the water healthy enough for human consumption and aquatic life protection?

Assessments that use chemical criteria are based on the presumption that if these criteria are not exceeded, then the uses are attained. As toxics are increasingly controlled through additional chemical criteria and whole effluent toxicity testing, regulatory agencies and the public wonder if these controls have resulted in a healthy indigenous biological community of plants and animals.

Water chemistry data and criteria are powerful tools in regulating water quality. They are used to measure the pollutant removal effectiveness of treatment technologies and quality assessments of surface and ground waters. These techniques have been and will continue to be fundamental to pollution control for point sources through discharge permits.

However, our ability to determine the overall health of natural systems is limited. As the U.S. Environmental Protection Agency (EPA) and selected

States have made clear through guidance (U.S. Environ. Prot. Agency, 1990) and regulations (Ohio Environ. Prot. Agency, 1988), the best approach to assessment is an integrated one in which the strengths of each assessment tool are emphasized. Biological tools are most effective in assessing biological integrity. Where water quality problems are detected, chemical criteria are best at controlling pollution sources. Biology should not be used as the sole basis for controls, nor should water chemistry be considered the sole basis for assessment.

Numeric criteria provide a quantitative measure of performance. In a society that is driven by numbers in everything from speed limits to school grades, they seem necessary. However, the quantitative approach raises a particular dilemma for both freshwater and marine biologists—how to characterize the quality of the aquatic community numerically while recognizing the inherent complexity of natural systems. The issue is the degree to which biotic integrity can be quantified while still retaining scientific validity.

Jim Karr, who developed the Index of Biotic Integrity (IBI) (Karr et al. 1986), and others have demonstrated that numerical interpretation of natural systems can be done without sacrificing scientific validity. The IBI concept does not constitute a new approach to biological assessment. Rather, it has provided a new way of reporting the results that make it easier for biologists to communicate scientific information to regulatory agencies, the regulated community, and the public. The IBI provides a vehicle for bringing biology out of the file drawer and into the hands of decisionmakers.

Many numerically based assessment tools have been developed for marine and estuarine environments. It is up to the States to apply these tools to the management of marine and estuarine waters so that they can better answer the question: Is the water healthy?

Biocriteria Program — Delaware

Delaware is testing a numerically based biological assessment tool. This program is designed to address all types of surface waters in the State, including rivers, ditches, ponds, estuaries, and wetlands, both tidal and nontidal. Initially, it has been focused on the use of benthic invertebrates as indicators of biotic integrity.

To manage this complex task, Delaware's surface waters have been divided into four major categories that are relatively homogeneous with regard to biological conditions. This division is based on three factors: physiographic characteristics or ecoregions (Omernik, 1987), tidal influence, and sampling equipment.

These regions and the assessment strategies to be applied to them are described as follows:

- Freshwater/nontidal—piedmont ecoregion: Kick net in riffles using EPA Rapid Bioassessment Protocol III (Plafkin et al. 1989); salinity 0 ppt.
- Freshwater/nontidal—coastal plain ecoregion: D-frame net swept along banks (under development); salinity 0 ppt.
- Freshwater/tidal (under development). Salinity less than 5 ppt.
- Marine/estuarine—Depth stratified sample using box or tube cores; salinity greater than 5 ppt.

Marine and Estuarine Biocriteria Program

The program to develop biocriteria for estuarine and marine waters is initially based in the Inland Bays region of southern Delaware: the Indian River, Rehoboth, and Little Assawoman bays. This focus is in large part the result of intense development pressure in these areas as evidenced by their designation as a National Estuary Program; a 40 percent increase in population over the last 10 years; the development in 1990 of a water use plan to help manage the multiple uses of water within the watershed and the designation of the region as an outstanding water resource in State water quality standards. These designations have focused State efforts in the Inland Bays region, including nonpoint source activities under section 319 and regulated activities, including those permits for point source discharges, marina projects, and activities affecting subaqueous lands and wetlands.

The recently adopted State marina regulation (Del. Dep. Nat. Resour. Environ. Control, 1990b) has spurred the development of biological indicators in marine and estuarine systems. The regulation requires marina developments to address several living resource components: wetlands, subaqueous lands, shellfish beds, submerged aquatic vegetation, and benthic resources. The latter component requires assessment of benthic invertebrate communities using a method developed by Luckenbach, Diaz, and Schaffner (Luckenbach et al. 1988) (Fig. 1).

MARINA REGULATIONS

Benthic Resources

- "Benthic resources are protected as a matter of policy because of their importance in the food chain and their value as commercial and recreational food sources.
- The status of the benthic community must be assessed by the applicant using frequency, diversity and abundance measures approved by the Department. As a part of this determination, the rapid bioassessment techniques of Luckenbach, Diaz and Schaffner (1989) will be used by the Department to characterize benthic communities. Taxonomic and biomass data specific to this methodology shall be collected. Only areas scoring 0–3, on a relative scale of 0–8, will be considered for marina siting. The Department may modify this methodology as experience is gained in applying these techniques in Delaware waters."

Figure 1.—Delaware Department of Natural Resources and Environmental Control marina regulations.
— See also Figure 2.

Delaware is in the process of testing and modifying this methodology in State estuaries. These data will be evaluated with regard to establishing numeric biocriteria in State water quality standards.

Methods

The rapid assessment technique developed by Luckenbach, Diaz, and Schaffner is based on the premise that a healthy benthic community is characterized by large, deep-dwelling organisms, primarily animals from the Annelida (worms) and Mollusca (clams) orders. A benthic community that is dominated by small animals from families that are characteristic of unstable environments is an indicator of impact or stress.

The method has been tested in the lower Chesapeake Bay and been shown to be an indicator of biotic integrity (Luckenbach et al. 1988). Sampling requires recovery of a sediment sample intact to allow sectioning with depth. The fraction in the top 5 centimeters is processed separately from the sample from 5 to 15 cm. The sample collection is rapid, requiring no more than 30 minutes at each station. The cost of lab processing is approximately $100 to $200 for each sample (both top and bottom). Numerical scores are calculated from these data and the benthic community is defined according to Figure 2.

Total Score	*Benthic Community Character*
0–1	"Poor" health, highly disturbed, early successional, poor water quality *or* other severe disturbance
2–3	"Poor" to "Fair" health, moderately disturbed, perhaps recovering community, suggestion of poor water quality
4–5	"Moderate" to "Good" health, mid-successional stage
6–8	"Good" health, undisturbed, late successional community

Figure 2.—Benthic community scoring system.

The method uses a multi-variate approach based upon three pieces of information to derive a numerical score:

- Size determination—number of animals greater than 2 cm in length;
- Taxonomic composition—number of families characteristic of stable conditions; and
- Biomass—percent of the total biomass contained below the surface of the sediment (below 5 cm).

The physical habitat quality of the sediments is also evaluated. Measurements of percent sand and percent volatile residue are made along with qualitative information on the color and texture of the sediments and the presence of submerged aquatic vegetation. Generally, the procedure is most applicable to unvegetated bottoms. Sites with submerged aquatic vegetation may require a different scoring approach. Detailed water chemistry data are not collected. Scoring is performed according to the procedures presented in Figure 3.

Data Collection — Rehoboth Bay

Three types of data were considered most important for the development of biocriteria focused on benthos: benthic community, sediment type, and salinity. A review of historical data indicated that benthic resource and sediment type data have not been collected in the Delaware's inland bays since 1970 (Maurmeyer and Carey, 1986). Because of development that has occurred in the bays over the last 20 years, additional data collection was deemed necessary. The review of historical salinity data indicates that all of Rehoboth Bay is polyhaline (greater than 25 ppt). Therefore, the benthic data collected in Rehoboth Bay will not be affected

Phase I Scores		*Score*
Fauna present below five cm?	Yes	1
	No	0
Fauna below five cm greater two cm in maximum dimension?	Yes	1
	No	0

Phase II Scores	
Species present below five cm	*Score*
Only surface dwellers present (Spionidae, Capitelidae Oligochaeta)	0
Small burrowers and commensals, (Mactridae, Nereldae, Glyceridae Nephytiidae, Polynoidae, Syllidae, Cirratulidae, Phyllodocidae, Hesionidae, Pilargidae), but not those listed below.	1
Long-lived, large fauna (Tellinidae, Veneridae, Solenidae, Chaetopteridae, Onuphidae, Maldanidae, Terebellidae, Ophioroida)	2

Phase III Scores	
% Biomass below five cm	*Score*
0 – 1	0
1 – 10	1
10 – 30	2
30 – 60	3
60 –100	4

Figure 3.—Benthic community scoring metrics.

by changes in salinity. Benthic resource data were collected at four stations in Rehoboth Bay in July 1990 (Fig. 4).

This initial sampling had two objectives. First, the sampling tested the sensitivity of the method. Two stations were chosen in areas of intense human activity and two in areas protected from human activity. The second objective was to define the spatial heterogeneity of the data and the variability of the unit sampling effort (250 sq. cm of bottom). To address this objective, three replicates were collected at each station.

Results and Discussion

The results of the scoring are presented in Table 1. The biomass and size data are presented in Table 2, while the taxonomic composition data are presented in Table 3. Several conclusions can be drawn from the data.

■ Differences between impacted and unimpacted stations were not clearly distinguished. These differences would be more clearly defined by adjusting the calculation procedures. The method may need to be regionally customized.

■ Numerical scores ranged from 5 to 8, or all in the "good" to "excellent" range. Station 4, Sally's Cove, was significantly better in quality with regard to the criteria calculations, number of sensitive families, and percent of biomass in the bottom fraction than the other sites.

■ There is insufficient data on sediment type. Additional data on sediment type throughout the bay are needed to interpret the biological data.

■ For percent biomass calculations (Table 2), there was good correlation between annelids and whole samples, except large clams were present (Station 3). Future sampling will be focused in nonshellfish areas, and biomass calculations will be made using Annelids only.

Table 1.—Rehoboth Bay scores (Stations 1–4) (as revised 9/28/90).

	PHASES			
STATIONS	I	II[1]	III[2]	SCORE
State Park (sand)				
1	2	1	4	7
1-A	2	1	4	7
1-B	2	1	3	6
				$\bar{x}$ = 6.6
Composite[3]	2	1	4	7
Marina (mud)				
2	2	2	4	8
2-A	2	1	3	6
2-B	2	1	4	7
				$\bar{x}$ = 7.0
Composite	2	2	3	7
L&R Canal (mud)				
3	2	1	3	6
3-A	2	1	3	6
3-B	2	0	3	5
				$\bar{x}$ = 5.6
Composite	2	1	3	6
Sally's Cove (sand)				
4	2	2	4	8
4-A	2	2	4	8
4-B	2	2	4	8
				$\bar{x}$ = 8.0
Composite	2	2	4	8

Note: Based on Luckenbach/Diaz/Shaffner Rapid Assessment Procedure (Luckenbach et al. 1988).

[1]Families represented by the data that resulted in a one point score included four Annelids (Cirratulaidae, Nereidae, Phyllodocidae, and Syllidae) and one Mollusc (Mactridae). Families represented by the data that resulted in a 2 point score included three Annelids (Chaetoptaridae, Maldonidae, and Onuphidae) and two molluscs (Tellenidae and Veneridae).

[2]Phase III biomass calculations were based upon Annelids only due to dominance of one Mollusc in Station 3-B sample.

[3]Calculation of a single composite value for each station, based upon composite of the data for each station.

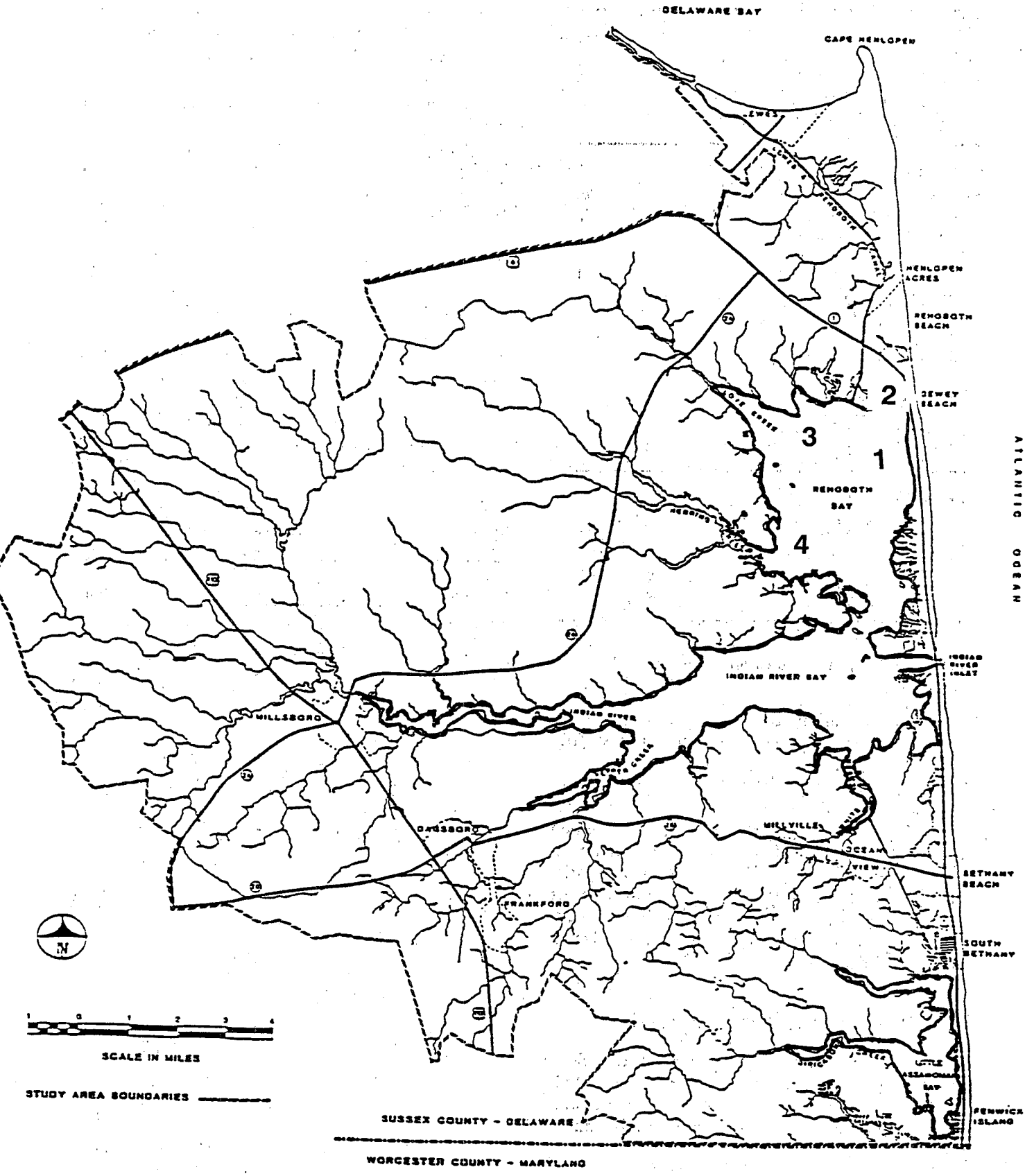

Figure 4.—Delaware Inland Bays and Rehoboth Bay sampling locations. (1) State Park; (2) Marina; (3) L&R Canal; (4) Sally's Cove.

Table 2.—Rehoboth Bay biomass data (as revised 9/28/90).

	Macroinfauna biomass as gross wet weight, and size distribution, Rehoboth Bay, July 1990									
						NO. 2 cm		% BIOMASS–BOTTOM		
	STATION	DATE	TAXON	BOTTOM	TOP	BOTTOM	TOP	ANNELIDS	WHOLE	COMPOSITE*
State Park	1	90/07/12	Annelida	0.712	0.330	9	0	68	64	
	1	90/07/12	Mollusca	0.000	0.097					
	1	90/07/12	Miscellaneous	0.070	0.007					
	1-A	90/07/12	Annelida	1.645	0.317	5	1	83	86	
	1-A	90/07/12	Arthropoda	0.000	0.001					68
	1-A	90/07/12	Mollusca	0.349	0.024					
	1-A	90/07/12	Miscellaneous	0.057	0.001					
	1-B	90/07/12	Annelida	0.501	0.425	8	2	54	53	
	1-B	90/07/12	Mollusca	0.000	0.022	22	3			
	1-B	90/07/12	Miscellaneous	0.000	0.004					
Marina	2	90/07/12	Annelida	0.748	0.450	15	4	62	61	
	2	90/07/12	Arthropoda	0.000	0.002					
	2	90/07/12	Mollusca	0.000	0.017					
	2-A	90/07/12	Annelida	0.439	0.539	5	5	45	45	
	2-A	90/07/12	Arthropoda	0.000	0.001					59
	2-A	90/07/12	Mollusca	0.000	0.005					
	2-B	90/07/12	Annelida	0.508	0.188	11	1	73	70	
	2-B	90/07/12	Arthropoda	0.002	0.012	31	10			
	2-B	90/07/12	Mollusca	0.000	0.013					
	2-B	90/07/12	Echinodermata	0.000	0.001					
L & R Canal	3	90/07/12	Annelida	0.169	0.114	7	0	60	48	
	3	90/07/12	Arthropoda	0.002	0.065					
	3	90/07/12	Mollusca	0.000	0.002					
	3	90/07/12	Miscellaneous	0.000	0.001					
	3-A	90/07/12	Annelida	0.246	0.246	1	1	50	41	53
	3-A	90/07/12	Arthropoda	0.002	0.039					
	3-A	90/07/12	Mollusca	0.000	0.078					
	3-A	90/07/12	Miscellaneous	0.001	0.000					
	3-B	90/07/12	Annelida	0.194	0.188	3	2	51	8	
	3-B	90/07/12	Arthropoda	0.003	0.066	11	3			
	3-B	90/07/12	Mollusca	0.000	2.022	*Ilyanassa obsoleta* (1 spec.)				
	3-B	90/07/12	Miscellaneous	0.000	0.007					
Sally's Cove	4	90/07/12	Annelida	1.322	0.231	6	1	85	85	
	4	90/07/12	Arthropoda	0.001	0.050					
	4	90/07/12	Mollusca	0.225	0.000					
	4-A	90/07/12	Annelida	0.658	0.149	11	0	81	82	
	4-A	90/07/12	Arthropoda	0.001	0.022					
	4-A	90/07/12	Mollusca	0.112	0.002					84
	4-B	90/07/12	Annelida	0.818	0.147	9	0	85	80	
	4-B	90/07/12	Arthropoda	0.000	0.035	26	1			
	4-B	90/07/12	Mollusca	0.020	0.021					
	4-B	90/07/12	Chironomidae	0.001	0.000					
	4-B	90/07/12	Miscellaneous	0.000	0.004					

Source: DNREC, Div. of Water Resources, Dover, 1990.
*Annelids, only.

■ There was a fair degree of spatial heterogeneity in the biomass and size distribution data. Surveys using a 3-replicate design at 250 sq. cm per replicate will continue to be conducted.

■ The method allows comparison with historical data using straight grab sampling by combining the top and bottom fractions. Therefore, the data are easily comparable with other studies using a straight grab sampling method.

Reference Conditions

It is easy to score biotic integrity numerically as shown above. It is more difficult to set the threshold or criteria for water quality standards. Criteria are needed to determine whether actions should be taken to restore degraded conditions or maintain existing quality.

The process of setting criteria in freshwater streams has used two basic approaches: regional reference streams that are determined to be "least impacted" and upstream–downstream comparisons. Clearly, an upstream–downstream approach is not applicable to marine and estuarine systems. Therefore, establishing a set of regional references is necessary.

This approach may be problematic in that it may simply define the "best of what is left" rather than what is attainable. In other words, the "best of

what is left" may be impacted when compared to conditions within a larger region. This is especially true when assessing small systems with a limited pool of reference conditions from which to choose. For example, it is difficult to say if Station 4 (Sally's Cove) in Rehoboth Bay is impacted because of large-scale development in the region.

This type of sampling bias could drastically affect the derivation of biocriteria in estuaries and alter the technical and political decisions made to manage these resources. Unfortunately, the behavior of ambient biological systems is difficult to predict. Otherwise, we could crank coefficients into a model to tell us the biological community that is attainable under various scenarios. Clearly, an empirical or observed approach is therefore necessary.

Blindly implementing controls and observing what is attainable is costly, time-consuming, and wasteful. To date, the use of "least impacted" natural systems to derive biocriteria has worked in those States (Ohio and Maine) that have developed biocriteria. When dealing with complex natural systems, we may have no choice but to strive to attain "the best of what is left." The only question that remains is the spatial scale that is used. The pool of estuaries within Delaware is clearly not large enough, while using all the estuaries in the United States does not recognize major differences in estuaries on the Atlantic, Pacific, and Gulf coasts.

The selection of references for estuaries will require a regionally coordinated approach, not only in the selection of "least impacted" sites but also in the development and use of standard data collection methods. Unfortunately, coordinating the many diverse groups involved (States, estuary programs, local governments, researchers, and academics) will not be easy.

EPA can play a vital role in facilitating this coordination. Ongoing EPA programs that could contribute include the Biocriteria Development Program, the Environmental Monitoring and Assessment Program (EMAP) (U.S. Environ. Prot. Agency, 1990b) and local programs such as the National Estuary Program and the Chesapeake Bay Program. The provinces used in EMAP, as shown in Figure 5, may provide a framework for managing the development of biocriteria for estuaries on a regional scale.

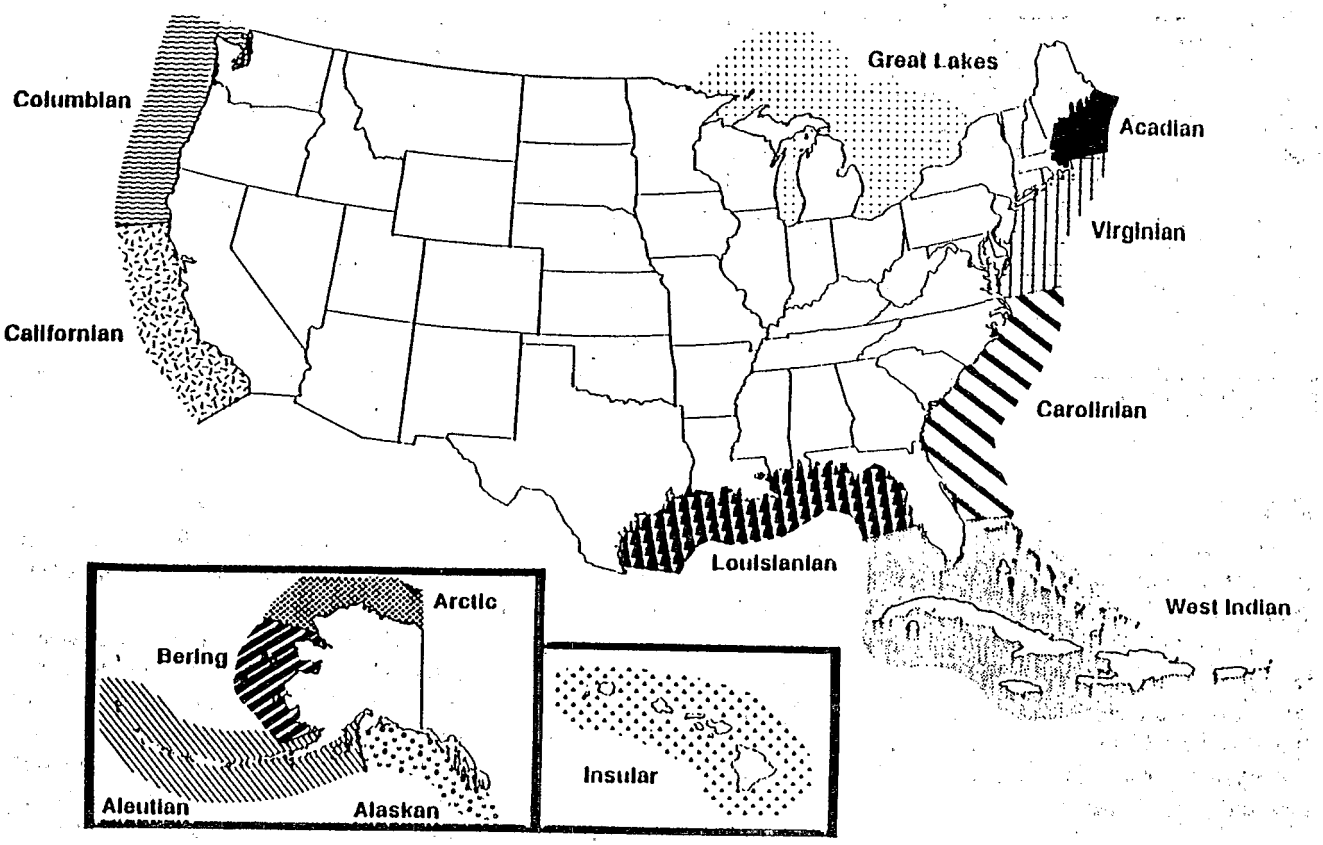

Figure 5.—EMAP Physiographic provinces.

Table 3.—Rehoboth Bay taxonomic data summary (Indicators of good/excellent quality).

RESULTS–ALL STATIONS		(BELOW 5 CM) FOUND IN REHOBOTH BAY
Annelida		
Polychaeta	(Segmented worms)	
** 1. Chaetopteridae		X
* 2. Cirratuladae		X
* 3. Glyceridae		
* 4. Hesionidae		
** 5. Maldonidae		X
* 6. Nephytidae		
* 7. Nereidae		X
** 8. Onuphidae		X
* 9. Phyllodocidae		X
* 10. Pilargidae		
* 11. Polynoidae		
* 12. Syllidae		
** 13. Terebellidae		X
Mollusca		
Pelecypoda	(Bivalves)	
* 14. Mactridae		X
** 15. Tellinidae		X
* 16. Solenidae		
** 17. Veneridae		
Echinodermata		
Ophiuroida	(Brittle stars)	
** 18. All Families		
	Total	9

RESULTS BY STATION (TOTAL NUMBER, NUMBER OF FAMILIES)
Station 1 — 7, 2
Station 2 — 8, 3
Station 3 — 2, 2
Station 4 — 23, 4

Source: DNREC, Div. of Water Resources, Dover, 1990.
*1 pt. score
**2 pt. score

The first step in this process is to draw together representatives from government, research, and academia to help standardize the collection methods and select sites for data collection, including the selection of references. In this way, data can be collected over the next several years to support the derivation of biocriteria in the future. The development of biocriteria requires a long term commitment. Through a coordinated effort, we can produce quantitative biocriteria for estuaries to help answer the question, is the estuary healthy?

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The Puget Sound Wetland Restoration Monitoring Protocol

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ABSTRACT

A systematic approach for measuring estuarine wetland function, particularly in wetland restoration and mitigation projects, has been lacking; consequently, the development of the "ecotechnology" of estuarine wetland restoration and creation has proceeded haphazardly. To remedy this situation, the Urbanized Estuary Mitigation Working Group (UEMWG) developed a protocol to quantitatively assess the function of estuarine wetlands and associated habitats for fish and wildlife. The goal of the protocol is to initiate systematic, on-site measurement of estuarine wetland function for fish and wildlife utilization by assessing attributes of the habitats identified as being functionally important to fish and wildlife. The information gathered is added to the data base on the ecotechnology of estuarine wetland construction. The protocol specifies parameters, measurement methods, and statistical evaluation criteria for assessing the level of functioning of habitats. This information provides the groundwork for development of biological criteria for evaluating the quality of estuarine wetlands in the Pacific Northwest and can be used as a benchmark for gauging effects of development and mitigation on wetlands.

Introduction

The demand for wetland restoration and creation is increasing at a rate beyond the ability of present "ecotechnology" to effectively implement or manage (Zedler, 1986). In a recent analysis of 35 projects receiving wetland development permits requiring mitigation (< 5 percent of all permits) in Washington, Kunz et al. (1988) documented that only 68 percent of the lost wetland types were replaced. This level of mitigation is in line with that occurring on a national basis (Kusler et al. 1988).

Failure to mitigate wetlands loss and damage may be attributed to two principal problems: (1) a lack of technical knowledge of wetlands structure and function; and (2) an inability of regulators and managers to uniformly assess mitigation projects and their outcome (Cooper, 1987; Kunz et al. 1988). To advance the technology of wetland construction, a relevant and scientifically sound data base that includes samples from both natural and developed systems is needed.

The protocol outlined here describes and recommends techniques for quantitatively measuring

of habitats, species, or attributes could be sampled than might have originally been recommended. Comparison between restored and natural systems must be made with caution, because natural systems have developed over long periods of time (e.g., hundreds of years for salt marshes), and even the best restored systems may not reach the reference level in a lifetime (Frenkel and Morlan, 1990).

The quality of habitats could be assessed relative to criteria (e.g., mean prey densities, seasonal dynamics in the mean prey densities) established for reference conditions in these systems. The protocol utilizes a selected set of measurable parameters that are indicators of habitat quality. These parameters are based on recommendations for assessing pollution impacts (Gray, 1981) and for determining the important members of a biological assemblage that are responsible for its structure (i.e., Paine, 1966). The procedure used to develop the present regional protocol was efficient and involved regional scientists. This latter fact increases the probability of reaching a large data base for a region and enhances the likelihood of development of a credible protocol. Because the species guilds and the levels of attributes are regionalized, development of a similar protocol for other regions would require a process similar to that used for the Pacific Northwest.

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Relationships Among Water and Sediment Contamination Toxicity and Community Responses in the Trinity River, Texas

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The community structure of the fish and benthic macroinvertebrates was measured quarterly over an 18 month period along a 200 mile section of the Trinity River in Texas. Chemical analyses of the water and sediment were conducted at each sampling station during each survey. Water and sediments were evaluated for toxicity via chronic bioassay using *Ceriodaphnia dubia* (water), fathead minnow (water and sediment), *Chironomus tentans* (sediment), Microtox and *Corbicula fluminea* (*in situ* water). A synthesis of the taxonomic analyses, water/sediment chemical data, and toxicity tests results provided insight into factors regulating faunal distributions. Probably the single greatest complicating factor in establishing associations between these parameters is the lack of habitat equality between the stations. In the absence of stress from either point or nonpoint pollutants, one might anticipate that the community structure would be the same. However, given the distance over which the stations were distributed in this study and the likelihood of finding equal habitat in the system uncomplicated by anthropogenic sources is unrealistic and, therefore, the likelihood of finding significant associations is also reduced. A rank sum analysis approach was used to evaluate the relationships between the biological, physical, and chemical metrics collected during the study. Data from this study suggest that the ranking scheme technique can provide insights on temporal and spatial relationships of point and nonpoint source impacts.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Designing Surveys to Assess Biological Integrity in Lakes and Reservoirs

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ABSTRACT

Successful approaches have been developed during the past decade for monitoring biological conditions in streams. Much less progress has been made in assessing biological conditions in lakes and reservoirs. Development of assessment approaches for these water bodies presents several challenges. These include the need to (1) identify the important physical and biological variation within and between water bodies; (2) develop field sampling methodologies that provide quality field data useful in defining ambient conditions; (3) specify reference conditions to provide expectations against which sample locations can be evaluated; and (4) develop methods of data analysis and synthesis that best reflect ambient environmental conditions. Assessment methods should be grounded in ecological principles, and be sensitive to the full range of human influences (pollution) on aquatic ecosystems; that is, they should be sensitive to degradation caused by both chemical contaminants and other impacts of human society. Using fish sampling data from the Tennessee Valley Authority, this study sought parameters that reflect basic ecological relationships in reservoirs. Our first series of metrics developed as an index of biotic integrity includes species richness and composition; fish health; reproductive guilds; individual biology; population structure; and community trophic structure. Although much work remains to be done in the development of biological assessment approaches in lakes and reservoirs, this research demonstrates that sufficient knowledge is currently available to make more informed decisions about the protection and management of these important water resources.

Background

The goal of monitoring is to accurately depict the conditions of the sample environment in an effort to assess the degree and causes of biological degradation, if any. If a balanced biological community is the expectation, then deviation from that condition could result from pollution, defined in the Clean Water Act of 1977 (PL 95-217) as "the manmade or man-induced alteration of the chemical, physical, biological, or radiological integrity of water." Thus, pollution is not narrowly defined as chemical contamination. but also includes any human action that degrades a water resource.

Chemical analysis of water samples has long been used for evaluation of water resources because of the ease of sampling and the apparent rigor conferred by sample analysis in a controlled laboratory environment. However, rigorous analytical quality does not compensate for the weaknesses of chemical sampling. Samples are representative of conditions at the sampling site for only a brief period. Transitory divergence from those conditions, such as during a runoff event or intermittent industrial release may be missed. Finally, the high level of natural variation in chemical characteristics requires that substantial changes occur before statistical inferences about change can be made.

The addition of biological monitoring does not entirely avoid these problems. However, judicious use of biological monitoring can, because of the long life cycles of individual organisms, provide a

more integrative view of the cumulative impact of many human influences. In addition, knowledge of the age, size, and trophic structure of sampled populations integrates biological conditions over periods that exceed the life of individual organisms.

Historically, studies of lake systems have included aspects of biological dynamics more often than have surveys of streams. Connections between nutrients and phytoplankton abundances in lakes have been explored by limnologists for decades, and attempts to link nutrients, phytoplankton activity (e.g., chlorophyll *a*) and lake morphoedaphic indexes are common (Hutchinson, 1967; Wetzel, 1983). Others have explored connections between abiotic factors and fish communities of lakes (Johnson et al. 1977; Tonn and Magnuson, 1982; Matuszek and Beggs, 1988). All emphasize the bottom-up determination of biological conditions. Recent evidence suggests that aquatic ecosystem dynamics are determined by a complex interaction of bottom-up and top-down regulation (i.e., limitation of target species by processes at lower trophic levels or higher trophic levels, respectively) of populations and variation in the physical environment (Tonn and Magnuson, 1982; Northcote, 1988; Karr et al. in press). Thus, knowledge of nutrient or phytoplankton abundances may not be sufficient to ensure understanding of biological condition.

Fortunately, this improved understanding of the dynamics of lakes comes at a time when society and the regulatory agencies are calling for a revolution in monitoring programs. Continuing declines in the quality and quantity of water resources despite extensive regulatory efforts demonstrate the inadequacies of existing programs (Gen. Acc. Off. 1977; Karr and Dudley, 1981; Nat. Res. Counc. 1987; U.S. Environ. Prot. Agency, 1987, 1988a,b, 1989, 1990a; Karr, 1987, 1991). Calls for restructuring of existing monitoring programs and acceleration of the development and application of promising biological monitoring techniques are common (U.S. Environ. Prot. Agency, 1987), and substantive progress is being made in defining the conceptual underpinnings of biological assessment (Karr, 1987; U.S. Environ. Prot. Agency, 1990a) and developing better methods for assessment (Karr, 1991, Karr et al. 1986; Ohio Environ. Prot. Agency, 1988; Plafkin et al. 1989; Davis, 1990).

During the past decade, successful approaches for monitoring ambient biological conditions have been developed for streams. These new assessment methodologies are based on long-established principles. Kolkwitz and Marsson (1908) demonstrated that pollution-tolerant forms replaced less tolerant species in degraded environments. Richardson (1928) noted that degradation was better indicated by tracking species composition and relative abundances than by the often dramatic changes in absolute abundances of individual species. Patrick (1950) and Cairns (1974) called for use of biological communities in assessment of water resources. Advances in streams may be useful in guiding the development of biomonitoring in lakes.

Four factors have contributed to rapid advances in biomonitoring in the last decade (Karr, 1991): (1) recognition that past approaches have not protected water resources; (2) development of integrative ecological indexes; (3) development of regional approaches to establishing ecological expectations; and (4) assessment of cumulative impacts of numerous, often small, societal activities.

An index of biotic integrity (IBI) developed nearly 10 years ago (Karr, 1981) was designed to measure the extent to which a stream fish community approximates an excellent natural community. IBI was specifically developed for biological monitoring of small- to medium-sized streams. As a result, the sampling protocol was established to account for patterns of variation in stream communities at several spatial and temporal scales, a goal made easier by the linear nature of habitat conditions—pools, riffles, and raceways—in small streams. Although the situation is more complicated in large rivers with floodplains that may include braided channels, log piles, islands, floodplain lakes, and other side channel habitats, IBI has been used successfully in large rivers as well (Hughes and Gammon, 1987; Gammon et al. 1990; Ohio Environ. Prot. Agency, 1988; Rankin and Yoder, 1990).

Effective biomonitoring depends on a sampling scheme that covers the local mosaic of habitats. With fish sampling, coverage requires sample distances that vary with stream size (Karr et al. 1986; Ohio Environ. Prot. Agency, 1988). In contrast to fish sampling, invertebrate sampling protocols often specify sampling from riffle areas only (Plafkin et al. 1989). However, some invertebrate specialists believe that sampling of invertebrates should extend to at least three habitats in locations where conservation issues predominate (Jenkins et al. 1984). Use of riffle-only collections has been challenged by Brooker (1984) and Cuff and Coleman (1979) also preferred a multihabitat sampling design. Both fish and invertebrate-based indexes are robust only so far as the sampling protocol specified provides a sample that is representative of the local biological community.

The Ohio Environmental Protection Agency (1988) adopted a protocol that modifies fish sampling methods according to stream size. This ap-

proach calls for use of backpack electrofishing or seining in small streams, small boat-mounted generators or land-based generators with long electric lines for mid-sized streams, and electrofishing equipment mounted in 12-foot to 16-foot boats for larger rivers. Specific criteria were established for each sampling method following detailed testing to ensure that representative samples are collected independent of river size and sampling method.

The general IBI approach has now been used in over 35 states, in several provinces of Canada, and in France. In addition to state efforts (Ohio Environ. Prot. Agency, 1988), a number of federal agencies have adopted IBI and its derivatives as tools for assessment of running-water resources (e.g., Plafkin et al. 1989; Hunsaker and Carpenter, 1990; Hirsch et al 1988; Saylor and Scott, 1987).

Establishing biological criteria for lakes and reservoirs presents several challenges. These include

1. gaining knowledge of the environmental factors that determine the characteristics of the resident biotic community;

2. defining the attributes of relatively undisturbed systems, especially as a function of basin size and morphometry, in much the same way that stream communities vary with stream size and valley (channel) type; and

3. developing efficient and reliable metrics to determine how and to what extent human activities influence the structure of the sampled communities.

Classifying Aquatic Ecosystems

Gaining an understanding of the ecological dynamics that characterize the water resource system is the first step in planning a sampling program to evaluate biological integrity. Two major classes of aquatic ecosystems exist: lotic (flowing water) or lentic (standing water); that is, streams and lakes.

In flowing water systems, stream morphometry and size are the dominant variables of ecological significance. At the scale of square decimeters, the upper or lower, upstream or downstream surfaces of rocks experience significant heterogeneity of flow. Along a few hundred meters of stream channel, the habitat may alternate among three major habitats—pools, raceways, and riffles. Finally, longitudinal changes occur as water flows downhill and streams fuse to form larger and larger rivers (stream order). A similar gradation occurs within standing water systems. Beginning with temporary ponds, it extends to the deepest and longest lived lakes. Basin shape and size influence the relative areas of (1) wetlands with semiaquatic and emergent macrophytes; (2) littoral zone with emergent and submerged macrophytes; and (3) pelagic or open water areas.

On long time scales, surface waters are dynamic as streams meander and lakes accumulate sediments. At shorter time scales, identifiable characteristics can be associated with a community's position along a lake or stream habitat gradient. A prominent difference between streams and lakes is that habitat gradients extend in every direction from a lake's center, while stream habitats occur in linear sequences as water flows downhill. Surface water connections are present along the size gradient in streams, while lakes of different size within a region need not share such connections. The first steps in designing an appropriate lake monitoring program are to (1) define the position along the morphometric size gradient; (2) determine the biological attributes to be sampled; and (3) select the techniques that will ensure the collection of quality data, while avoiding artifacts resulting from inconsistencies in sampling methods.

Establishing Biological Criteria for Lakes and Reservoirs

Planning for ambient biological monitoring in lakes and reservoirs requires integrating knowledge of biological patterns within those systems with understanding of the limitations of different sampling techniques. Moreover, effective evaluation of human impacts on these systems requires some knowledge of how human actions might affect those biological communities.

Preliminary application of biological monitoring to lake, reservoir, and even estuarine environments has already provided useful insights regarding monitoring programs (Greenfield and Rogner, 1984; Dionne and Karr, in press; Miller, 1988). Although superficial similarities exist among these types of water bodies, differences require unique monitoring approaches.

Lakes

Lakes are natural environments, so reference data are usually available for determining the expected condition of biological communities. As many re-

searchers have shown, lakes exhibit variation in physical and biological characteristics regionally and as a function of size and depth configuration, pH, biogeographic context, and other factors (Johnson et al. 1977; Tonn and Magnuson, 1982; Matuszek and Beggs, 1988). Size and basin morphometry are especially important because they determine the extent of wetland, littoral, and pelagic areas. Morphometry, productivity, and presence of tributary streams influence winterkill frequency, and thus duration of numerous aspects of community structure. The complex of habitats that are present affect the biotic community and success of different sampling gears.

Because habitat heterogeneity is a strong correlate of basin morphometry, lake volume, depth, and other factors embodied in the lake morphoedaphic index are critical in determining the attributes of the lake biota. Habitat heterogeneity in lakes contributes to ecosystem dynamics through the addition of species complexes associated with each habitat; heterogeneity also supports taxa that depend on the juxtaposition of two or more habitats.

Reservoirs

Examples of biological assessment of lentic waters presented here come from recent applications with reservoirs, because our experience in lakes is limited. Reservoirs differ from lakes in a number of ways that can be attributed, directly or indirectly, to control of flow regime. The primary uses of impounded waters—hydropower, flood control, and navigation (Counc. Environ. Qual. 1987; Voightlander and Poppe, 1989)—all produce unnatural variation in flow rates and water levels that have major impacts on the biota. Rapid, often extreme, changes in water level create a barren "intertidal zone" in place of the littoral vegetation and woody debris that normally provide habitat structure in lakes (Aggus, 1971; Groen and Schroeder, 1978).

The loss of littoral habitat structure also results in the loss of the plant- and debris-colonizing invertebrates that are important prey for many fish species (June, 1976; Strange et al. 1982; Crowder and Cooper, 1982; Killgore et al. 1989; Schramm and Jirka, 1989; O'Brien, 1990). Unprotected banks in turn create turbid water conditions. High discharge rates result in passage of great numbers of planktonic organisms (including pelagic fish eggs and larvae) out of the system (Cowell and Hudson, 1967; Walburg, 1971). Patterns of discharge also influence temperature and oxygen gradients both within and downstream of reservoirs. As a result, the normal seasonal stratification and turnover that typically drive natural lake processes can be highly perturbed (Wunderlich, 1971; Cole and Hannan, 1990; Ford, 1990).

Because reservoirs are created by flooding river floodplains and uplands rather than by filling a natural depression or basin, shorelines are often highly dendritic, with a high ratio of shore length to water volume in contrast to that of many lakes. This factor contributes to high turbidity in reservoirs (Kimmel et al. 1990; Marzolf, 1990; O'Brien, 1990), and may also result in greater influence of the runoff associated with various human land uses. Permanent or inconsistent flooding may destroy the tributary spawning habitats often used by riverine fishes (Richards et al. 1986; Walburg, 1977).

The contrast between reservoirs and lakes provides complementary systems for the development of monitoring schemes. In some regions, natural and impounded lakes occur in the same biogeographic areas. In these situations natural lakes may provide reference data for assessment of the overall impact of flow management on reservoir health. Recommendations for changes in water level management based on these comparisons could then be implemented and reservoir response measured. By the same token, assessment of reservoir health by comparison with healthy lakes can help identify common impacts on lakes. For example, bank erosion similar to that found in reservoirs can result from lakeside forestry, agriculture, housing, urban development, and recreation. Loss of lake habitat structure through removal of aquatic plants and woody debris to improve aesthetic or recreational appeal also creates reservoir-like conditions.

Reservoir IBI

Initial efforts to develop an IBI for Tennessee Valley Authority (TVA) reservoirs use a reservoir classification based on fish communities (McDonough and Barr, 1977) and reflect the geographic region, elevation, size, and function of the impoundments. The small, high-elevation Appalachian storage reservoirs used for flood control in the Blue Ridge Mountains and in the Upper Holston River Valley form two distinguishable classes or groups. The lower elevation "Large Storage/Upper Mainstem" (hydropower and either navigation or flood control) reservoirs form a third group. The "Lower Mainstem" reservoirs (hydropower and navigation) extending westward for 400 river miles to the confluence of the Tennessee and Ohio rivers form the final group.

The TVA has maintained a cove rotenone program since the 1940s to sample fish (Dionne and Karr, in press). The practice has been standardized

in recent years to provide reliable quantitative data. Coves are blocked off with nets, rotenone is dispersed throughout the cove, and fish are collected, identified, counted, and weighed. The result is a detailed record of species composition, abundances, and biomass for the entire fish community. This study is based on these samples for the initial exploration of reservoir assessment procedures.

Metric Development

The selection process for reservoir metrics follows the same concepts that guided the selection of IBI metrics for streams. To create an index that is sensitive to many causes of degradation, metrics must represent the extent to which fish communities diverge from an optimal condition. Efforts to reach this goal began with analyses of the TVA's cove rotenone data. The choice of metrics was based on presumed important features of the population: community and trophic structure of the reservoir fishes (Table 1). At the population and community levels, metrics were defined to measure the total numbers of individuals (Metric 13) and species (Metric 1), as well as important taxonomic (Metrics 2-3) and functional groups (Metrics 4-9). Some metrics are likely to change as the reservoir IBI is tested and refined. Species designations for tolerance, intolerance, and trophic guild required for metrics 4-8 were based on information provided by the TVA (Saylor, 1990). Details of the metrics are described in Dionne and Karr (in press).

Our selection of metrics allows evaluation of the species richness, dominant taxa, population structure, reproductive habits of reservoir fish, individual biology, fish health, and community trophic structure. For the stream IBI, most metrics are not sensitive over the entire range of degradation (Karr et al. 1986; Karr, 1991). For example, darters are found only in streams of intermediate to high quality, so they cannot reflect different levels of degradation at the low end of the quality scale. Conversely, fish with external signs of disease occur only in highly degraded systems, so the proportion of diseased fish in streams with low and intermediate levels of degradation will typically be 0 percent.

The final reservoir IBI will contain the set of metrics from each category (Species Richness and Composition, Trophic Composition, Fish Abundance and Condition, and Reproductive Composition; Table 1) that most effectively assesses the health of the ecosystem on a scale from very poor to excellent. The effectiveness of the final index will be a function of its biological sensitivity and its relative cost.

The presence of shad is an important feature of fish assemblages in reservoirs. Young-of-year (YOY) gizzard shad (*Dorosoma cepedianum*) and threadfin shad (*Dorosoma petenense*) often dominate both fish numbers and biomass, frequently comprising well above 50 percent of total fish biomass (Zeller and Wyatt, 1967; Jenkins, 1967; Noble, 1981; Downey and Toetz, 1983). In the Tennessee River mainstem reservoirs, young-of-year bluegill can be equally

Table 1.—Preliminary metrics for reservoir index of biotic integrity based on cove rotenone sampling.

	RATING CRITERIA					
	FOREBAY			INFLOW		
	5	3	1	5	3	1
Species Richness and Composition						
1. Total species number	≥33	23–32	<23	≥29	22–28	<22
2. Number of small cyprinid and darter species	≥ 6	4–5	≤ 3	≥ 5	3–4	≤ 2
3. Number of sucker species	≥ 5	3–4	≤ 2	≥ 5	3–4	≤ 2
4. Number of intolerant species	≥ 5	3–4	≤ 2	≥ 4	3	≤ 2
5. Percent individuals as tolerant species	<27	27–53	≥54	<33	33–65	≤66
Trophic Composition						
6. Percent individuals as specialized benthic insectivores*	≥8.4	4.2–8.3	≤4.1	≥ 2	1	0
7. Percent individuals as omnivores*	<27	27–53	≥54	<27	27–53	≥54
8. Percent individuals as piscivores*	≥18	9–17	< 9	≥28	14–27	<14
9. Percent individuals as YOY shad and bluegill	<33	33–63	≥64	<40	40–69	≥70
10. Percent individuals as adult shad and bluegill	<26	26–51	≥52	<27	27–53	≥54
Reproductive Composition	Will be included when data available.					
11. Percent of species as plant and rock substrate spawners						
12. Number of migratory spawning species						
Fish Abundance and Individual Health						
13. Total number of individuals**	≥14,000	7–13,999	<7,000	≥20,000	10–19,999	<10,000
14. Fish health score (TVA)	Data not availble in historical samples.					

YOY = Young-of-year.
*Excluding YOY shad and bluegill.
**Excluding shad and bluegill.
Source: Dionne and Karr, in press.

important in cove fish assemblages, although they do not share the dominant status of young-of-year shad in open water. All three species depend on zooplankton as a food source at this stage in their life cycles, although the shad can also consume phytoplankton (Scott and Crossman, 1973; DeVries and Stein, 1991). These forage species comprise a large proportion of the available prey for piscivores in some reservoirs, but large populations do not consistently lead to enhanced populations of their predators (Jenkins and Morais, 1978; Ziebell et al. 1986; DeVries and Stein, 1991).

Given their great tendency to dominate reservoir standing stock, knowledge of shad ecology is essential to understanding reservoir fish community and trophic structure, as well as to recognizing differences in the ecology of reservoirs and lakes. Recent work on ecological interactions between shad, bluegill, and largemouth bass in Ohio reservoirs containing one or the other shad species (DeVries et al. 1991; DeVries and Stein, 1991) indicate that bottom-up (competition between shad and zooplankton for phytoplankton, competition between shad and bluegill for zooplankton) and top-down processes (predation by largemouth bass on bluegill) combine to determine reservoir community structure.

The relative roles of these two processes is dictated by the timing of spawning of shad and bluegill, which determines their sequence of appearance as larvae in the limnetic zone to feed on plankton food resources, and their subsequent growth and survival. This same complex of ecological processes most likely drives systems where both gizzard shad and threadfin shad occur together. In these systems, competition for zooplankton and phytoplankton between the two shad species, size-selective predation on the two species by piscivores, and relatively frequent winterkills of threadfin shad must be added to the interactions observed by DeVries and colleagues (DeVries and Stein, 1990, 1991; DeVries et al. 1991).

If little is known about the role of shad in reservoir community structure, even less is known about their function in natural lakes. This may in part be explained by the fact that few natural lakes exist through much of these two species ranges, especially for the cold-sensitive threadfin shad. Nevertheless, the gizzard shad is found well into the latitudes of natural glacial lakes, as far north as the Great Lakes region of North America. In spite of this, we are not aware of evidence that shad regularly dominate fish standing stock in lakes. The potentially important variations in reservoir and lake ecosystems responsible for this contrast may be explained by differences in habitat structure on a scale discernable to individual fish.

Physical structure in the form of aquatic vegetation or woody debris in the littoral zone is often greatly reduced in reservoirs. This may lead to reduced success of piscivores (e.g., largemouth bass, pike, muskellunge) known to associate with structured littoral habitat (Scott and Crossman, 1973; Diana et al. 1977; Fish and Savitz, 1983; Chapman and MacKay, 1984; Savino and Stein, 1989), which in turn prevents this trophic level from achieving the numbers required to exert top-down control of reservoir shad populations. DeVries et al. (1991) suggest that competition for zooplankton between larval shad and bluegill in the limnetic zone of Stonelick Reservoir led to low survival and limited return migration of juvenile bluegill to the littoral zone, and subsequent poor growth in their largemouth bass predators. In addition to these documented interactions in the limnetic zone, loss of littoral benthic and plant-associated invertebrate communities may have a negative influence on potential shad predators and competitors.

The complex interactions between piscivores, shad, and other planktivores and invertebrate-feeding fish in natural lakes may well be mediated by littoral habitat structure and associated invertebrate productivity. Bluegill in lakes use vegetated littoral habitats as a predator refuge, and use both the littoral and open water as foraging habitats, depending on their body size, piscivore density, and habitat-specific foraging success (Mittelbach, 1981; Werner et al. 1983a,b). Small shad forage on both phytoplankton and zooplankton in open water, and large gizzard shad feed on detritus. The timing of spawning for all these species is influenced by the spring rise in water temperature, and this can determine the outcome of interactions between new year classes (DeVries et al. 1991). Perhaps the lack of littoral structure and associated invertebrate productivity in reservoirs, as well as the influence of fluctuations in water level on water temperature and available spawning habitat, all contribute to the dynamics of shad, their predators, and their competitors in these ecosystems.

Metric Rating

Defining a reference site is a major challenge in the ecological monitoring of reservoirs. No objectively defined, "healthy" (i.e., free from human disturbance) reference systems are available to provide benchmark values for index metrics. As a class, reservoirs differ in important ways from natural rivers and lakes, precluding the use of natural surface waters as reference sites. In the Tennessee River system, all reservoirs are subject to artificial fluctuations in water level and flow regime, and

many have been exposed to substantial point and nonpoint sources of pollution, especially on the mainstem. Thus, no set of "pristine" reservoirs is available within the system to serve as undisturbed reference sites. As understanding of Tennessee Valley impoundments develops, it may be possible to establish hypothetical reference values. For the present our approach is to generate metric values for samples from a number of ecologically similar reservoirs, and use the range of values obtained to assign high, intermediate, and low ratings (5, 3 and 1, respectively) to metric values from the study samples. This approach (Fig. 1) is modeled after the stream IBI "maximum species richness line" developed by Fausch et al. (1984). To generate IBI scores for a number of mainstem impoundments, data from the "Lower Mainstem" reservoirs were pooled. The scores are based on data collected since 1970 with standardized techniques on the lower mainstem.

A number of metrics from cove rotenone samples exhibited trends according to distance of the sample site from the dam. For example, the "total number of individuals per hectare" metric declines with distance from the dam in Chickamauga Reservoir (Dionne and Karr, in press), reflecting the gradient in physico-chemical characteristics from the more lacustrine region near the dam to the more riverine region upstream (Siler et al. 1986; Thornton, 1990). These regions are classified as forebay, inflow, and an intermediate transition zone, defined as the region where suspended sediment settles out of the water column. To account for longitudinal heterogeneity, sites were rated based on their position within the reservoir.

Implementing the Reservoir IBI

Information necessary for inclusion of the two reproductive metrics and the fish health metric are not yet available. Hence, these results are based on only 11 of the 14 proposed reservoir IBI metrics. An example of reservoir IBI scores is presented for a mainstem impoundment, Wheeler Reservoir (Fig. 2). IBI scores are more variable from year-to-year than is typical of stream systems. This variability may stem from the sampling design, and may also reflect a basic difference in temporal variation of ecological parameters between streams and impounded rivers, parallel to the trend in streams of

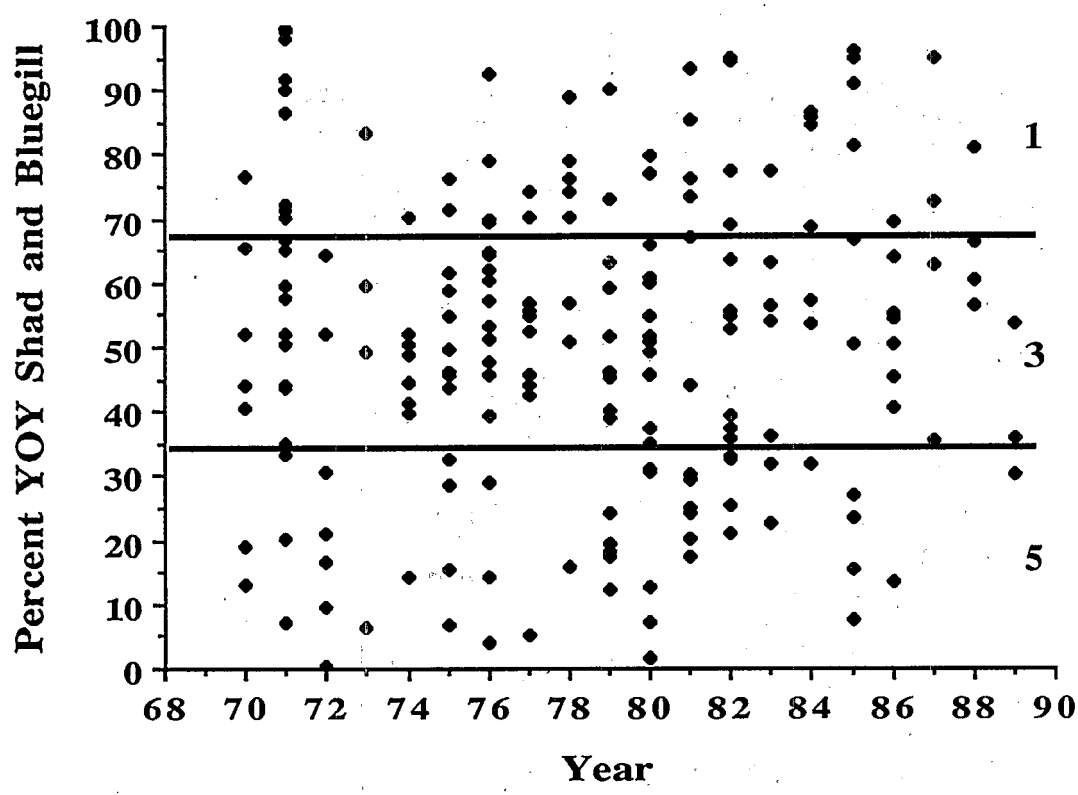

Figure 1.—An index of biotic integrity metric scatter plot for all coves sampled from 1970 in lower mainstem reservoirs of the Tennessee River. The range of possible values for the "percent of individuals as young-of-year (YOY) shad and bluegill" metric is divided into three equal intervals. Metric values within these intervals are rated as 1, 3, or 5, representing low, intermediate, and high ecological health, respectively. Plots such as this were constructed for all reservoir metrics, and used to rate metrics for individual cove samples. (Dionne and Karr, in press).

greater variability for disturbed than undisturbed sites (Karr et al. 1986; Rankin and Yoder, 1990). Perhaps the repeated (annual) destructive sampling at sites using rotenone produces perpetually unstable "young" communities; that is, these samples may represent transient assemblages analogous to an early successional plant community maintained by frequent disturbance. For these and other reasons (Dionne and Karr, in press), cove rotenone sampling does not provide the best approach for monitoring the effects of increasing human use of Tennessee River resources.

TVA Reservoir Biomonitoring

Recognizing the need for more representative sampling methods, TVA recently (since 1988) implemented a fish community assessment program using boat electroshocking, experimental gill nets, and hydroacoustics in all mainstem and selected tributary reservoirs. The electroshocking sampling regime consists of 10 timed runs (10 minutes each) along shore in the forebay, transition zone, and inflow of each impoundment, for a total of 30 samples, collected each autumn. The 10 samples are distributed among the major habitat types in each area (rip rap, rock, bluff, gravel bank, mud bank, submerged brush, and vegetation). Because electroshocking is nondestructive, samples can be collected along developed shoreline, and direct assessment of the influence of local human disturbance on the aquatic resource is possible.

The IBI metrics developed from the cove rotenone data should be readily modified for use with the new electroshocking data. However, because qualitative and quantitative differences exist between cove rotenone and electroshocking samples, the IBI scores from the two sampling methods are not directly comparable. Electroshocking samples are biased because some fish species and size classes are more effectively shocked than others. However, the maximum water depth that can be sampled without specialized equipment is less than 2 m. Fish species with small body size and benthic habit are the most seriously under-represented, followed by the young-of-year of all species. Electroshocking samples are less frequently dominated by young-of-year shad because many sampling runs do not intercept their schools. Finally, electroshocking data based on timed runs (as opposed to measured runs of a set distance) do not produce fish density estimates, but only the relative abundance of fish species. Thus, all metrics based upon fish densities must be adapted for use with relative abundances. For this reason, researchers may be advised to switch to distance-based samples. Future metrics may also include information from experimental gill-netting (relative abundance of larger deep water fish species) and hydroacoustics (offshore shad densities).

As mentioned earlier, many reservoirs are characterized by a long, irregular shoreline composed of a main river channel and numerous embayments formed by flooding of tributary streams. Yet, little progress has been made in understanding hydro-

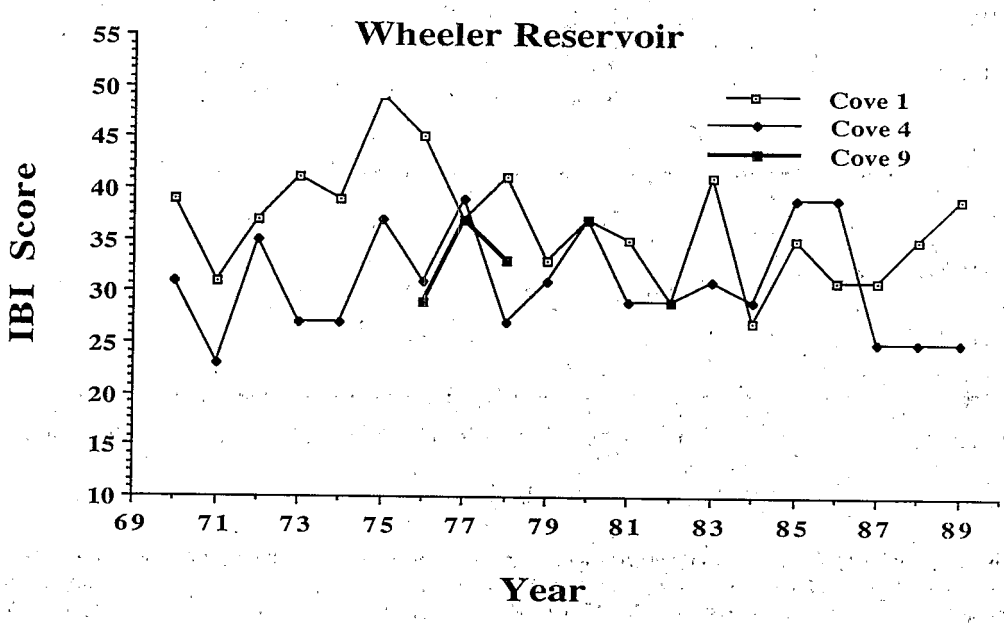

Figure 2.—Temporal variation in index of biotic integrity (IBI) scores for coves sampled repeatedly with rotenone in Wheeler

logical and ecological interactions between main channel and embayments. Because a large share of the human use of reservoir water resources occurs in embayments, it is important to know whether the impact of these uses remains localized, or extends from embayments into the main channel.

In the mainstem of the Tennessee River, impoundments exhibit "plug" flow, where the water of the main channel travels through the reservoir as a cohesive unit. Marked differences in temperature, chlorophyll, and turbidity levels often occur between embayments and the main channel (Baxter, 1977; Kennedy et al. 1982; Butkus, 1989). However, little is known about the contribution of each embayment to overall discharge, or about the active and passive movements of invertebrates and vertebrates between the main channel and embayments. Mixing of embayment and channel water and biota change with the seasonal hydrologic cycle. Are there periods during the year when high tributary discharge flushes water, animals, and sediments (along with the existing pollutant load) into the main channel? How often and under what conditions do water, animals, and sediment from the main channel back up into embayments?

These mixing processes influence the impact of human use on overall reservoir health. Similarly, knowledge of the interactions of wetland, littoral, and pelagic zones in lakes is basic to understanding how human activities affect lake or reservoir ecological health.

Development of Lake IBI

An index designed to measure the biotic integrity of lakes can be constructed according to the general scheme outlined for reservoirs. Because the ecology of a reservoir is in some ways like that of a disturbed lake, the type of metrics included in a lake IBI would be similar to those developed for reservoirs. However, the details of the lake IBI metrics necessarily depend upon lake size and shape, taxa of fish present, and other factors such as presence of wetland and littoral vegetation, and chemical composition. Lake and reservoir biomonitoring would benefit from a comprehensive approach combining major taxa such as fish, invertebrates, and plankton. Unfortunately, teams assessing biological integrity rarely include a broad range of taxonomic perspectives. Rather, experts in each group too frequently debate the merits of their taxon instead of working cooperatively for more integrative assessments.

As the reservoir IBI is developed and tested, extension of this approach to the assessment of natural lake systems would be useful. Much of what is being learned about reservoirs can be applied directly to lakes, and what is learned about lakes would provide an important perspective on water resource conditions created by reservoir impoundment. Following impoundment, reservoirs typically experience a rapid growth in production followed by a decline in fishery and other biotic potential (June, 1976; Kimmel and Groeger, 1986, O'Brien, 1990). IBI should be useful in tracking and identifying the factors responsible for this cycle. Such information would be instrumental in efforts to extend the period of increased production by control of water levels and flow. This is relevant because the construction of new reservoirs continues (Counc. Environ. Qual. 1987), and the future of existing reservoirs is debated at the time of relicensing.

More generally, both natural and impounded lakes are increasingly affected by human activities. For the most part, the biological consequences of these impacts remain unquantified until they reach critical levels. To determine an effective course of water resource management, development of methods for monitoring the biological response of reservoirs and lakes to human influence is essential. Only then is it possible to detect the consequences of chronic impacts and the onset of degradation in healthy systems; likewise, effective monitoring approaches are required to assess the response of degraded systems to management strategies.

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Fish Assemblages as Indicators of Environmental Quality in Chesapeake Bay

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ABSTRACT

Standard indicators of ecological integrity have not been developed for general use in estuaries. One promising indicator is based on fish assemblages, which can be sampled, identified, and interpreted with relative ease and moderate cost. By focusing on multispecies analysis, it may be possible to diminish the problems associated with great temporal variations in the occurrence of individual species, and to provide more comprehensive ecological information. An Index of Biotic Integrity (IBI) developed from long-term beach seine survey records is applicable to a wide range of salinity. The index appears responsive to temporal and spatial patterns of contaminant loads and water quality. Long-term trends and covariation in the relative abundance of 19 species of fish that are captured consistently in seine surveys were analyzed in combination with established management goals for water quality and fisheries to examine the potential of forecasting ecosystem recovery in the bay. Sampling was also expanded to several previously unmonitored tidal tributaries to further evaluate the IBI. The project examined a variety of approaches to clear interpretation, presentation, and use of fish assemblage information in environmental management of estuaries.

Introduction

The value of biological indicators and ecological measures of environmental quality is well-established, both conceptually and in practice (Karr, 1987; U.S. Environ. Prot. Agency, 1990). In nontidal aquatic systems, community properties such as diversity, relative abundance of functional or trophic groups, numerical dominance, and so forth, long have been used as evidence for anthropogenic degradation or its absence. Until recent years, the bulk of this type of analysis was confined to communities of invertebrates and microorganisms.

In the past decade, fish communities have been recognized as excellent indicators of environmental quality in freshwater streams, to the point where at least one fish community measure, the Index of Biotic Integrity, or IBI (Karr, 1981, 1987; Fausch et al. 1984; Angermeier and Karr, 1986) has been incorporated into some State water quality regulatory pro-

grams (U.S. Environ. Prot. Agency, 1990). In estuaries and marine waters, much of the effort in sampling biotic communities has been devoted to plankton and benthic invertebrates, and a large part of the analysis appears to have been devoted to understanding processes, or to comparative descriptions. Apparently, there has not been a widespread effort to evaluate community-based measures, especially of fish, and to apply them to questions of anthropogenic impacts in tidal waters.

Recognition that estuaries and coastal waters have suffered degradation of water quality, habitat loss, and concurrent losses of fisheries and other aquatic resources, has resulted in greatly increased attention to understanding and managing these systems. National and regional research and management programs have been initiated with the goal of restoring the quality of large bodies of water such as Chesapeake Bay, and protecting living aquatic resources from pollution, habitat destruction, and overharvesting. Biologically based indicators will be essential to these programs for (1) determining priority areas for management, (2) measuring the effectiveness of management actions and progress toward restoration goals, and (3) predicting the ecological consequences of management scenarios (Karr, 1987). Although States are required by Federal law to incorporate biological criteria into their water quality standards, a guidance document for estuaries probably will not be published before 1995 (U.S. Environ. Prot. Agency, 1990).

Clearly stated estuarine biological criteria are obviously essential to carry out regulatory mandates. It would also be quite useful to represent the condition of these complex ecosystems by means of a composite index or simple graphics, so that managers and nonspecialists can readily evaluate and compare information, establish goals, and set priorities for remediation. The problem is to develop concise, understandable statistics that also are ecologically meaningful, representative, reproducible, and can be generated routinely without massive investments in data collection. For estuaries and coastal waters, which are open systems with large physical, chemical, and biological gradients on various spatial and temporal scales, this is a difficult challenge.

Biological indicators of water quality have been developed based on fish assemblages of northern Chesapeake Bay. Goals for the use of these indicators are: (1) to identify ecological degradation in specific areas of the bay and its tidal tributaries, and (2) to focus environmental management programs on specific areas based on validated ecological concern.

It is hoped that this project will be helpful in establishing numerical biological criteria for estuaries. Specific goals are to: (1) explore the potential of fish assemblages as biological indicators in tidal waters of Chesapeake Bay and tributaries, (2) develop and test rapid, cost-effective techniques for biological assessments in the estuary, (3) define the reference condition and regional scales over which direct comparisons can be made; (4) develop techniques for identifying habitat factors associated with degradation; and (5) develop techniques for predicting multispecies responses to attainment of water quality and ecological restoration goals.

Methods

The area for this study is the tidal tributaries of Maryland's Chesapeake Bay (Fig. 1). The Chesapeake Bay is the focus for a multijurisdictional restoration and protection program dedicated to comprehensive management of the estuary and its living resources (Chesapeake Exec. Counc. 1989). The Chesapeake watershed supports a full range of land uses, including large urban areas, intensive agriculture, and extensive forests. Major concerns for the condition of the estuary include rapid population growth and development, eutrophication, and declining fisheries.

Salinity in northern Chesapeake Bay ranges from freshwater (< 0.5 ppt) at the head of the bay and in the upper tributaries to about 20 ppt near the Virginia border. The mainstream of the bay and the major tributaries include large, tidal, fresh oligohaline (0.5-5.0 ppt) and mesohaline (5.0-18 ppt) areas. Sampling programs are conducted in all of these salinity zones.

Data were analyzed from two somewhat different kinds of habitats: (1) major tributary areas (Nanticoke, Choptank, and Potomac Rivers and the head of the bay) sampled by the Estuarine Juvenile Finfish Survey (EJFS) (Cosden and Schaefer, 1988) and (2) small subestuaries (Severn, South, Magothy, and Wicomico Rivers) sampled by the Small Tributary Finfish Monitoring Project (STFMP). Both sampling programs are conducted by the Maryland Department of Natural Resources.

The EJFS began in 1954; sampling methods and most of the sites have been consistent since 1958. The sampling gear is a 30.5 m x 1.2 m, 6.35 mm mesh bagless seine. Two seine hauls are taken at each of 22 fixed, permanent stations (Fig. 1) in the major tributaries once a month during July, August, and September. All fish captured are identified and counted. The EJFS was designed to measure striped bass (*Morone saxatilis*) recruitment, but over the long term, the survey has provided consistent data on at

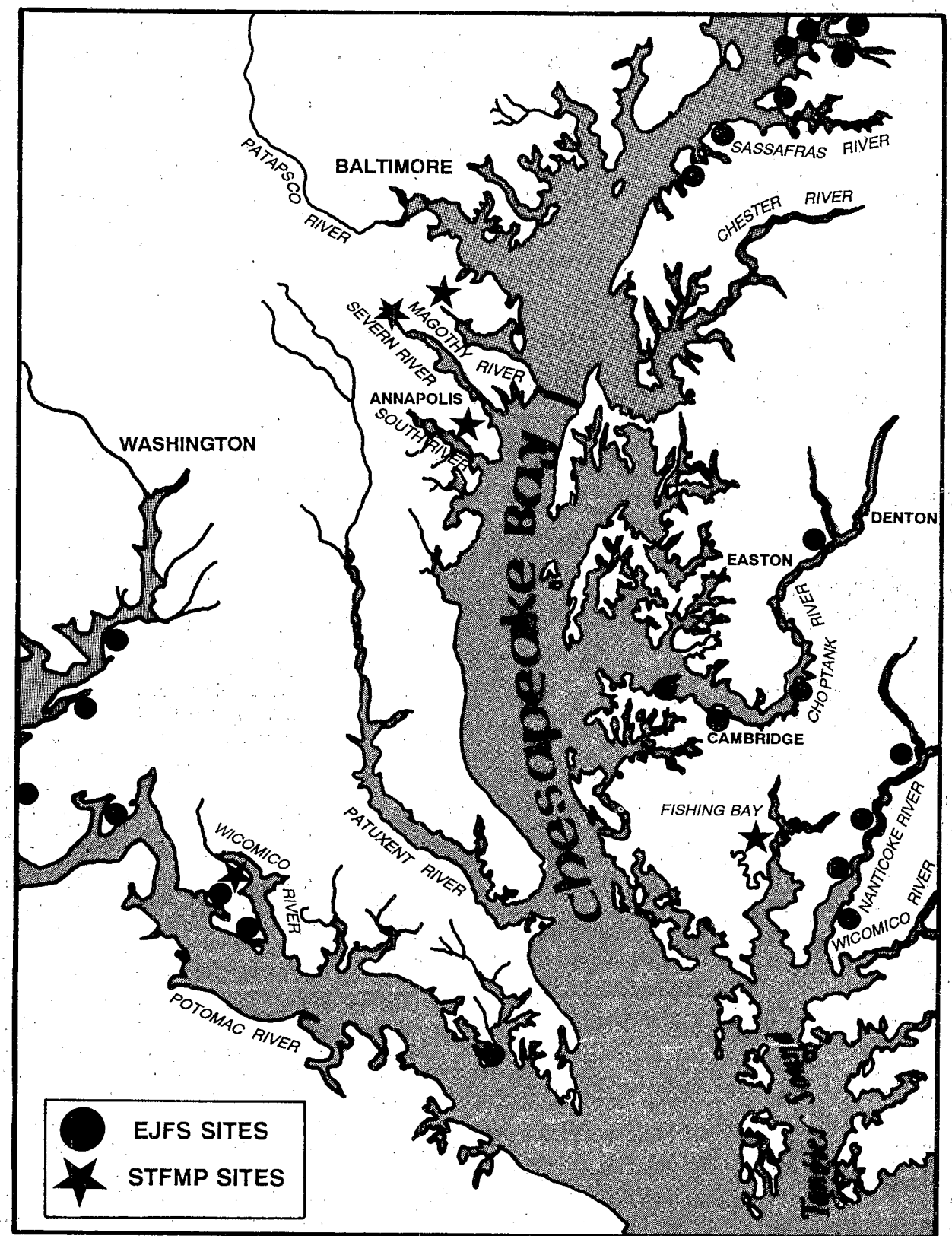


Figure 1.—Sampling locations in northern Chesapeake Bay. EJFS: Estuarine Juvenile Finfish Survey. STFMP: Small Tributary Finfish Monitoring Project. Five stations, evenly spaced along the tributary axis, are monitored in each of the STFMP tributaries.

least 19 species of fish. Salinity, temperature, and physical habitat characteristics are recorded at each site during each sampling period.

The STFMP employs the same methods as the EJFS (30.5 m beach seine, fixed stations) with the addition of midwater and bottom trawls (in channel areas) and near-shore bottom trawls at each station. Sampling areas are shown in Figure 1; five evenly spaced stations have been established along the axis of each tributary from its mouth to near the head of tide. Bottom nets are box trawls with a 3.05 m headrope; bodies and cod ends of midwater trawls have a 1.53 x 1.53 m square opening; both nets have 12.7 mm stretch mesh. All fish captured are identified and counted and the number of fish with obvious physical anomalies (lesions, damage, parasites, and so forth) is recorded. Temperature, salinity, pH, and dissolved oxygen are measured at surface, midwater, and bottom depths of the channel at each station; physical habitat characteristics are recorded at each site. A detailed habitat assessment protocol is under development for the STFMP.

The data collected have been analyzed to produce five types of information: (1) development of a prototype IBI, (2) analysis of long-term trends in species abundance and the IBI, (3) interpretation of interspecies covariation in terms of groups of species with similar sensitivities to water quality and habitat conditions, (4) graphic community "snapshots" at decade-long intervals, and (5) the potential of predicting community composition from knowledge of long-term trends and established management goals.

Index of Biotic Integrity

A prototype IBI, comprised of 12 metrics describing the fish community was developed from a 23-year subset of the EJFS data. Rather than defining a reference station, the reference condition was taken to be the upper third of the long-term distributions of each metric from seven stations in the Potomac River estuary, after adjusting for the effects of salinity. Salinity adjustments made for metrics that correlated significantly with salinity were made by removing regression equations from the data and scoring the distributions of residuals. Fish abundance data were summed over each year's sampling period (three "rounds") before computing the IBI. The IBI was evaluated for long-term trends at all 22 permanent EJFS stations, and tested by comparing STFMP data from a tributary with a relatively undeveloped watershed (Wicomico River) to data from a more degraded tributary (Severn River).

Long-term Trends

In addition to evaluating trends in the IBI, the study examined 30-year trends in relative abundance of 19 species of fish caught routinely (i.e., there were few seine samples that failed to capture at least one individual of the species) by the EJFS. Linear, quadratic, and cubic regressions of log-transformed annual mean catch per unit effort were computed against time for the whole upper bay area. The focus was on long-term trends over a large geographical area, and in similarities in temporal patterns in the abundance of species groups. The expectation was that common trends among species would reflect common responses to environmental changes related to anthropogenic stresses.

Interspecies Covariation

In addition to evaluating common trends, correlation and cluster analysis were used to estimate similarities among species. The working hypothesis was that the 19 species would be grouped into a few categories that would reflect, qualitatively, different types of tolerances. It was also expected that strong correlations between species would reflect common responses to future changes and provide a basis for predictions.

Graphic Analysis

Histograms and star charts were used to illustrate community composition at different points in time. For example, three-year means of $\log_{10}$ catch per seine haul for the 19 common species from the EJFS were graphed as histograms. For the reference period (1958–60), the species were arrayed from left to right in order of declining abundance. Later periods were shown in the same way, except that the reference period species order was maintained. This technique was also used to compare degraded areas to a reference area. Star charts were used to display essentially the same information as the histograms, except that species were color coded for greater visual impact.

Prediction

The project explored the potential for predicting community composition from knowledge of trends, species covariation, and management goals. Long-term management goals have been established for two of the species in the analysis: striped bass and American shad (*Alosa sapidissima*). Attain-

ment of these goals was assumed, and a combination of interspecies linear regressions and intervention analysis was used to develop a view of the fish community 10 years hence.

Results

A combination of two variants of the prototype IBI performed reasonably well over all of the permanent EJFS stations. It was necessary to substitute two metrics in the IBI for the freshest tidal areas (salinity < 2 ppt) because of the near absence of marine- and estuarine-spawning fish. Otherwise, the salinity adjustment approach appeared to work well. The IBI showed significant difference between the Wicomico and Severn Rivers (Fig. 2). Nine of 12 IBI metrics received higher scores in the Wicomico River "reference" tributary. The great variability of dissolved oxygen in the Severn River (Fig. 2) probably is symptomatic of the tributary's water quality problems, and may be related directly to the poorer fish community and low IBI. The IBI showed significant long-term trends at several EJFS stations; these trends appeared to be consistent with our knowledge of pollution loads over this period; however, quantitative information on loads generally was not available.

Significant 30-year trends were found for 15 of the 19 common EJFS species. Examples of trend patterns are shown in Figure 3. Pairwise species correlations and cluster analysis also identified groups of species that had similar or inverse patterns of abundance. Anadromous and semianadromous species (striped bass, white perch (*Morone americana*), alewife (*Alosa pseudoharengus*), blueback herring (*Alosa aestivalis*), and American shad) were grouped together strongly both by clustering and correlation. This group is characterized by similar life histories and sensitive early life stages, which are spawned in fresh, or nearly fresh water. The juveniles of these species (the life stage captured in these surveys) appear to be diagnostic of less degraded conditions. More anadromous species and more individuals of these species are found in the Wicomico River (the STFMP reference tributary) than in the other STFMP tributaries.

A second strong grouping, based upon correlation analysis, included gizzard shad (*Dorosoma cepedianum*), a freshwater plankton feeder, menhaden (*Brevoortia tyrannus*), a marine-spawning plankton feeder, mummichog (*Fundulus heteroclitus*), an estuarine resident species, and spot (*Leiostomus xanthurus*), a marine-spawning benthic feeder. Trend and cluster analysis grouped these latter species somewhat differently, but at least two of these

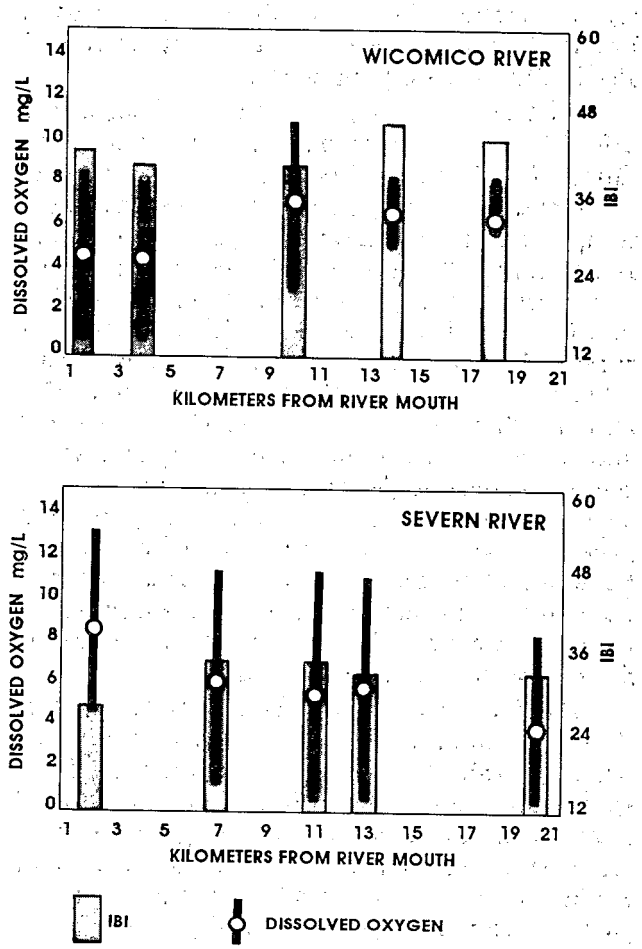


Figure 2.—Prototype Index of Biotic Integrity and dissolved oxygen (circles are means and bars are ranges) at STFMP stations in the Wicomico and Severn Rivers. Data are from the summer of 1989.

species occurred in the same group in all analyses. These species also correlated negatively with species in the anadromous group.

Graphic analysis of community structure at three periods in time showed great differences between the 1958–60 reference period and later decades (Fig. 4). The 1978–80 pattern appeared to be the most disrupted. The forecast of the community in the year 2000 (Fig. 4) showed a return to a condition more similar to the reference period than the 1978–80 or the 1987–89 periods.

Discussion

The results presented here are preliminary, and are intended only to show the potential of fish community analysis for representing temporal and spatial patterns of the quality of estuarine habitats. Con-

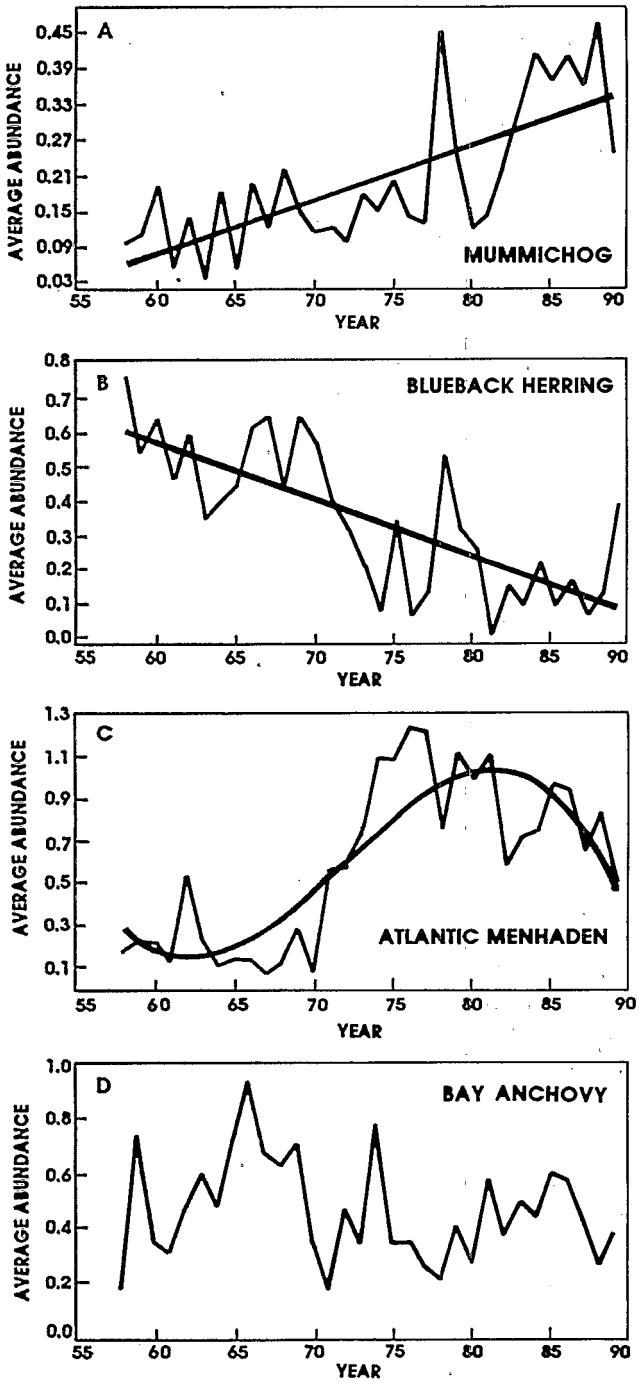


Figure 3.—Examples of trend patterns for common species captured by the EJFS. The "average abundance" is the Bay-wide (22 stations) annual mean of $\log_{10}$ catch per seine haul. A – tolerant species; B – sensitive (anadromous) species; C – pattern that may indicate eutrophication trends; D – species with no trend.

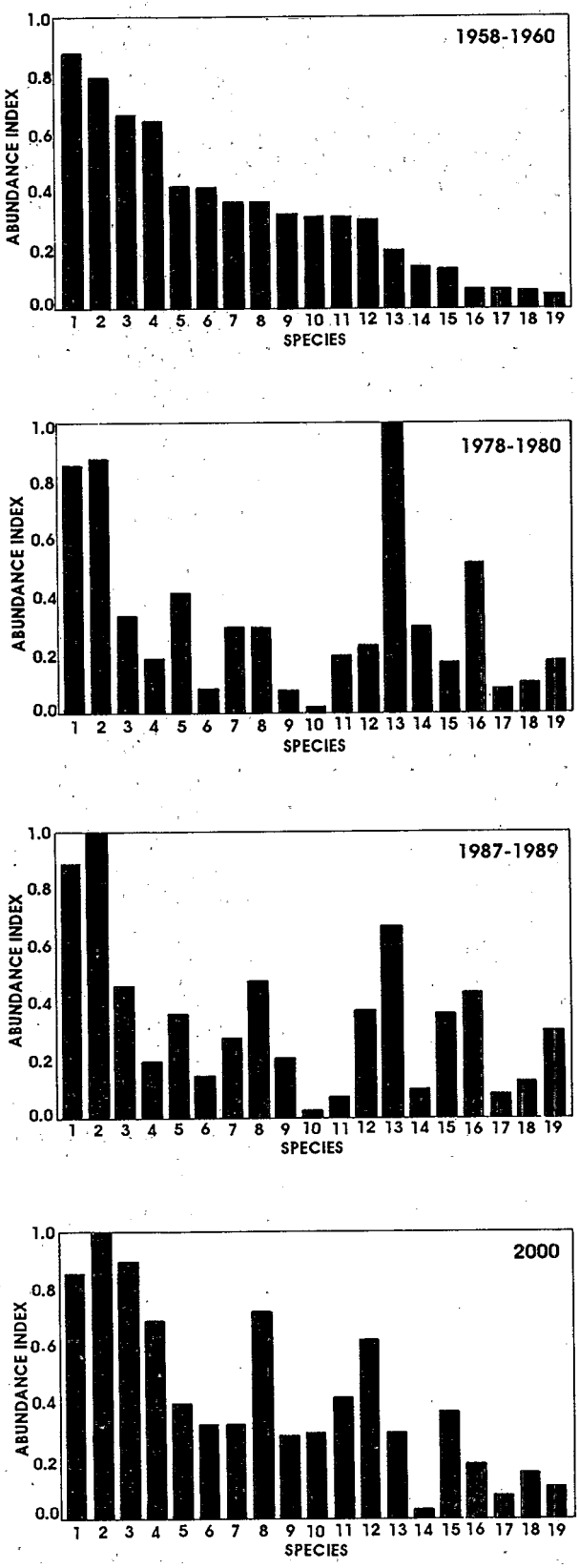
clusions to date support the idea that sampling fish with methods commonly used in Chesapeake Bay will provide very useful data for establishing estuarine biological criteria, for monitoring, and for setting management priorities.

The prototype IBI needs additional evaluation and testing, although the general approach appears to be sound. The data and field experience gained over the past two years of the STFMP will be used to fine-tune the IBI.

The analysis of species covariation points to at least two groups of species that appear to be indicative of water quality conditions. The anadromous group is indicative of good water quality, while the menhaden–mummichog–spot-gizzard shad group appears to represent pollution-tolerant species. The similar long-term trends in this latter group suggest that they have functioned as opportunists in Chesapeake Bay, perhaps filling niches vacated by more sensitive species that have declined in response to declining water quality and overfishing.

The presence, absence, or abundance of indicator species often has been used in water quality assessments to simplify sampling and interpretation. This analysis suggests a few indicator species. The species with the largest number of significant associations with other species (correlation analysis) were the blueback herring (anadromous group) and the menhaden (tolerant group). It would be better, however, to identify indicator species that are not subject to fishing pressure. The mummichog is a strong candidate to represent the tolerant group. Both trend and correlation analysis suggested the Atlantic needlefish (*Strongylura marina*) and the banded killifish (*Fundulus diaphanus*) as possible indicators of the anadromous, or sensitive group, but this judgement was not supported by cluster analysis. Given the natural variability of these systems, drawing conclusions about biotic integrity entirely from the presence or abundance of one or two species are not recommended.

The design of fish-based biological monitoring programs for estuaries will require different approaches than currently are used in fresh water systems. The requirement for complete sampling of the fish community (Karr, 1981) will have to be relaxed, for example. Electrofishing or exhaustive seining or trawling of large, tidal, saline systems is not practical. The gear used in the surveys discussed here is selective for small species and juvenile fish of larger species. The beach seine does not adequately represent benthic or pelagic species, but is selective for species that prefer neritic habitats. The use of small trawl nets as in the STFMP, in combination with seines, appears to give a better, but still incomplete, representation of the total assemblage of fish. However, the sampling biases that are inherent to work in estuaries do not necessarily preclude community analysis as long as sampling methods remain consistent over time. It must be recognized that only a



1	White perch
2	Atlantic Silverside
3	Striped Bass
4	Blueback Herring
5	Bay Anchovy
6	Atlantic Needlefish
7	Spottail Shiner
8	Tidewater Silverside
9	Banded Killifish
10	American Shad
11	Alewife
12	Striped Killifish
13	Atlantic Menhaden
14	Rough Silverside
15	Mummichog
16	Spot
17	Hogchoker
18	Silvery Minnow
19	Gizzard Shad

Figure 4.—Community structure at three points in time (three-year means of Baywide $\log_{10}$ catch per seine haul), and community structure projected to the year 2000. Species numbers are keyed to the 1958-60 reference period.

few "slices" of the total fish assemblage are sampled at any one time and place. This study indicates that these slices contain sufficiently consistent information about environmental quality not only to identify degraded waters, but even to begin to make some predictions about the ecological future of areas, given some knowledge of former conditions, present conditions, and quantitative management goals.

Fish sampling in estuaries, with the designs presented here, is rapid and inexpensive. Sampling for the STFMP, for example, generally requires two work days for two biologists with a small boat to sample each of the monitored tributaries (each with five stations). Because fish are identified and counted in the field, no laboratory processing of samples is required and data return is immediate. Those who use the data, however, must be willing to wait for a full season's sampling before obtaining final results, except in extreme cases (for example, where no fish are found during sampling).

Arguments against the use of fish as definitive biological indicators in estuaries include (1) the incomplete sampling problem discussed above, (2) the fact that many of the fish captured are migratory and do not necessarily reflect immediate conditions at the location sampled, (3) incomplete knowledge of the life histories and environmental tolerances of many of the species captured, and (4) the confounding effects of environmental variation and fisheries on the abundance of species. For those familiar with these problems, any one of these constraints might appear to be fatal to consistent interpretation of this data. However, this study suggests that this type of analysis is robust because the interpretation depends on *patterns of communities*, or at least consistent portions of communities, rather than individual species.

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Selection of Biological Indicators for Integrating Assessments of Wetland, Stream, and Riparian Habitats[1]

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ABSTRACT

Biological indicators were compared to physical and chemical parameters for assessing the effects of human disturbance in wetlands, streams, and riparian habitats. Two watersheds were studied in central Pennsylvania, one relatively undisturbed and one disturbed by agricultural and residential development in the lower sections. Methods based primarily on the structure and functional groupings of biological communities were used to compare the intensity of impacts. Avian similarity indices and response guilds reflected differences in habitat condition within the wetland and riparian components of watersheds. Neotropical migrants and species that have specific habitat requirements were more abundant in the reference watershed. Edge and exotic species occurred more frequently in disturbed areas. Fish and benthic macroinvertebrate communities varied between lentic and lotic waters, and between disturbed and undisturbed reaches of streams. More warmwater fish and omnivorous species were present in the disturbed watershed. Wetlands with flowing water supported macroinvertebrate taxa similar to streams, whereas wetlands with standing waters contained more pollution-tolerant species. The forested watershed provided habitat for four functional feeding groups of stream invertebrates (scrapers, shredders, collectors, and predators), whereas streams of the agricultural watershed contained primarily herbivores (scrapers and collectors). Biological monitoring, using a variety of community-based indicators, may be useful for detecting the degree of habitat disturbance and identifying areas in need of restoration.

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Introduction

The ultimate objective of the Clean Water Act is to restore and maintain the chemical, physical, and biological integrity of the nation's waters (section 101). The initial actions designed to achieve that objective focused primarily on monitoring the quality of surface waters as defined by their chemical constituents. More recently, greater attention has been directed toward a more balanced approach that adds biological and physical parameters to the assessment tool box (e.g., Karr and Dudley, 1981; Plafkin et al. 1989).

As the agency with primary responsibility for implementing provisions of the Clean Water Act, the U.S. Environmental Protection Agency (EPA) has begun to emphasize the importance of using biological criteria to assess the health of surface waters. EPA is directing states to adopt narrative biological criteria as part of their water quality standards (U.S. Environ. Prot. Agency, 1990). The primary advantage of biological indicators is that they presumably integrate the impacts of water pollution and habitat disturbance over time. This continuous record typically is not available from chemical sampling protocols. Whereas chemical parameters have proved useful for monitoring point discharges into surface waters, biological and physical measures appear to be better for assessing the effects of more dispersed impacts such as nonpoint source runoff, incremental losses of wetlands, and changes in land use along riparian corridors and throughout watersheds. Thus, as the Clean Water Act recognized initially, a multifaceted assessment using chemical, physical, and biological parameters will provide a comprehensive measure of ecological integrity for the nation's waters (U.S. Environ. Prot. Agency, 1990).

Given the importance of monitoring multiple parameters, it should be equally obvious that streams, rivers, and lakes should be studied simultaneously with their wetland and upland surroundings. The interface between surface waters, wetlands, and adjacent uplands is usually referred to as a riparian zone (Hunt, 1985). This zone, defined here as the soil, flora, and fauna within the 100-year floodplain, serves as critical habitat for a great diversity of species, and buffers the stream channel from point and nonpoint sources of pollution. This study is based on the assumption that the linkages among the biota, land use, and hydrology can be used to monitor the environmental health of watersheds. Consequently, we studied the biological, physical, and chemical characteristics of both in-stream and wetland–riparian components of two watersheds in an effort to understand these linkages. This paper documents the different responses of biotic communities to anthropogenic disturbances that affect both wetland–riparian areas and in-stream conditions.

Methods

Study Area

This study compared two watersheds in central Pennsylvania from 1987 to 1990 to investigate how biotic communities are altered when a watershed is disturbed. One of these was relatively undisturbed and was used as a reference watershed; the other had been disturbed by agricultural and residential development in the lower sections. Methods based primarily on the structure and functional groupings of biological communities were used to compare the intensity of impacts. The degree of disturbance occurring within watersheds was determined by analyzing land use patterns and hydrologic changes regardless of the specific origins of those impacts.

The undisturbed, or reference, watershed was White Deer Creek and its associated tributaries, located in rural areas of Centre and Union Counties, Pennsylvania. White Deer Creek has a drainage area of 117 km^2 and flows into the West Branch of the Susquehanna River (the first 89 km^2, within Bald Eagle State Forest, were used as the study region). Limited forestry operations as well as seasonal fishing and hunting are the only major activities within the watershed. Forested habitat covered 94 percent of the watershed, 1 percent of the area was wetland, 4 percent was partially disturbed (shrub/brush and old field), and 1 percent was disturbed area (gravel pit, barren and minor agriculture; Fig. 1; Croonquist, 1990).

Little Fishing Creek and its tributaries (109 km^2), located within agriculturally-dominated portions of Centre and Clinton Counties, Pennsylvania, constituted the disturbed watershed. Little Fishing Creek drains into Fishing Creek and eventually into the West Branch of the Susquehanna River. Little Fishing Creek flows into agricultural and residential areas along midreach and mainstem channels where the riparian and wetland zones have been altered substantially. Livestock freely roamed in and out of the stream, causing much bank erosion and siltation that had degraded both terrestrial and aquatic habitats. Forested habitats covered 71 percent of the watershed, 57 percent of which was in the upper, protected regions. Undisturbed wetlands comprised < 1 percent of the watershed, and 13 ha of the 28 ha (46 percent) of wetlands were in head-

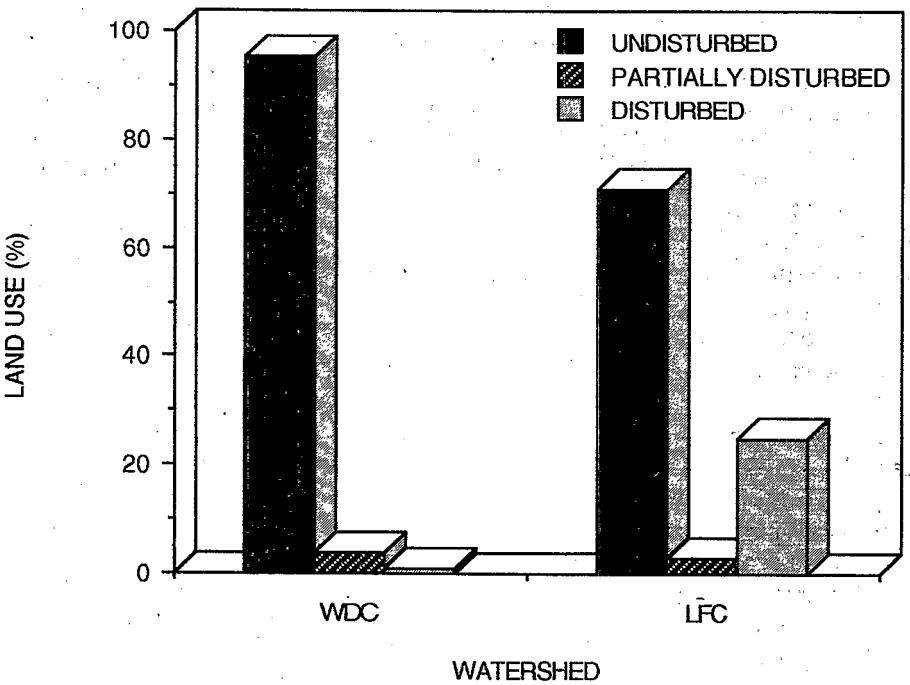

Figure 1.—Land use and cover for White Deer Creek and Little Fishing Creek.

water (undisturbed) regions. Partially disturbed habitats (shrub/brush, old fields, and partially disturbed wetlands) covered 4 percent. Over 25 percent of the watershed consisted of disturbed habitats (agriculture, residential, commercial); 94 percent of these disturbed habitats were in the middle and mainstem sections (Fig. 1; Croonquist, 1990).

Because of the large size of both watersheds and the variability of habitat from headwaters to mainstems, each watershed was divided into four hierarchical sections based on mean annual discharge and stream order. Hierarchical sections were headwater, second order tributary, midreach channel, and lower mainstem channel, with associated wetlands within each section. Three study sites were selected within each section, providing 12 study sites per watershed, 24 sites in total (Croonquist, 1990).

A geographic information system (GIS) was used to characterize land use of each watershed and each hierarchical section; however, headwaters and small tributaries could not be differentiated with the GIS, so they were grouped together. Land uses were assigned to three categories—undisturbed, partially disturbed, and disturbed. Undisturbed land use types included all forested and wetland types, except disturbed emergent and lacustrine wetlands along the middle and mainstem areas of Little Fishing Creek. Partially disturbed land use types included old fields, shrub/brush, and the previously mentioned emergent and lacustrine wetlands. Disturbed types included agriculture and development (either residential or industrial (Croonquist, 1990). The sites contained in the two middle hierarchies of Little Fishing Creek were modified to give greater weight to land disturbance rather than discharge alone.

Stream Habitat Assessment

A standardized rapid bioassessment protocol compiled by Plafkin et al. (1989) was used to characterize physical stream habitat. This method estimates general land use and physical stream characteristics such as stream width, depth, flow, and substrates to arrive at a relative assessment score ranging from severely degraded (0) to excellent (132). A total score was obtained for each study site and mean scores were compared among hierarchies within each watershed and between watersheds during the fall of 1989. In addition, we quantified the degree of sediment embeddedness in stream substrates using a 0.1 m^2-ring made of white electrical cable. The ring was tossed randomly five times at each site. Sediment area and depth were visually estimated (nearest 5 percent).

Biological and Water Chemistry Sampling

Biological and water quality sampling were conducted at each of the 24 study sites from October 1987 through September 1989 (see Brooks et al. 1990; Brooks and Croonquist, 1990; and Croonquist, 1990 for detailed descriptions of sites and methods). Each of the 24 study sites contained 3, 100-m transects. The riparian transect was located along the riparian zone (0 - 2 m from the bank) parallel with the stream channel. A second, the wetland transect, began at one end of the riparian transect and extended 100 m from and perpendicular to the channel through the adjacent wetland/upland zone. A third transect for aquatic sampling was located in the stream channel parallel to the riparian transect.

Water chemistry sampling and censusing for birds were done periodically for two years to estimate seasonal changes. Conductivity, alkalinity, pH, and temperature were used to characterize the water quality of streams and wetlands. Once each year total nitrogen (NO_3 mg/L) and total phosphorus (PO_4 mg/L) were measured in stream samples at all 24 study sites. Bird censuses consisted of 5-minute point counts at every other sample plot (every 50 m), totaling five point counts per sample site (Croonquist, 1990). All species heard and seen within a 25-m radius were recorded, which created an effective sampling area of 0.2 ha per plot and 1.0 ha for each site. Fish (electrofishing) and benthic macroinvertebrates (collected by Surber sampler in stream and Ekman dredge in wetlands) were sampled during the spring and summer of 1989. Fish were sampled in nine sites from White Deer Creek and 11 sites from Little Fishing Creek because of the absence of sufficient water in some headwater sites. Additional sampling of mammal, herpetile, and plant communities was done simultaneously, but are reported elsewhere (Brooks et al. in press; Croonquist, 1990; Croonquist and Brooks, in press).

To detect changes in biological communities among sites and between watersheds, we examined the number of species present, the amount of overlap between communities (Jaccard's Coefficient of Community; Jaccard, 1912), and the kinds of species present based on response guilds (see Brooks and Croonquist, 1990), a modified Index of Biological Integrity (Karr, 1981; Fausch et al. 1984), and other functional groupings (Plafkin et al. 1989) (Table 1).

Table 1—Criteria of response guilds for avian communities.

RESPONSE GUILDS	SCORES
Wetland Dependency	
Obligate species (> 99% in wetlands)	5
Facultative wet (usually in or near wetlands)	3
Facultative (wetlands not essential)	1
Facultative dry (occasional or no use)	0
Upland (> 99% in uplands)	0
Habitat Specificity	
Alpha species—stenotypic, specialist	5
Gamma species—landscape dependent	3
Beta species—generalist, edge	1
Trophic Level	
Carnivore, specialist (restricted diet)	5
Carnivore, generalist	4
Herbivoré, specialist (e.g., nuts, nectar)	3
Herbivore, generalist	2
Omnivore (plants or animals)	1
Species Status	
Endangered, endemic, of concern	5
Commercial, recreational value	3
Other native species	1
Exotic	0
Seasonality	
Neotropical migrant	5
Short-distance migrant	4
Year-round resident	3
Nonbreeding season resident only	2
Migratory transient	1
Occasional	0

Source: Brooks and Croonquist, 1990.

Results and Discussion

Stream Habitat Assessment

Stream habitat assessment values provided information as to the quality of riparian and in-stream habitat within and between watersheds (Table 2). There was a negative correlation between land use disturbance and habitat quality within Little Fishing Creek ($r = -0.652$, $p = 0.05$) and between watersheds ($r = -0.771$, $p = 0.05$; Croonquist, 1990). Along White Deer Creek, the headwater section had the lowest mean score (92 ± 7.2) and mainstem had the highest (130 ± 2.1), but all sections had relatively high scores. Sites with greatest anthropogenic disturbance, the mainstem sites of Little Fishing Creek, had the lowest mean score (45 ± 5.8; Croonquist, 1990). Student's t-test showed that the mean score of White Deer Creek mainstem was significantly higher (i.e., higher quality habitat) than the mean score of Little Fishing Creek mainstem ($p = 0.05$, d.f. = 10). Sediment embeddedness decreased down the hierarchy of White Deer Creek, presumably because of increasing water velocity and lack of sediment inputs from the forested watershed. Sediment embeddedness increased substantially in the lower reaches of Little Fishing Creek where cropping and grazing were the dominant land uses (Table 3). Thus, as physical disturbance increased, habitat quality decreased (Croonquist, 1990).

Water Chemistry

During the two years of study, White Deer Creek had a mean pH of 6.3 ± 0.7, mean conductivity of 34.6 ± 44.8 μS/cm, and mean water temperature of 9.2 ± 5.4 °C. Little Fishing Creek had a mean pH of 7.1 ± 0.8, mean conductivity of 153.8 ± 458.4 μS/cm, and mean water temperature of 9.8 ± 6.1 °C (Table 4). Water quality comparisons between the two watersheds were confounded somewhat because of a change in substrate type within Little Fishing Creek. Both watersheds had sandstone substrates along upper regions. As they flowed into the valleys, however, the substrate of Little Fishing Creek changed to limestone, whereas White Deer Creek remained sandstone. Mean water temperatures increased slightly through both watersheds, but did

Table 2—Stream habitat assessment scores for each hierarchy in White Deer Creek (reference) and Little Fishing Creek (impacted).

WHITE DEER CREEK		LITTLE FISHING CREEK	
STUDY SITE	SCORE	STUDY SITE	SCORE
Headwater		Headwater	
Kemmerer Trail	100	Dismal Swamp	79
Sand Spring Run	90	Fulton Gap	61
Mile Run	86	Camp Kill	112
Mean ± S.D.	92 ± 7.21		84 ± 25.87
Tributary		Undisturbed Middle	
Camp Site	110	Dam Site	78
Black Gap	115	Hecla Gap	121
Kettle Hole	107	Krislund Camp	113
Mean ± S.D.	111 ± 4.04		104 ± 22.87
Midreach		Disturbed Middle	
Beaver Dam	106	Lee's Gap	108
McCall Dam	127	Mingoville	88
Clark Trail	121	Deitrich's Trib.	47
Mean ± S.D.	118 ± 10.82		81 ± 31.1
Mainstem		Mainstem	
Gauging Station	131	Hublersburg	42
White House	132	Syndertown	42
Dump Site	128	N.J. Farm	52
Mean ± S.D.	130 ± 2.08[a]		45 ± 5.77[a,b]

Source: Croonquist, 1990.
Note: Mean score and standard deviation are given for each watershed section (Croonquist, 1990).
[a]Comparisons of mean scores between watersheds are significantly different (Student's t-test, $p = 0.05$, d.f. = 10).
[b]Pearson correlation test results show negative correlation between assessment score and land-use disturbance within Little Fishing Creek ($r = -0.652$, $p = 0.05$), and negative correlation between assessment score and land-use disturbance between undisturbed sites at the lower half of White Deer Creek and disturbed sites at the lower half of Little Fishing Creek ($r = -0.771$, $p = 0.05$).

Table 3—Percent sediment embeddedness for each hierarchy in White Deer Creek (reference) and Little Fishing Creek (impacted) watersheds.

HIERARCHY	WHITE DEER CREEK PERCENT	LITTLE FISHING CREEK PERCENT	CHI-SQUARED VALUE
Headwater	68	65	0.1
Tributary/Undis. Mid.	37	37	0.0
Midreach/Dist. Mid.	10	37	73[a]
Mainstem	3	77	1,825[a]

[a]$p < 0.05$

not differ significantly between watersheds (Table 4). Concentrations of nitrates and phosphates did not vary significantly within nor between watersheds during the two years (Table 5). The lower reaches of Little Fishing Creek did not have the high concentrations of nitrate and phosphate that were expected from the surrounding farms. However, samples were taken only once in the fall of each year, and therefore the effects of runoff of chemical fertilizers in agricultural areas probably were not adequately represented (Croonquist, 1990).

Biological Communities

Watersheds had similar communities in the upper regions where land use patterns were similar. Community similarities and functional guilds began to diverge in the lower reaches of the disturbed watershed. Birds were more indicative of these changes than in-stream fauna (Tables 6, 7, and 8). Overall, the reference watershed had somewhat fewer species than the disturbed watershed. The numbers of vertebrate species found in the undisturbed watershed versus the disturbed watershed were, respectively: 94 vs. 110 birds and 16 vs. 20 fish. The greater number of species observed in the disturbed watershed probably was due to the abundance of edge (for birds) and warmer water temperatures (for fish) in the lower reaches of Little Fishing Creek. Few differences were observed between headwaters of each watershed because conditions were similar for both forested habitat (Fig. 1), and streams (Tables 2, 3, 4, and 5).

Birds were indicative of changes in wetlands and riparian areas (Fig. 2, Table 8). Avian response guilds, when used individually or in combination, reflected the disturbance patterns of the landscape. The following avian guilds provided the most information for characterizing differences in the biotic communities between the undisturbed and disturbed watersheds: habitat specificity, trophic level, and seasonality (Brooks et al. 1990). Resident and

Table 4—Mean water quality and quantity values, and their standard deviations, from 1987–89 for White Deer Creek and Little Fishing Creek watersheds.

WATERSHED/HIERARCHY	pH	CONDUCTIVITY (µS/CM)	TEMP. (°C)	STAGE (m)
White Deer Creek				
Headwater	6.0 ± 0.6	34.9 ± 50.3	8.2 ± 4.2	−1.6 ± 5.9
Tributary	6.5 ± 0.2	41.3 ± 54.7	8.9 ± 4.5	−3.4 ± 8.4
Midreach	6.6 ± 0.2	37.3 ± 50.3	10.7 ± 5.8	−0.8 ± 8.1
Mainstem	6.6 ± 0.2	52.9 ± 70.0	10.9 ± 6.2	2.7 ± 11.7
Little Fishing Creek				
Headwater	6.7 ± 0.5	90.0 ± 123.9	9.6 ± 5.6	0.8 ± 9.9
Undisturbed middle	6.9 ± 0.6	92.0 ± 103.7	10.1 ± 5.7	−2.7 ± 10.7
Disturbed middle	7.4 ± 0.3	159.3 ± 131.7	11.2 ± 6.0	−2.3 ± 9.3
Mainstem	7.7 ± 0.3	199.8 ± 74.6	12.5 ± 7.2	−1.4 ± 14.7

Table 5—Mean values (n=3), and standard deviation, of total nitrate (NO_3 mg/L) and total phosphate (PO_4 mg/L) from water samples of each hierarchy of White Deer Creek and Little Fishing Creek watersheds.

HIERARCHY	TOTAL NITRATE 9/88	TOTAL NITRATE 12/89	TOTAL PHOSPHATE 9/88	TOTAL PHOSPHATE 12/89
White Deer Creek				
Headwater	0.67 ± 0.19	0.68 ± 0.71	0.024 ± 0.016	0.005 ± 0.006
Tributary	0.97 ± 0.12	0.75 ± 0.77	0.008 ± 0.006	0.004 ± 0.004
Midreach	0.40 ± 0.05	0.28 ± 0.03	0.014 ± 0.005	0.005 ± 0.002
Mainstem	0.72 ± 0.36	1.35 ± 0.61	0.068 ± 0.108	0.004 ± 0.001
Little Fishing Creek				
Headwater	0.48 ± 0.46	1.32 ± 0.35	0.025 ± 0.017	0.068 ± 0.095
Undisturbed middle	0.23 ± 0.06	1.13 ± 0.39	0.015 ± 0.009	0.010 ± 0.003
Disturbed middle	0.40 ± 0.17	0.65 ± 0.39	0.025 ± 0.012	0.012 ± 0.007
Mainstem	0.72 ± 0.38	1.03 ± 0.33	0.052 ± 0.025	0.017 ± 0.003

Table 6—Metrics used to assess fish communities in White Deer Creek and Little Fishing Creek.

CATEGORY	METRIC	SCORING CRITERIA 5	3	1
Species Richness and Composition	1. Total no. of fish species			
	2. Proportion of sculpins			
	3. Number cyprinid species			
	4. Number adult trout spp.			
	5. Number intolerant spp.			
	6. Proportion suckers	<5	5–20	>20
Trophic Composition	7. Proportion individuals as omnivores	<20	20–45	>45
	8. Proportion total insectivores	>80–100	>40–80	0–40
	9. Proportion Salmonids	>5	5–1	<1

Key: Integrity classes for total IBI scores are: excellent (58–60); good (48–52); fair (40–44); poor (28–34); and very poor (12–22). Expectations for metrics 1–5 vary with stream size and region (Karr, 1981; Fausch et al. 1984).

neotropical-migrant breeders that had specific habitat requirements and/or were carnivorous (e.g., woodland warblers, such as cerulean warbler, *Dendroica cerulea*, black-throated blue warbler, *Dendroica caerulescens*, and northern waterthrush, *Seiurus noveboracensis*) decreased in percentage only down the hierarchy of the disturbed stream. Edge (blue jay, *Cyanocitta cristata*) and exotic (house sparrow, *Passer domesticus*) species were found in greater abundance in the disturbed watershed. Neotropical migrants with high guild scores for habitat specificity formed about 25 percent of the community in upper regions of both watersheds. These species formed only 5 percent of the mainstem community of the disturbed watershed, but remained a major component of the undisturbed watershed community.

Aquatic communities paralleled the trends observed for physical habitat assessments of in-stream and riparian areas despite the modest sampling effort. The results of the IBI were more indicative of changes in fish communities than results from the community coefficients (Fig. 3, Tables 6 and 7). The IBI suggested that fish communities of the lower

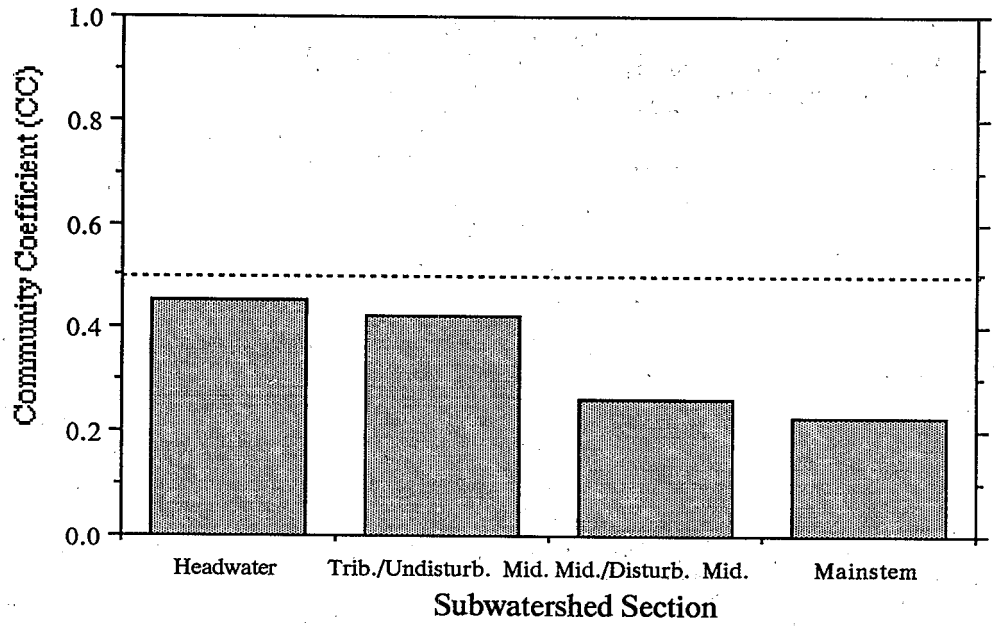

Figure 2.—Avian community coefficient values between watershed sections of White Deer Creek and Little Fishing Creek, for the riparian transect.

Table 7—Mean Index of Biotic Integrity (IBI) for fish in each hierarchy in White Deer Creek (reference) and Little Fishing Creek (impacted) watersheds.

HIERARCHY	WHITE DEER CREEK SCORE (CLASS)	LITTLE FISHING CREEK SCORE (CLASS)	CHI-SQUARED VALUE
Headwater	48 ± 0[a] (good)	20 ± 28[b] (very poor)	2.67
Tributary/Undis. Mid.	48 ± 0 (good)	41 ± 2 (fair)	2.99
Midreach/Dis. Mid.	47 ± 1 (good)	45 ± 9 (fair to good)	3.63
Mainstem	42 ± 2 (good)	35 ± 1 (poor to fair)	9.77[c]

[a]Mean ± SD.
[b]Large standard deviation because one site contained no fish.
[c]$p < 0.05$.
Key: Integrity classes for total IBI scores are: Excellent (58–60); good (48–52), fair (40–44); poor (28–34); and very poor (12–22).

Table 8—Percent composition (%) of selected response guilds of birds. Results given by hierarchical section of White Deer Creek and Little Fishing Creek.

	HEADWATER		TRIBUTARY[a]		MIDREACH[b]		MAINSTEM	
	WDC	LFC	WDC	LFC	WDC	LFC	WDC	LFC
Exotic (Status = 0)	2	4	0	3	0	7	0	8
Edge (Habitat Specificity = 1)	39	51	42	47	46	66	55	68
Permanent Resident + Edge (Seasonality = 3 & Hab. Spec = 1)	19	29	21	28	25	41	31	45
Habitat Specific (Habitat Specificity = 5 or 3)	61	49	58	53	54	34	45	32
Neotropical Migrant (Seasonality = 5)	41	40	42	32	32	24	41	20
Wetland Dependent (Wetland Dependency = 5, 3, or 1)	36	37	32	35	33	30	33	31
Neotropical Migr. + Habitat Specific (Season. = 5 & Hab. Spec = 5 or 3)	29	23	28	18	19	9	22	6

[a]Tributary section of White Deer Creek and undisturbed middle section of Little Fishing Creek.
[b]Midreach section of White Deer Creek and disturbed middle section of Little Fishing Creek.
Note: Numbers underlined represent large differences in percent composition between watersheds (Croonquist and Brooks, 1991).

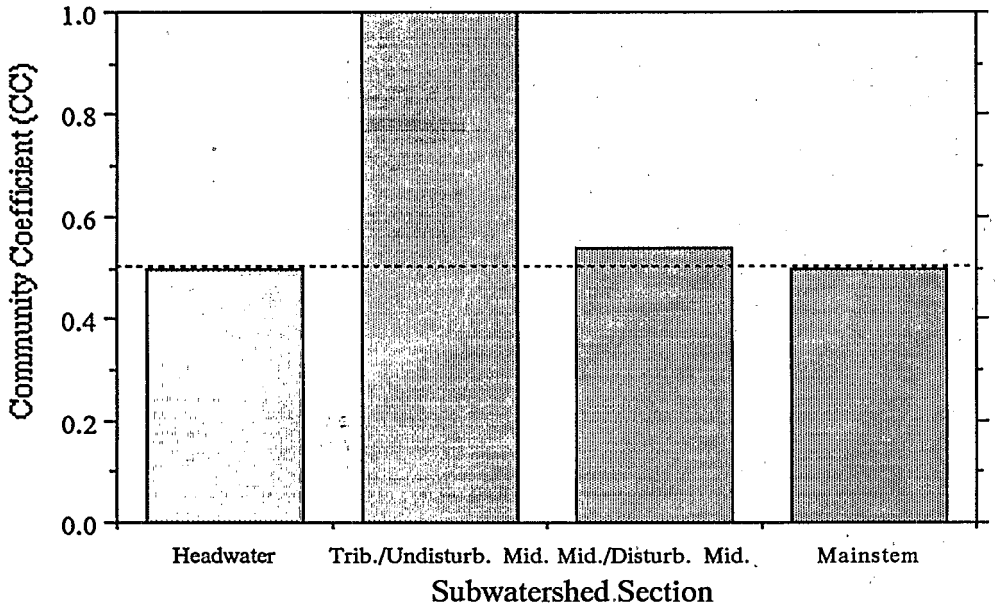

Figure 3.—Fish community coefficient values between watershed sections of White Deer Creek and Little Fishing Creek.

reaches of Little Fishing Creek, where disturbance was greatest, were negatively impacted (Tables 6 and 7). The lower regions of the disturbed watershed, which had degraded water quality, severely disturbed stream banks, and increased sediment loads on the stream bottom, supported more warmwater fish species (centrarchids) and pollution-tolerant omnivores (white sucker, *Catostomus commersoni*, and horned chub, *Nocomis biguttatus*). More sensitive species, such as salmonids and other insectivores (blacknose dace, *Rhinicthys atratulus*), were more abundant in the reference watershed.

Storage and sorting problems with the macroinvertebrate samples prevented the use of a quantitative assessment method such as those suggested by Plafkin et al. (1989); however, some qualitative trends were apparent. Wetlands with flowing water supported macroinvertebrate taxa similar to those of streams (ephemeropterans, plecopterans, tricopterans), whereas wetlands with standing waters contained hydracarinids, dipterans, annelids, and pelecypods. The forested watershed provided habitat for four functional feeding groups (scrapers, shredders, collectors, and predators), whereas streams of the agricultural watershed contained primarily herbivores (scrapers and collectors).

In summary, the subtle impacts of habitat disturbance in the riparian zone were apparent through investigations of physical, chemical, and biological parameters. Differences between reference and disturbed watersheds were more obvious in the riparian corridor (e.g., stream habitat assessment and avian response guilds) than for in-stream conditions (e.g., water chemistry, fish, and in-stream macroinvertebrate communities). However, the in-stream parameters were sampled at much lower frequencies. Changes in fish and macroinvertebrate communities were most obvious where physical conditions of the stream were severely degraded, as in the mainstem of Little Fishing Creek.

Protection of wetlands, streams, and riparian corridors is critical for maintaining biological diversity of terrestrial and aquatic species. Biological monitoring, using a variety of community-based indicators, may be useful for detecting the degree of habitat disturbance and identifying areas in need of restoration when used in conjunction with physical and chemical indicators. This three-parameter approach can be used to target restoration efforts toward portions of watersheds where recovery is feasible.

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Biological Survey Study Design Considerations When Representing Biointegrity and Evaluating Non-Attainment of Designated Uses

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Biological survey study design is critically important in biocriteria development. The design must be scientifically rigorous to provide for legally defensible data and be biologically relevant to detect problems of regulatory concern. It is not financially nor technically feasible to completely evaluate an entire ecosystem at all times, selecting community components, the time, season, station location, methods to measure the community of interest, and a quality assurance and quality control program are important to the success of a biocriteria program. When using biological surveys to establish what the state of biointegrity is and to determine if designated uses are being attained there are several considerations that should be taken into account. An introduction to quality assurance and quality control considerations as well as the role data quality objectives have in helping focus biological survey design will be discussed. An overview will be given on things to keep in mind when designing biological surveys like selecting aquatic community components, designing biological surveys to measure these aquatic community components, metric selection and sampling design. The discussion of these topics will follow the Biological Criteria National Program Guidance for Surface Waters.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Significance of Change in Community Structure: A New Method for Testing Differences

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ABSTRACT

Structural changes in biotic communities often precede detectable adverse effects on ecosystem process rates. Management concerns may focus on changes in abundance, the loss of important species, or changes in the composition or diversity of groups of taxa. Community structure data are high in information content, but may be difficult to analyze and interpret. Bioassessment methods have recommended use of multivariate procedures for examining differences in community similarity, but the number of species (variables) in community samples and the limited number of replications make application of multivariate methods problematic; the number of degrees of freedom will usually be fewer than the number of variables and the covariance matrix will be singular. Additionally, some species present at one site will be absent at other sites, especially those that are impacted, thus invalidating assumptions of normality. An analytical method is needed that can use community information for inferential analysis of environmental effects without violating assumptions of statistical models. To use community structure information, species data must first be reduced to measures of similarity (or distance) among replicates. Measures (indices) for assessing community similarity can be based on presence-absence, ranked or rated abundance, and relative or absolute abundance of taxa. A permutation procedure involving a large number of random switches of similarity measures is used to build a probability distribution of the ratio of mean between treatment or location analysis of variance procedures. Assuming that a null hypothesis of no difference among locations is rejected, follow-up analyses can indicate the locations or treatments that are different and can identify the influence of species whose presence, absence, or abundance greatly affects analyses. This research has used the permutation method and binary, ranked, and continuous data to examine community changes in multispecies laboratory experiments and field evaluations of periphyton and invertebrate communities. Unlike descriptive multivariate methods or rating/score methods, the permutation method provides an inferential test of hypothesized differences between reference sites to suspected impact sites. The technique is an objective means of determining differences and identifying the taxa indicative of community differences.

Introduction

Evaluating biological changes in stressed ecosystems is based on the hypothesis that human influence alters the sustainability of ecosystem services. Controversy over the conceptual basis for ecosystem protection has followed the topical, favoring paradigms of ecology: studies that once focused on describing the structure of ecosystems gave way to process studies examining sources and flows of energy and materials. Increasing evidence suggests, however, that aquatic ecosystem structure changes before process accommodations are detectable (Odum, 1985, 1990; Schindler, 1987; Schaeffer et al. 1988; Pratt, 1990). Process measures are robust and change primarily with the availability of substrates (nutrients, dead organic matter) rather than the biological machinery that processes these substrates. Where stresses do not significantly alter the supply of substrates, process measures show little impact (Levine, 1989).

Evaluations of aquatic community structure have re-emerged as an important facet of environmental impact analysis and have already been incorporated into the regulatory framework in several States. The establishment of biological criteria for waters implies that methods are available to detect significant changes in community structure. Taxonomic lists incorporating the presence, absence, or abundance of species at particular locations of interest have high information content. The means for dealing with this information vary considerably depending on the questions under study. This paper presents methods for examining community structure information using inferential procedures for testing hypotheses of community change.

Early in this century, the ubiquity of organic pollution and its effects on aquatic communities led to the classification of organisms by enrichment (or low oxygen) tolerance, the classic Saprobian system (Kolkwitz and Marsson, 1908). In the early 1970s, questions focused on changes in biotic diversity, and information theory (Shannon and Weaver, 1949) was applied to the comparisons of community taxa abundance. However, information theory indices were often sensitive to the number of taxa (Green, 1979) and aroused considerable debate about the relationship between biotic diversity and community stability (Hurlbert, 1971).

Current impact analysis makes use both of improved knowledge of stream ecology and our ability to deal with complex data sets. Understanding of the distribution, tolerance, and habits of aquatic species is now much clearer (e.g., Lowe, 1974; Hilsenhoff, 1982; Cummins and Klug, 1979; Karr et al. 1986). A variety of systematic procedures for evaluating taxonomic structure is available (Metcalfe, 1989; Cairns and Pratt, in press). Other descriptive procedures allow comparison of collections according to taxonomic composition and abundance using community similarity or multivariate distance measures (e.g., Gauch, 1982; Pielou, 1984; Digby and Kempton, 1987). Such procedures have been used to examine community differences along environmental gradients that include habitat alterations and pollutants (e.g., Pratt et al. 1985; Whittier et al. 1988). However, these descriptive procedures do not provide rigorous, inferential methods for testing differences among communities.

In 1986 the Environmental Protection Agency moved to standardize and improve biological methods for assessing and monitoring surface waters. An important product for this effort was a guidance document on rapid biological assessment focusing on community structure of fish, macroinvertebrates, and algae (Plafkin et al. 1989). Methods for assessing fish communities were based on the work of Karr and colleagues (1986), while those for macroinvertebrates were based both on historical methods and the development of additional methods analogous to those for fish. Much of this work was incorporated into methods now used by the state of Ohio in assessing biological water quality (Ohio Environ. Prot. Agency, 1987).

Rapid bioassessment procedures are systematic means for collecting and evaluating community and habitat structure information. The procedures have various uses, including determining attainability of water uses and characterizing the degree of use impairment. The procedures recognize the potential for regional variation in ecosystem condition and performance (Omernik, 1987). Site comparisons are made across limited environmental gradients to either an upstream reference site community, or to a regional reference community. Methods for assessing macroinvertebrate communities use the extent of similarity to the reference community as one of several community assessment metrics. While the rapid bioassessment procedures are rational means for evaluating community data, they are not inferential. The indices developed usually lack any estimate of variance and, therefore, cannot provide a statistical comparison among sites. Additionally, the use of multivariate procedures to compare communities violate assumptions of normality when taxa are present at some sites but not others.

Regardless of the mechanism by which biological criteria are developed, community comparisons and inferential procedures will be needed to detect changes in biological structure. The methods de-

scribed in this paper can be used to compare communities to regional reference communities or to upstream reference sites and can provide an inferential test of hypotheses of no difference among communities. Studies of biological quality will need to demonstrate rigor in the quality of sampling, the identification of species, and the analysis of results.

Methods

The analytical method presented here tests for differences in community structure among sites or treatments (see Smith et al. 1990). The analyses require the selection of a measure of community similarity-dissimilarity or distance. Following the construction of a matrix comparing community measures by sites or treatments, a permutation procedure is used to repeatedly and randomly switch measures among the site or treatment categories. Switching similarity measures is equivalent to switching data vectors (i.e., switching all data for one treatment replicate). Statistical comparisons are based on the relationship of between treatment similarity to within treatment similarity, analogous to analysis of variance procedures. Follow-up analyses examine pairwise comparisons of treatment categories and identify the influence of particular species on the chosen community measure.

Community Similarity—an Overview

The array of methods for estimating community similarity is diverse and will not be reviewed in detail here. Legendre and Legendre (1983) list 27 community metrics. With reference to ecological studies, indices and methods for comparing communities are succinctly reviewed by Pielou (1984) and Digby and Kempton (1987). Certain indices are recommended by Plafkin et al. (1989), but most indices have uncertain or unknown statistical properties and few have been rigorously studied for their sensitivity in detecting community change.

Two aspects of the assessment of community similarity are worth noting. First, both similarity measures (comparisons of species overlap between physical samples) and distance measures (distance between samples in multidimensional species space, often a complement of similarity) are available to examine the relationship among replicates from different sites. Second, presence–absence (binary), relative abundance, and actual or absolute abundance are simply scales that weight the importance of a species. In binary data the weightings are 1 (present) and 0 (absent). Relative abundance data may be presented as abundance rankings (usually scaled as integer values between 0 and 10) or as proportional abundance (scaled by the total number of individuals counted in each replicate). Actual or absolute abundance data weight individual species according to the actual number of individuals counted and weight different replicates by the total number of individuals. Other measures of abundance may be used and include such estimators as biomass, biovolume, cover, and importance value.

Binary data underestimate the importance of dominance changes, but are more closely related to expectations of falling species numbers in stressed communities. Binary data are comparatively easier to obtain because time is not consumed enumerating individuals, a laborious task for small taxa (e.g., algae, protozoa, micrometazoa). Where the scale of sampling is large compared to the size of organisms, relatively complete sampling can be assumed. This is usually the case in sampling microorganisms. However, when communities of larger taxa such as macroinvertebrates or fishes are used, evidence of the adequacy of sampling (e.g., a species-area or species-effort curve) is needed to determine the appropriate scale and number of replicates. When the number of taxa is large, binary data are sufficient to detect changes in community structure.

The example analyses presented here use binary data or rated abundance, although the methods are applicable to indices derived from weightings with approximately continuous data such as individuals per species or continuous data such as biomass or biovolume per species. Obviously, enumeration of individuals ignores size differences among species; however, these estimates of population sizes within replicate physical samples are appropriate for comparisons among sites or treatments. The analyses are appropriate for the analysis of multispecies laboratory experiments and for comparisons of replicate samples among field sites.

Data Requirements

The starting point for analyses is the development of a taxonomic summary of species presence (or abundance) in replicate physical samples by treatment or site. Replicate physical samples of the within-site or within-treatment conditions are required. The number of replicates may vary, but at least three or four replicate samples per site are necessary to obtain adequate estimates of variability. Taxa may be identified to any practical taxonomic level, although the rigor of the analyses and the conclusions that can be drawn will depend on the level of taxonomic precision chosen. Family-

level classification will likely increase similarity while species-level taxonomy will decrease similarity among replicates. Two example data sets are shown in Tables 1 and 2.

Selection of Community Measure

As previously mentioned, the selection of community association measures will, in part, influence the outcome of analyses. Similarity measures are based on comparisons of shared and unique taxa between pairs of samples (Fig 1). Examples of commonly used measures of both similarity and distance are shown in Table 3. Negative matches (taxa failing to occur in both samples) are often ignored in similarity measures because the absence of a particular taxon may not be judged important (Roback, 1974). However, when taxa have particular indicator value, the absence of a taxon in two samples may make the association between the samples stronger. This is especially useful when important taxa are expected at study sites. For example, the absence of red oak trees at two sites might be considered important in associating those

		Sample i	
		Species present	Species absent
Sample j	Species present	a	b
	Species absent	c	d

Figure 1.—Association matrix used to compute similarity measures.

sites, and this importance should be reflected in the choice of the community similarity measure. Binary, ranked, and continuous data can be used to form many of the indices.

Some measures or their complements are metric; that is, they satisfy the triangle inequality principle. Other measures that fail to satisfy the triangle inequality are termed semimetric (Legendre and Legendre, 1983). Semimetric measures may have less predictable statistical properties. A difference of a given magnitude between semimetric coefficients may not have the same meaning for all values of the

Table 1.—Sample data set showing ranked abundance of taxa (scaled 1–5) by treatment and replicate.

TAXON		TREATMENT 1	2	3	4	5	6
A	sp1	0 0 1	0 0 0	0 1 0	0 1 0	3 3 3	0 4 4
A	sp2	5 5 5	4 4 4	3 3 3	2 2 2	1 1 1	1 1 1
C	sp1	1 1 1	1 1 1	0 1 1	1 1 0	1 0 1	0 1 0
C	sp2	1 1 1	1 1 1	1 1 0	0 0 1	0 1 0	0 0 0
C	sp3	0 0 0	1 0 0	0 0 0	1 1 1	1 0 0	1 1 1
C	sp4	1 1 1	0 1 1	1 0 1	0 1 1	0 3 3	5 5 5
C	sp5	1 1 1	1 1 1	1 1 0	1 1 0	0 0 0	1 0 0
D	sp1	0 0 0	0 0 0	0 0 0	1 1 0	1 1 0	1 0 1
F	sp1	1 1 1	1 1 1	0 1 1	0 1 1	1 1 1	1 0 1
F	sp2	1 1 1	1 1 1	1 1 1	1 1 1	0 1 0	1 1 0
G	sp1	0 0 1	0 0 0	0 0 1	0 2 2	3 3 3	0 4 4
G	sp2	1 1 1	0 1 1	0 1 0	1 0 1	1 0 0	0 0 1
G	sp3	1 1 1	0 1 1	1 0 0	1 1 0	0 0 1	0 1 0
G	sp4	1 1 1	1 1 1	0 0 1	0 1 0	0 0 0	1 0 1
M	sp1	1 1 1	1 1 1	1 0 1	1 0 0	0 0 1	1 0 0
M	sp2	1 1 1	1 1 0	1 1 0	1 0 1	1 1 1	0 1 1
N	sp1	5 5 5	4 4 4	3 3 3	2 2 2	1 1 1	1 1 1
N	sp2	1 1 1	0 1 0	1 1 1	1 0 0	0 0 1	0 1 1
N	sp3	1 1 1	0 1 1	0 0 1	0 1 1	1 0 0	0 0 0
N	sp4	0 0 0	0 0 0	1 0 0	1 0 1	0 1 1	1 0 1
N	sp5	1 0 0	1 1 1	1 0 1	0 0 0	0 0 0	0 0 0
N	sp6	1 1 1	1 1 1	1 1 1	0 0 1	1 1 1	0 0 1
N	sp7	1 1 1	1 1 1	0 0 1	1 0 1	0 0 0	1 1 1
P	sp1	0 1 0	0 1 0	1 0 0	0 1 0	1 0 1	1 0 0
S	sp1	1 0 0	1 0 0	1 1 0	0 0 0	0 0 0	1 1 0
S	sp2	0 1 0	1 0 1	0 1 1	0 0 0	1 0 0	0 0 0
S	sp3	0 1 0	1 1 0	0 0 0	1 1 0	0 0 1	0 0 0
S	sp4	1 1 1	0 1 1	1 0 1	0 1 1	0 1 0	1 1 1
S	sp5	1 1 1	1 1 0	1 1 0	1 1 0	1 1 0	1 1 0
S	sp6	5 5 5	4 4 4	3 3 3	2 2 2	1 1 0	1 0 1
Total taxa		22 22 24	19 20 23	19 17 16	17 16 19	16 15 15	17 17 15

Note: The ordered treatment values correspond to increasing concentrations of copper.

Table 2.—Data from Table 1 rearranged by frequency of occurrence of taxa in replicates.

TAXON	TREATMENT 1	2	3	4	5	6	FREQUENCY
A sp2	5 5 5	4 4 4	3 3 3	2 2 2	1 1 1	1 1 1	18
N sp1	5 5 5	4 4 4	3 3 3	2 2 2	1 1 1	1 1 1	18
S sp6	5 5 5	4 4 4	3 3 3	2 2 2	1 1 0	1 0 1	16
F sp2	1 1 1	1 1 1	1 1 1	1 1 1	0 1 0	1 1 0	15
F sp1	1 1 1	1 1 1	0 1 1	0 1 1	1 1 1	1 0 1	15
C sp4	1 1 1	0 1 1	1 0 1	0 1 1	0 3 3	5 5 5	14
M sp2	1 1 1	1 1 0	1 1 0	1 0 1	1 1 1	0 1 1	14
N sp6	1 1 1	1 1 1	1 1 1	0 0 1	1 1 1	0 0 1	14
C sp1	1 1 1	1 1 1	0 1 1	1 1 0	1 0 1	0 1 0	13
S sp4	1 1 1	0 1 1	1 0 1	0 1 1	0 1 0	1 1 1	13
S sp5	1 1 1	1 1 0	1 1 0	1 1 0	1 1 0	1 1 0	13
N sp7	1 1 1	1 1 1	0 0 1	1 0 1	0 0 0	1 1 1	12
C sp5	1 1 1	1 1 1	1 1 0	1 1 0	0 0 0	1 0 0	11
M sp1	1 1 1	1 1 1	1 0 1	1 0 0	0 0 1	1 0 0	11
N sp2	1 1 1	0 1 0	1 1 1	1 0 0	0 0 1	0 1 1	11
C sp2	1 1 1	1 1 1	1 1 0	0 0 1	0 1 0	0 0 0	10
G sp4	1 1 1	1 1 1	0 0 1	0 1 0	0 0 0	1 0 1	10
G sp2	1 1 1	0 1 1	0 1 0	1 0 1	1 0 0	0 0 1	10
G sp3	1 1 1	0 1 1	1 0 0	1 1 0	0 0 1	0 1 0	10
G sp1	0 0 1	0 0 0	0 0 1	0 2 2	3 3 3	0 4 4	9
N sp3	1 1 1	0 1 1	0 0 1	0 1 1	1 0 0	0 0 0	9
A sp1	0 0 1	0 0 0	0 1 0	0 1 0	3 3 3	0 4 4	8
C sp3	0 0 0	1 0 0	0 0 0	1 1 1	1 0 0	1 1 1	8
N sp4	0 0 0	0 0 0	1 0 0	1 0 1	0 1 1	1 0 1	7
P sp1	0 1 0	0 1 0	1 0 0	0 1 0	1 0 1	1 0 0	7
D sp1	0 0 0	0 0 0	0 0 0	1 1 0	1 1 0	1 0 1	6
N sp5	1 0 0	1 1 1	1 0 1	0 0 0	0 0 0	0 0 0	6
S sp1	1 0 0	1 0 0	1 1 0	0 0 0	0 0 0	1 1 0	6
S sp3	0 1 0	1 1 0	0 0 0	1 1 0	0 0 1	0 0 0	6
S sp2	0 1 0	1 0 1	0 1 1	0 0 0	1 0 0	0 0 0	6

Table 3.—Exemplary measures of community similarity. Formulas are based on terminology shown in Fig. 1.

COEFFICIENT	FORM	CLASS	REFERENCE
Simple matching	$\frac{a + d}{a + b + c + d}$	metric	Digby & Kempton, 1987
Jaccard	$\frac{a}{a + b + c}$	metric	Jaccard, 1901
Czekanowski	$\frac{2a}{2a + b + c}$	semimetric	Czekanowski, 1909 Sorensen, 1948
Margalef	$\frac{a(a+b+c+d)}{(a+b)(a+c)}$	semimetric	Margalef, 1958
Community loss	$\frac{c}{a + b}$	nonmetric	Courtemanch & Davies, 1987

Note: Measures whose complements satisfy the triangle inequality are termed metric. Those that do not are semimetric. Measures whose complements can take negative values are termed nonmetric.

coefficients. Still others are nonmetric because their complements can be negative, and the application of such coefficients may be problematic.

Many other measures of both community similarity or distance can be used (Legendre and Legendre, 1983; Digby and Kempton, 1987; Plafkin et al. 1989). Among the more familiar to ecologists and pollution biologists are Euclidian distance, Manhattan (city block) distance, Bray-Curtis measure, Canberra measure, correlation coefficient complement, Morisita's index, and Pinkham and Pearson measure. A general form for community measures (and their complement distance measures) is given by Gower (1971).

Analysis Method

A similarity matrix for all possible replicate pairs is constructed using a chosen metric. A test statistic is computed comparing the mean similarity of replicate objects within "treatments" to the between treatment similarity. If the test statistic for data is

$$L(\text{data}) = \overline{B} / \overline{W} \qquad (1)$$

where $\overline{B}$ is the mean between treatment similarity and $\overline{W}$ is the mean within treatment similarity, then this statistic can be compared to one derived from a permutation procedure in which coefficients in the

similarity matrix are randomly switched a large number of times (1,000). A test statistic L (permute) is recalculated as above after each permutation of the matrix. The distribution of L (permute) can be used to determine if L (data) can be differentiated from L (permute) at a given alpha (α). For example, if 1000 random switches (permutations) are made then one would reject a null hypothesis of no difference in community similarity at $\alpha = 0.05$ if L (data) were more extreme than 950 (95%) of the L (permute) values. Because the total similarity (T) is a constant for any given matrix, the component B and W similarities may also be used as test statistics.

Similar arguments can be made for the use of distance measures rather than community similarity. However, in multidimensional space, the location of similar samples is associated with a small distance; dissimilar samples would be more widely separated. Therefore, the principles of using distance measures as measures of similarity would apply to testing community differences, but the expected direction of change in the randomly switched matrix would be opposite to expectations for similarity measures.

Follow-up Analyses

Assuming that a hypothesis of community similarity among sites or treatments is rejected, several follow-up analyses are possible. One approach would be to conduct multiple comparisons using the permutation procedure on treatment pairs. This approach would test differences between individual treatments or sites. However, if the number of replicates is small (as it often is in field studies), detection power is limited by the fact that only a small number of unique permutations of the similarity matrix are possible.

A second useful follow-up analysis is to determine the relative contribution of each taxon to community similarity. The effect of taxa on similarity can be determined by computing the effect on similarity of removing each taxon. In this analysis, removing common taxa will reduce total similarity. Taxa adding heterogeneity to the matrix (and so decreasing similarity) will increase total similarity when removed. Identification of these taxa permits an inspection of the data matrix to identify taxa that may appear or disappear in ceratin treatments or sites. When coupled with a data matrix sorted by the total frequency of occurrence of taxa, these identifications become easier (e.g., Table 2). Additional follow-up analyses are summarized by Smith et al. (1990).

Community Data Analysis

The hypothetical data set in Table 1 was constructed (based on actual data of effects of copper on periphyton algae) to demonstrate a method for detecting changes in community composition between sites or treatments. The presence of taxa was determined from 500 cell counts of preserved samples. The ordered treatments (1-6) ranged from controls (<10 μg Cu/L) to 80 μg Cu/L. The data matrix has been edited to include only the 30 most common taxa and to magnify abundance differences among taxa.

Results

Example Analysis—Similarity

Based on presence–absence data shown in Table 1, a similarity matrix was constructed using the Jaccard measure. Communities have higher similarity if they share a greater proportion of their total species. A portion of this matrix is shown in Figure 2. Rectangular blocks in the table mark similarity coefficients comparing samples between treatments. Triangular areas lying along the matrix diagonal are coefficients showing the within treatment similarity. Computation of within and between treatment similarity was followed by 1,000 permuta- tions randomly switching matrix coefficients to produce a hypothetical distribution of within-treatment similarity (Fig. 3).

These analyses showed that the critical within-treatment similarity based on $\alpha = 0.05$ was 0.5155. This compared to the actual with-treatment similarity of 0.5271. Further, the number of permutation similarity values that were more extreme than this observed similarity was 17, corresponding to a p-value of 0.018 ($p = [17+1]/1000$)). Therefore, the hypothesis of no difference in similarity among treatments is rejected.

Follow-up analyses showed that similarity of treatments to controls (group 1) decreased with increasing copper levels (Fig. 4) and that common species strongly affected community similarity. Species with moderate negative or positive influence are typically those that are either eliminated in higher treatments (e.g., C. sp2) or that become more frequent in replicates at high copper levels (e.g., C. sp3, D. sp1). Based on multiple pairwise comparisons, only treatments 2 and 5 could be differentiated.

Treatment/Replicate													
1A	1B	1C	2A	2B	2C	3A	3B	3C	...	6A	6B	6C	
1	.80	.83	.64	.88	.78	.67	.58	.63	...	.50	.48	.44	1A
	1	.80	.62	.92	.75	.58	.56	.60	...	.48	.41	.43	1B
		1	.52	.80	.71	.54	.58	.63	...	.44	.54	.56	1C
			1	.62	.58	.48	.59	.50	...	.50	.36	.33	2A
				1	.75	.64	.50	.60	...	.48	.41	.43	2B
					1	.48	.46	.71	...	.44	.31	.38	2C
						1	.48	.40	...	.52	.43	.35	3A
							1	.38	...	.32	.41	.38	3B
								1	...	.42	.39	.48	3C

Figure 2.—Part of the matrix of Jaccard similarity coefficients from the examination of data in Tables 1 and 2.

Effect of Measure

Comparable analyses to those presented were done using only presence–absence data to determine Euclidian distance among treatments. Distance analyses provide estimates of community dissimilarity: dissimilar communities are more widely separated in multidimensional space. These analyses are essentially identical to those using presence–absence based similarity (Fig. 5). The within-treatment distance estimate is less extreme than 7 of 1,000 permutation-based within-treatment values ($p = 7+1/1000 = 0.008$), so the hypothesis of no difference in distance among treatments is rejected.

Follow-up analyses identified the same patterns found using analysis with Jaccard's coefficient (Fig. 6). Evaluation of these data using several other presence–absence based metrics revealed essentially similar patterns, although other metrics weight common species more heavily and so produce coefficients of greater magnitude.

Effect of Data Type

While presence–absence data were effective in detecting differences among treatments, data evaluated by rank abundance resulted in greater detection power. For example, the rankings shown in Table 1 weight only 6 of 30 species; three become less abundant across treatments, and three become more abundant. Examination of the weighted data using Euclidian distance showed clear treatment differences (Fig. 7) and allowed detection of additional differences between treatment pairs (Fig. 8).

Discussion

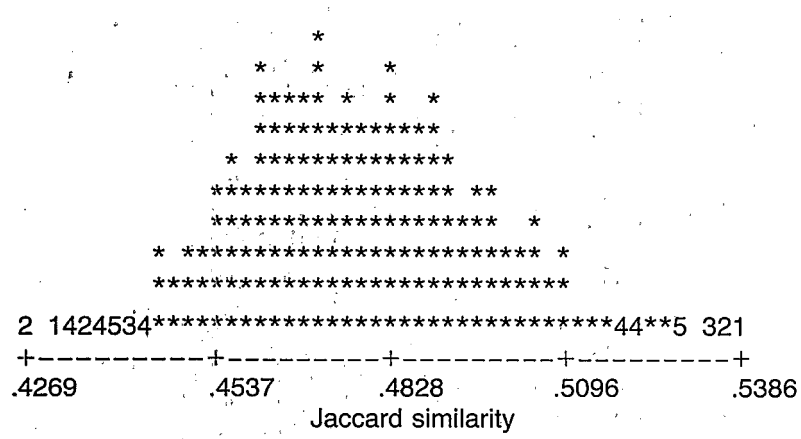
The structure of biological communities is vulnerable to a host of potential adaptations in response to environmental stressors ranging from pollutants, to physical stresses, to habitat modification. The analysis of populations within communities often presents equivocal data: some species increase in abundance while others decrease. In extreme cases, some species are locally extirpated, reducing community heterogeneity. In other cases, intolerant species are replaced by stress-tolerant forms.

The application of community similarity and related analyses provides a means for comparing sites under differing conditions. The above methods for critically examining replicate communities at differing sites or in multispecies experiments provide a means of inferentially locating and evaluating community change, and identifying the species influencing the change in community structure.

Data Type and Effect of Measure

Measures based on binary (presence–absence) data limit the ability of investigators to detect community differences. Analyses that place more weight on common species in community pairs (for example, Czekanowski) increase the absolute value of the similarity index, but provide no greater detection power for comparing communities. Additionally, the complements of these measures are typically semimetric and are, therefore, less useful indicators of relative difference (distance) among communities. Measures such as the Coefficient of

A



Total similarity	73.7955	mean	0.4823
Between similarity	64.3072	mean	0.4763
Within similarity	9.4883	mean	0.5271

Notes: The critical value is the 950th value.
The critical mean within similarity is 0.51545.
Reject hypothesis of no effect if this value is smaller than the observed mean within similarity.
Number > observed within 17.

B

	Treatment					
	1	2	3	4	5	6
1	1.0000	1.0051	.9690	.8659	.7022	.7608
2	1.0051	1.0000	1.0334	.8996	.6832	.7421
3	.9690	1.0334	1.0000	.9900	.9607	.9552
4	.8659	.8996	.9900	1.0000	1.0414	1.1360
5	.7022	.6832	.9607	1.0414	1.0000	.9639
6	.7608	.7421	.9552	1.1360	.9639	1.0000

Note: Lambda values for pairwise comparisons, ratios of mean between to mean within similarity. Values near 1 indicate little difference between groups.

Figure 3.—(A) Distribution of permutations of within similarity. (B) Pairwise comparisons (Lamda values) showing progressive differences between treatments.

Community Loss are nonmetric and require additional investigation before they can be recommended for detecting community differences. Measures that place emphasis on missing species are useful only when the base species list includes important indicator species.

Measures based on binary data require species loss and gain to detect differences in community structure and are not sensitive to changes in abundance. However, when the number of species examined is high (as for microbial communities), detection of community differences is greater that for less diverse assemblages such as fishes. Schindler (1987) has recently recommended small, rapidly reproducing species with poor dispersal capabilities as efficient environmental monitors. Additional evidence suggests that structural changes in ecosystems consistently process changes (Pratt, 1990).

Measures that incorporate ranked, relative, or absolute abundance as weightings for taxa in com-

A

Species removed		Influence
A	sp2	−4.675
C	sp2	1.087
C	sp3	1.417
D	sp1	1.459
F	sp1	−1.956
F	sp2	−1.885
N	sp1	−4.675
N	sp3	1.332
N	sp4	1.331
N	sp5	1.774
N	sp6	−1.109
P	sp1	1.330
S	sp1	1.492
S	sp2	1.508
S	sp3	1.544
S	sp6	−2.735

B

Trt.	Trt.	Critical value	Between similarity	
1	2	.38724	.73406	
1	3	.39258	.59541	
1	4	.39516	.52651	
1	5	.38613	.43858	
1	6	.39156	.47645	
2	3	.39058	.55150	
2	4	.39742	.47437	
2	5	.38703	.37148	*
2	6	.38590	.40479	
3	4	.38267	.40726	
3	5	.39143	.41105	
3	6	.39180	.41033	
4	5	.39739	.43892	
4	6	.38616	.48076	
5	6	.38786	.42378	

Note: The critical values below are for the multiple comparisons on the mean between similarity. Reject if the mean between similarity is smaller than the critical value. Asterisk (*) denotes detected difference.

Figure 4.—(A) Species influence scores for select species from Tables 1 and 2. Scores indicate the magnitude and direction of effect on community similarity when the species is removed from the analysis. (B) Pairwise tests of treatment differences based on between-treatment similarity. Differences are detected at p = 0.1.

munity samples have improved the ability to detect differences in structure. In these cases, measures are sensitive to changes in abundance, not simply to gain or loss of taxa. Distance measures are effective in detecting the separation of samples in multidimensional (multispecies) space; however, no rigorous analysis comparing the relative powers or sensitivities of these measures has been used to detect biotic community differences that result from stress.

Competing Analytical Methods

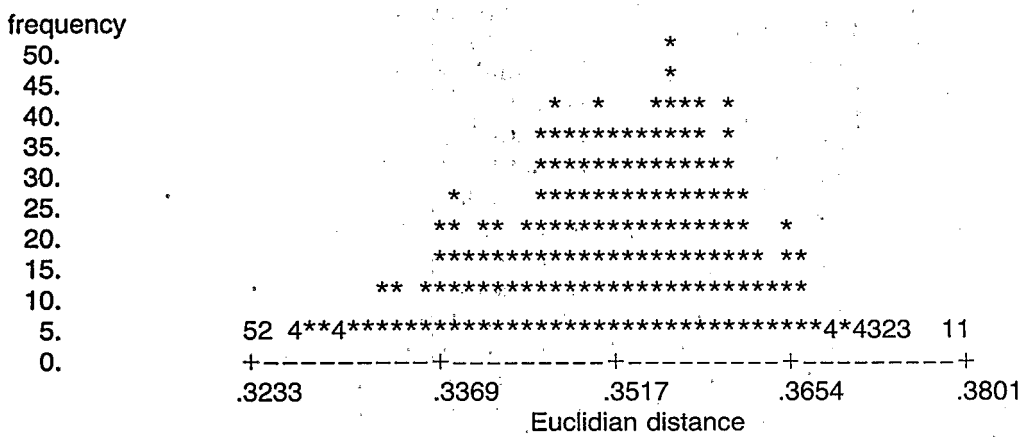
Several other methods for evaluating community structure data have been used to determine ecosystem health. These methods include calculation of diversity indices or indices of biotic integrity, and the comparison of communities using cluster analysis.

Diversity indices compare sites based on species abundance or some other measure of the distribution of individuals or biomass for species present. The most commonly used indices are based in information theory (for example, Shannon and Weaver, 1949). While these indices provide numerical values for diversity, including values for replicate samples at a site, they do not account for the identity of the component taxa. That is , two samples might have the same information theory diversity and share no species in common. Diversity indices have value in comparing sites, and the computed index is often strongly correlated with the number of taxa (Green, 1979), suggesting that species richness alone is a sufficient measure of diversity. The concept of species diversity is often ignored in investigations where problem taxa are pooled (for example, chironomid midge larvae) so that the resulting index is computed from a mixture of taxa that are comprised of species, genera, and families.

Indices of biotic integrity provide a score for each of several biological criteria. Unlike diversity indices, where the identity of the taxa is ignored, the identity of collected taxa is of primary importance. Indices such as the fish-based Index of Biotic Integrity (IBI), (Karr et al. 1986) or the Invertebrate Community Index (ICI) (Ohio Environ. Prot. Agency, 1987) are based on a combination of concepts. These indices are computed by assigning integer scores to community metrics that are conceptually categorized.

For example, the IBI separates metrics according to species richness (all taxa and some indicators), trophic composition, and abundance and condition of specimens. Similar concepts are used in the ICI, although there is more reliance on indicator taxa. The scores assigned are derived from a subjective rating system based on determination of re-

A



Total distance	53.9703	mean	0.3527
Between distance	48.0703	mean	0.3561
Within distance	5.9000	mean	0.3278

Notes: The critical value is the 51st value.
The critical mean within distance is 0.33505.
Reject hypothesis of no effect if this value is smaller than the observed mean within distance.
Number < observed within 7.

B

	Treatment					
	1	2	3	4	5	6
1	1.0000	.9830	1.0712	1.1715	1.3650	1.2963
2	.9830	1.0000	.9841	1.0835	1.2646	1.2149
3	1.0712	.9841	1.0000	1.0057	1.0219	1.0236
4	1.1715	1.0835	1.0057	1.0000	.9789	.9296
5	1.3650	1.2646	1.0219	.9789	1.0000	1.0208
6	1.2963	1.2149	1.0236	.9296	1.0208	1.0000

Note: Lambda values for pairwise comparisons, ratios of the mean between to the mean within distance. Values near 1 indicate little difference between groups.

Figure 5.—(A) Distribution of permutations of within-treatment distance for the example using binary data. (B) Pairwise comparisons.

gional optima (or expected values). Statistical comparisons are not possible, although community similarity to a reference site is one of the metrics used in the ICI. The term "metric" as applied to IBI and ICI is unfortunate because the assigned scores lack the characteristics of true metrics (Legendre and Legendre, 1983; Digby and Kempton, 1987).

Cluster analyses share many common features with methods of community similarity and distance analysis (see Pielou, 1984; Digby and Kempton, 1987). Both hierarchical and nonheirarchical clustering methods, like ordination procedures, are essentially graphic tools that allow visualization of community relationships. These methods provide no inferential analysis, but are simply ways of comparing several objects (samples) according to several descriptors (variables such as species or other characters).

As such, they may be useful companion tools to the analytical methods described in this paper. For most multivariate inferential procedures, the covariance matrix cannot be singular. An exception occurs

A

Species removed		Influence
A	sp2	.000
C	sp2	−2.157
C	sp3	−2.209
D	sp1	−1.896
F	sp1	−1.137
F	sp2	−1.155
N	sp1	.000
N	sp3	−2.142
N	sp4	−1.997
N	sp5	−2.048
N	sp6	−1.486
P	sp1	−1.954
S	sp1	−1.909
S	sp2	−1.928
S	sp3	−1.921
S	sp6	−.788

B

Trt.	Trt.	Critical value	Between distance	
1	2	.38717	.24979	
1	3	.38667	.31570	
1	4	.38612	.35068	
1	5	.38813	.38352	
1	6	.38776	.37036	
2	3	.39018	.32776	
2	4	.38646	.36588	
2	5	.38768	.40385	*
2	6	.38789	.39370	*
3	4	.38773	.38046	
3	5	.38783	.36781	
3	6	.39041	.37327	
4	5	.38657	.35689	
4	6	.39088	.34328	
5	6	.38875	.35822	

Note: The critical values below are for the multiple comparisons of the mean between distance. Reject if the mean between distance is larger than the critical value. Asterisk (*) denotes detected difference.

Figure 6.—Follow-up analyses. (A) Species influence scores based on Euclidian distance. (B) Pairwise tests of treatment differences based on between-treatment similarity. Differences are detected at p = 0.1.

when the number of samples is fewer than the number of variables (species), as is often the case in environmental analysis. Assumptions of normal distribution of variables are often violated when species abundances are zero at some sites.

Summary and Conclusions

Choice of data type and measure is important and should be based on the hypothesis being tested. The similarity measure used affects the outcome of analyses. The numerical value of the similarity measure is influenced by the functional form of the measure, transformations applied to the data prior to analysis, and the type of data. Choice of data limits the type of changes that can be detected. For example, presence–absence data are used primarily to assess changes in species composition. Abundance data, in contrast, reveal decreases or increases in the relative abundance of species.

With some measures, dominant species may strongly influence the measure. Changes in the relative abundance of a dominant species may not result in the loss of a species; thus, measures based on presence–absence may not reflect changes in the dominant species. In fact, if a species undergoes a large change but is not absent, the species may have a positive (stabilizing) effect in a presence–absence measure. On the other hand, changes that result in loss of species may not be reflected in measures based on proportional abundance unless they are accompanied by strong changes in the relative abundance (the total relative abundance of the species absent must be moderate when they are present).

Biological criteria for evaluating individual and cumulative effects of stresses might be based on comparisons with regional or upstream reference communities, although often no acceptable upstream corollary can be found. The usefulness of biological criteria for evaluating ecosystem health will be determined by the scientific adequacy of the analyses applied for detecting community change. At the present time, critical evaluations of analytical tools are needed to determine the appropriateness of available measures for detecting community change. Critical detections cannot be made by indices that are only descriptive and lack measures of variability.

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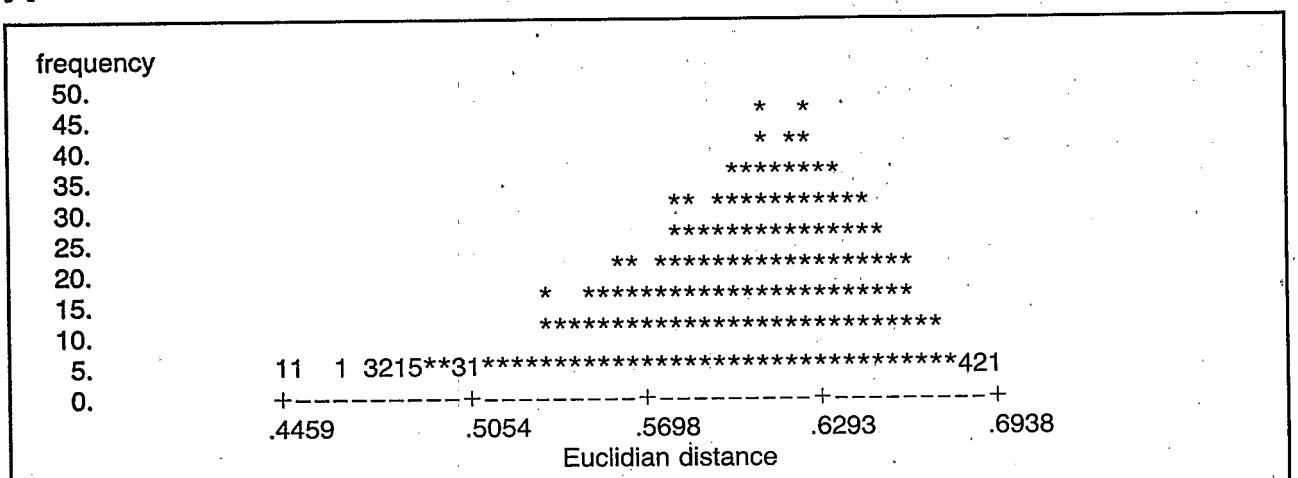
A



Total distance	92.8750	mean	0.6070
Between distance	86.1072	mean	0.6378
Within distance	6.7679	mean	0.3760

Note: The critical value is the 51st value.
The critical mean within distance is 0.52928.
Reject hypothesis of no effect if this value is smaller than the observed mean within distance.
Number < observed within 0.

B

	Treatment					
	1	2	3	4	5	6
1	1.0000	1.2057	1.5910	2.0498	2.8655	2.5655
2	1.2057	1.0000	1.1144	1.4945	2.2802	2.1458
3	1.5910	1.1144	1.0000	1.1155	1.6803	1.7064
4	2.0498	1.4945	1.1155	1.0000	1.3238	1.4395
5	2.8655	2.2802	1.6803	1.3238	1.0000	1.1116
6	2.5655	2.1458	1.7064	1.4395	1.1116	1.0000

Note: Lambda values for pairwise comparisons, ratios of the mean between to the mean within distance.
Values near 1 indicate little difference between groups.

Figure 7.—(A) Distribution of permutations of within-treatment distance for the example using rank abundance data. (B) Pairwise comparisons.

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A

Species removed		Influence
A	sp2	−5.915
C	sp2	−.674
C	sp3	−.746
D	sp1	−.592
F	sp1	−.387
F	sp2	−.337
N	sp1	−5.915
N	sp3	−.699
N	sp4	−.640
N	sp5	−.696
N	sp6	−.489
P	sp1	−.669
S	sp1	−.659
S	sp2	−.667
S	sp3	−.686
S	sp6	−7.015

B

Trt.	Trt.	Critical value	Between distance	
1	2	.78949	.30638	
1	3	.79348	.46888	
1	4	.77944	.63923	
1	5	.79077	.90158	*
1	6	.82022	.98559	*
2	3	.81480	.37119	
2	4	.79738	.52340	
2	5	.79219	.80488	*
2	6	.78752	.90666	*
3	4	.79658	.43593	
3	5	.79791	.66137	
3	6	.82326	.79031	
4	5	.79434	.54372	
4	6	.80127	.69137	
5	6	.78662	.53696	

Note: The critical values below are for the multiple comparisons of the mean between distance. Reject if the mean between distance is larger than the critical value. Asterisk (*) denotes detected difference.

Figure 8.—(A) Species influence scores based on Euclidian distance determined from rank abundance. (B) Pairwise tests of treatment differences based on between-treatment similarity. Differences are detected at p = 0.1.

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Errors in Errors in Hypothesis Testing

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ABSTRACT

Clear hypotheses are the basis for evaluating whether biological criteria are met in achieving environmental goals. Their logical structure reflects fundamental management concerns and scientific questions. Unfortunately, hypotheses are often tested or evaluated improperly in environmental studies. This paper demonstrates the importance of selecting the proper error term for statistical tests of impact or compliance. A straightforward, nontechnical discussion of the role of the error term in hypothesis tests shows how the choice of error term controls the question that is actually being asked. Simple simulation results demonstrate how the choice of the wrong error term can completely invalidate significance tests. This is an important issue for two reasons. First, the use of inappropriate error terms in environmental literature suggests that environmental scientists are not yet familiar enough with these concepts. Second, many packaged statistical programs automatically default to error terms that are not always appropriate, thus increasing the chance of producing erroneous hypothesis tests.

Introduction

Hypothesis testing is often considered one of the central activities of science, separating it from other technical and intellectual disciplines (Platt, 1964). The absence of clear hypotheses and unambiguous hypothesis tests forms the basis for much of the criticism of environmental studies (e.g., Beanlands and Duinker, 1983; Natl. Res. Counc. 1986, 1990; Fritz et al. 1980). Such critiques have resulted in an emphasis on planning environmental studies around null hypotheses that can provide the basis for conclusions about impacts and other changes.

This emphasis has, on the whole, been beneficial. However, too often null hypotheses are thought of as merely simplistic statements that express the opposite of an expected event. Thus, "the discharge will have no effect on fish populations" is typical of hypotheses of this type. At a somewhat more sophisticated level of study design, the importance of beta as well as alpha errors in hypothesis tests is taken into account (e.g., Henkel, 1976; Sokal and Rohlf, 1981). This has led to a recent focus on the utility of power tests and optimization analyses (e.g., Bernstein and Zalinski, 1983) that systematize the statistically efficient allocation of limited sampling resources.

Both these improvements have enhanced the rigor and utility of environmental studies. However, another fundamental issue of overriding importance affects study design and the ability to make logical inferences. This is the selection of the proper error term in hypothesis tests. The choice of error term controls the questions that are asked and affects, or should affect, the sampling or measurement design. In addition, inadvertently using the wrong error term can completely invalidate significance tests.

These ideas are not new and in fact are well known among statisticians. In spite of this, there is a consistent lack of attention to this issue and/or use of inappropriate error terms in environmental studies. This is particularly true in the gray literature of project reports and policy documents, suggesting that these ideas are perhaps inaccessible as presented in the statistical literature. They are therefore worth restating, in simpler terms and with straightforward examples that make them more understandable to practicing environmental scientists. Thus, as much as possible, this report avoids formal statistical terminology and definitions. Similarly, only the main ideas are presented, without the detail that a more developed treatment would include.

Error Terms Plain and Simple

The structure of the routine F test in the analysis of variance (ANOVA) illustrates some basic principles. The F test, which is based on the ratio of two variances, is analogous to a signal to noise ratio. The numerator of the test contains the signal plus background noise and the denominator of the test only the background noise (Equation 1).

$$F = \frac{\text{signal} + \text{noise}}{\text{noise}} \qquad \text{(Equation 1)}$$

If the numerator is larger than the denominator by a predetermined amount, it can be concluded that there is a change, impact, or effect (some kind of signal) that is not due to chance. Since this is a ratio test, the choice of the denominator term is critical.

The background noise in the F test is the variability in the system that stems from sources other than those that create the signal, but that could obscure, confound, or bias our perception of the signal. As one example, the success of a study of how some part of the environment responds to improvements in effluent treatment over a period of years could be affected by the background noise of natural temporal changes in the environment. Many important technical elements are involved in a formal statistical definition of the signal and noise terms. However, for these purposes, only the three general points are stressed.

1. The noise term can be made up of several kinds or sources of variability (see Fig. 1). Rarely does only one kind of variability contribute to the background noise in a system. For instance, in the effluent treatment example given, natural temporal variability can include diel, seasonal, yearly, and longer-term patterns, as well as less predictable disturbances.

2. The noise terms in the numerator and denominator of the F test should be identical, including the same sources of variability. The only difference between the variances in the numerator and denominator should be the presence of the signal term in the numerator. If the two noise terms are not identical, then the denominator will be artificially inflated or deflated and the significance test will be biased.

3. The sampling or measurement plan should be carefully designed to capture the important sources of background noise that cannot be controlled for. These cannot be included in the hypothesis test if they are not measured properly.

What is the Real Question?

Hypothesis tests can be thought of as evaluating questions about events or changes in the environment. Because they do this by comparing the potential signal to the background noise or error term (see Equation 1), different error terms result in different questions, even if the signal is identical in all cases. Lack of awareness of this fact leads to two related kinds of mistakes in environmental studies. First, researchers may in fact be asking one ques-

Table 1.—An illustration of how using different error terms in the significance test leads to actually evaluating quite different questions. Here, the signal stays constant and three alternative error terms are shown. The study design is described in Figure 1. The questions are deliberately constructed to reflect the signal to noise analogy of Equation 1. Thus, each question includes a description of the signal term and the background noise to which it is being compared. Changing the background noise changes the nature of the question.

STUDY DESIGN	SIGNAL	ERROR TERM	EQUIVALENT QUESTION
Figure 1	Condition X Location interaction	time	Question 1A
		time X location	Question 1B
		random sampling error	Question 1C

QUESTIONS

A: Is the change, from the before to the after, in the average difference between the control and impact locations larger than the random changes that occur over time in the study area as a whole?

B: Is the change, from the before to the after, in the average difference between the control and impact locations larger than the changes over time in the differences between locations?

C: Is the change, from the before to the after, in the average difference between the control and impact locations larger than the random sampling error at individual stations?

Elements of Sampling Plan	Schematic	Sources of Variation
• Sample impact and control locations		Condition: difference between the before and after conditions
• Sample several stations within each location	Impact Control Δ	Location: difference between control and impact locations
• Sample several instantaneous replicates at each station	Before: t_1 Δ : : t_n Δ	Condition X Location: change from the before to the after condition, in the difference between the control and impact locations
• Sample all stations several times in the before impact condition and also in the after impact condition	After: t_1 Δ : : t_n Δ	*Time: changes over time common to all stations
• Average data from the impact stations at each sampling time. Do the same for the control stations.		*Time X Location: changes over time in the differences between locations
• Calculate the difference between the impact and control averages to derive a difference score for each sampling time.		*Random Sampling Error: irreducible differences between samples collected at the same time and place

Figure 1.—The example study design used to illustrate the importance of selecting the proper error term. This study design is termed the BACI model (Before, After, Control, Impact) (Bernstein and Zalinski, 1983; Stewart-Oaten et al. 1986). The schematic shows the logical structure of the design, where the t's represent successive sampling times in each period and the Δ the difference between impact and control locations at each time. Important sources of variation are shown. Those marked by an asterisk are components of the background noise.

tion when they think they are asking another. Second, the question that in fact is being asked may be ecologically meaningless or irrelevant.

Figure 1 shows an example study design that is the basis for discussion in this section. Table 1 illustrates how specifying different error terms in this design leads to the evaluation of quite different questions. Typically, questions or null hypotheses specify only the signal term, equivalent to the first part of the questions in Table 1. In contrast to this, the complete questions in Table 1 include descriptions of both the expected signal and the background noise. Framing questions more thoroughly in this way makes it easier to identify and avoid the two kinds of mistakes described in the preceding paragraph.

In the example shown in Figure 1 and Table 1, the Condition x Location interaction is the numerator (or signal plus noise term) in the F test. This is because an impact has occurred if the difference between the impact and control locations, averaged over several times in the after condition, changes compared to the difference between the impact and control locations, averaged over several times in the before condition (Green, 1979; Bernstein and Zalinski, 1983; Stewart-Oaten et al. 1986). Figure 2 shows how this interaction, a change in the difference between control and impact, can be visualized. Even with this correct signal term, however, comparing it to the different error terms in Table 1 leads to quite different conclusions about whether an impact has occurred and about the nature of the impact.

For example, comparing this signal to the random sampling error indicates an impact has occurred if the Condition x Location interaction is large compared to the differences among samples taken at a single point in time and space. In contrast, using the Time x Location interaction (see Figs. 1 and 2) as the error term means that an impact has occurred only if the signal is large compared to the natural temporal variability in the differences between locations. This is more ecologically appropriate because it includes the natural temporal variability in the spatial comparison (between impact and control locations) that is the basis of the impact hypothesis (see Fig. 2).

Far from being an obscure statistical detail, choosing alternative error terms establishes quite different ecological criteria for deciding if an impact has occurred. By doing so, these different terms also establish quite different definitions of just what an impact is. In the first case, an impact will be any change larger than those that occur on a very small spatial scale at a single point in time. In the second case, an impact will be any change larger than those that occur between more widely spaced locations over short to moderate time scales. Since these alternative error terms will most likely differ in magnitude, a signal that would be a significant impact when compared to one would not be statistically significant when compared to the other.

The other potential error term shown in Table 1 raises similar issues. Using as the background noise the changes over time that are common to all stations (as suggested by Green, 1984, 1987) includes temporal variability but ignores the necessary spatial component of a study that compares two locations. It is easy to imagine a situation in which a large temporal change affects the entire study area (e.g., storm, El Nino, regional anoxia, population

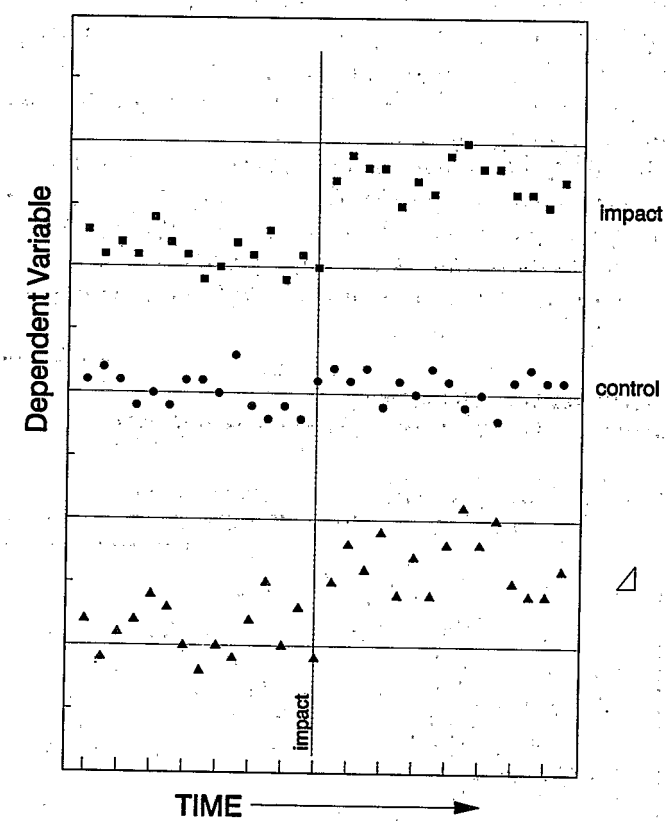

Figure 2.—Hypothetical representation of how values of a dependent variable would vary naturally through time at control and impact locations. As a result, the difference between the control and impact, designated by the Δ, would also vary. The variability in the difference or Δ is the Time x Location variance. The change in the Δ after the impact occurs is the Condition x Location interaction. In this design, the goal is to determine if the average Δ after the impact differs from the average Δ before the impact. The appropriate background noise for this test is the natural variability in the Δs, i.e., the Time x Location variance.

shifts) but does not erase the impact-related differences between control and impact locations. In this case, any decision about whether an impact exists depends entirely on the magnitude of the region-wide temporal changes, not on the size of the impact. The same size impact could be statistically significant or nonsignificant, depending on the extent of the temporal changes. This error term is therefore not very ecologically meaningful, since it compares the potential impact to a kind of variability that does not necessarily affect the difference between the control and impact locations.

Table 1 summarizes the use of these different possible terms in the context of the signal to noise ratio shown in Equation 1. The discussion of these examples shows that different error terms result in different ecological criteria for deciding what an im-

pact is. The examples also show that some of these criteria are not ecologically meaningful and that appropriate error terms must be selected carefully. Practitioners must also exercise care in this regard when using statistical software packages. In most packages, the default error term is the random sampling error and not all packages give users the option of defining error terms as needed.

When Significance Tests Lie

The preceding discussion has shown how the choice of error term influences the question that hypotheses tests are actually asking. This choice also affects the validity of statistical tests of significance. Tests performed with inappropriate error terms are essentially meaningless since the numerator of Equation 1 is being compared to an irrelevant denominator. Such faulty significance tests can be insidious since they appear outwardly well-founded. The following example presents a simple simulation study that illustrates the serious consequences of basing significance tests on incorrect error terms. It shows that using the incorrect error term can produce significant results a high percentage of the time even when the simulated data contain no impact.

The results of significance tests were simulated with the study design described (Figs. 1 and 2) using two different error terms. In each simulation, the data contained no impact, only random noise. In the first case, the correct error term was used, including both the Time x Location interaction and the random sampling error. In the second case, the interaction was omitted and the random sampling error was used alone. In both cases, the ratio between the random sampling error and the interaction was systematically varied. In the second case, this allowed investigation of what happens to the significance test as the error term reflects a greater or lesser proportion of the actual background noise in the data.

Line B in Figure 3 shows that the significance test using the correct error term finds significant results (i.e., a false impact) approximately 5 percent of the time, thus reflecting the true alpha level in the simulation of .05. Since the error term contains both components of the background noise, the test is not sensitive to their relative magnitudes. In contrast, Line A in Figure 3 shows that significance tests using only the random sampling error as the background noise, or error term, produced incorrect and highly variable results, even though the simulated data contained no impact.

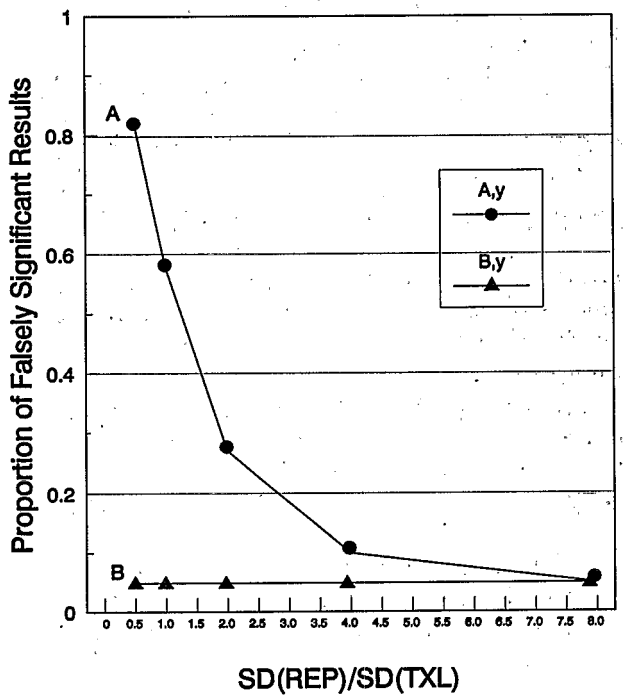

Figure 3.—Sensitivity of a significance test to two different error terms. Line A shows results of simulated significance tests using only the random sampling error (REP) as the error term. Line B shows results of simulated tests using the correct error term, including the standard deviation (SD) of both random sampling error (REP) and the Time x Location variability (TxL). The simulated data contain no impact, only random noise. Line A shows that the correct error term produces falsely significant results only 5 percent of the time, equal to the expected alpha level of false impacts. Line B shows that the rate at which the incorrect error term produces falsely significant results is extremely dependent on the ratio of the two background variabilities. (See text for further detail.)

The percentage of simulations that indicated a false impact depended on how much of the total background noise was made up by the random sampling error. Thus, when the random sampling error was roughly equal to the Time x Location interaction, the test produced falsely significant results a high percentage of the time (83 percent). It was not until the random sampling error became much larger than the Time x Location interaction (to the right on Fig. 3) that the percentage of falsely significant results fell and the test became more accurate. In this second case, the accuracy of the significance test depended, not on the size of any impact (there was none), but only on how much of the total background noise happened to be captured by the random sampling error.

This example demonstrates that a flawed significance test can produce a wide variety of results, depending on how inaccurate the error term is in

relation to the true background error. It should go without saying that statistical tests that produce such wildly variable results are worse than useless. They are dangerous because they can indicate an impact is present when in fact none has occurred.

Conclusions

Hypothesis testing is a critical step in determining whether environmental conditions have changed or whether compliance criteria have been met. Framing clear and specific null hypotheses helps in ensuring that hypothesis testing will produce valid and useful information. In this context, several points will help environmental scientists construct and carry out valid and powerful hypothesis tests.

1. Hypothesis tests are ratio tests, analogous to signal to noise ratios, in which the variance of an expected signal term is compared to the variance of a background noise term.

2. Null hypotheses will be more informative and accurate if they are framed in terms of both the expected signal and the background noise against which the signal will be compared.

3. The background noise, or error term, must be carefully structured to contain the correct components. If this is not done, the error term will be artificially inflated or deflated, leading to biased significance tests.

4. The choice of error term controls the question actually being asked by the significance test. Questions resulting from inappropriate error terms can be ecologically meaningless.

5. Using the wrong error term can lead to erroneous significance tests and to conclusions that an impact has occurred when in fact none exists.

While these concepts are familiar to statisticians, we have presented them, along with related examples, in straightforward and relatively nontechnical language. It is our hope that this will make them more accessible and understandable to environmental scientists who may not have the time or the background to interpret the statistical literature.

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The Integrated Biosurvey as a Tool for Evaluation of Aquatic Life Use Attainment and Impairment in Ohio Surface Waters

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ABSTRACT

The Ohio Environmental Protection Agency recently incorporated biological criteria ("biocriteria") into its water quality standards regulations. Numerical biological criteria were derived by utilizing the results of sampling conducted at "least impacted" regional reference sites. Fish and macroinvertebrate data from more than 300 Ohio reference sites were used to establish attainable, baseline expectations within the framework of an existing system of tiered aquatic life use designations. Attainment status is determined as being "full" (all biocriteria are met), "partial" (one organism group reflects attainment, but the other does not), or non (none of the biocriteria are met or one organism group reflects a poor or very poor condition). An attainment status table is constructed using these guidelines. The diagnosis of observed aquatic life use impairment relies on an integrated assessment of available biological, chemical, physical habitat, bioassay, pollution source, and general watershed information. This approach is employed extensively in the Clean Water Act section 305b reporting process and in support of regulatory program efforts. While all available biological and chemical criteria are utilized, considerable reliance is placed on the integrated interpretation of these data by the scientists who actually conduct the field sampling and evaluate the results. Detailed, site-specific knowledge of complex study areas in combination with these varied types of monitoring data is necessary to accomplish an environmentally accurate assessment. No single tool alone can accomplish this level and power of assessment. A common criticism of biosurvey information is that it lacks the ability to distinguish between different types, causes, and sources of impairment. The emergence of multimetric biological evaluation tools and a rigorous, standardized approach to field assessment has provided the detail necessary to establish biological response patterns and distinguish between general impact types. The Ohio Environmental Protection Agency is currently working to develop biological "response signatures" that consist of key response components of the biological data that consistently indicate one type of impact over another. Further refinement of this tool should have a profound influence on both site specific and statewide assessments and should be an important consideration in some of the biocriteria policy issues that are currently being debated.

Introduction

The monitoring of surface waters and evaluation of the biological integrity goal of the Clean Water Act have historically been dominated by nonbiological measures such as chemical/physical water quality (Karr et al. 1986). While this approach may have fostered an impression of empirical validity and legal defensibility, it did not sufficiently measure the ecological health and well-being of the aquatic resource. This point was demonstrated in a comparison of the abilities of chemical water quality criteria and biological criteria to detect aquatic life impairment based on ambient monitoring in Ohio. Of the 645 waterbody segments analyzed, biological impairment was evident in 49.8 percent of the cases where no violations of chemical water quality criteria were observed (Ohio Environ. Prot. Agency, 1990a).

While this "discrepancy" may be remarkable on the surface, the reasons are many and complex. Biological communities respond to and integrate a wide variety of chemical, physical, and biological variables in the environment, whether of natural or anthropogenic origin. These include several factors that chemical water quality criteria alone cannot adequately discriminate or detect; two examples are the habitat and siltation. Often it is the cumulative combination of chemical and physical factors that result in impaired biological community structure and function.

The Ohio Environmental Protection Agency recently adopted biological criteria in its water quality standards regulations. These biocriteria are based on a system of tiered aquatic life uses from which numerical criteria were derived using a regional reference site approach (Ohio Environ. Prot. Agency, 1987, 1990b). The numerical expressions of biological goal-attainment criteria are essentially the end product of an ecologically complex derivation and assessment system. While numerical biological indices have been criticized for oversimplifying complex ecological processes, the need to distill such information to commonly comprehended expressions is both practical and necessary. The advent of "new" generation evaluation mechanisms such as the Index of Biotic Integrity (IBI) (Karr, 1981; Fausch et al. 1984; Karr et al. 1986), the Index of Well-Being (Iwb) (Gammon, 1976; Gammon et al. 1981), the Invertebrate Community Index (ICI) (Ohio Environ. Prot. Agency, 1987) have filled important theoretical gaps left by previous indices.

Such multimetric evaluations extract ecologically relevant information from biological community data while preserving the opportunity to analyze such data on a multivariate basis. The problem of biological data variability is also addressed within this system. Variability is controlled by specifying standardized methods and procedures, compressed through the application of multimetric evaluation mechanisms, and stratified by accounting for regional and physical variability and potential. This has yielded evaluation mechanisms such as the IBI and ICI that have acceptably low, replicate variability (Rankin and Yoder, 1990).

Ecoregional Biocriteria and Determination of Use Attainment

Biological criteria in Ohio are based on two principal organism groups, fish and macroinvertebrates. Numerical biological criteria for rivers and streams were derived by utilizing the results of sampling conducted at more than 300 "least impacted" reference sites. This information was used within the existing framework of tiered aquatic life uses to establish attainable, baseline expectations on a regional basis. Resultant criteria for two of the "fishable, swimmable" uses, Warmwater Habitat (WWH) and Exceptional Warmwater Habitat (EWH), are shown in Figure 1.

Procedures for determining the use attainment status of Ohio's lotic surface waters were also developed. Using the numerical biocriteria as defined by the Ohio water quality standards, use attainment status is determined as follows:

- **FULL** — use attainment is considered full if all of the applicable numeric indices exhibit attainment of the respective biological criteria.
- **PARTIAL** — at least one organism group exhibits nonattainment of the numeric biocriteria, but no lower than a "Fair" narrative rating, and the other group exhibits attainment.
- **NON** — none of the applicable indices exhibit attainment of the regional biocriteria; or, one organism group reflects a "Poor" or "Very Poor" narrative rating, even if the other group exhibits attainment.

A use-attainment table based on these rules is constructed on a longitudinal mainstem or watershed basis. Data included in the table are sampling location (river mile index), biological index scores, the Qualitative Habitat Evaluation Index (QHEI) score, attainment status, and comments about important site-specific factors such as proximity to pollution sources. The following examples demonstrate the use of the biological criteria as an assessment tool and the overall biosurvey design as an integrated diagnostic approach.

Blacklick Creek

Table 1 shows a completed attainment table for Blacklick Creek located in the East Corn Belt Plains ecoregion of central Ohio. The lower section of this stream is impacted by a privately owned and poorly operated wastewater treatment plant. The results are typical of a small stream (50 sq. mi. drainage area) impacted by municipal sewage—full attainment upstream, nonattainment downstream, with eventual recovery to partial and full attainment. Field observations included sewage

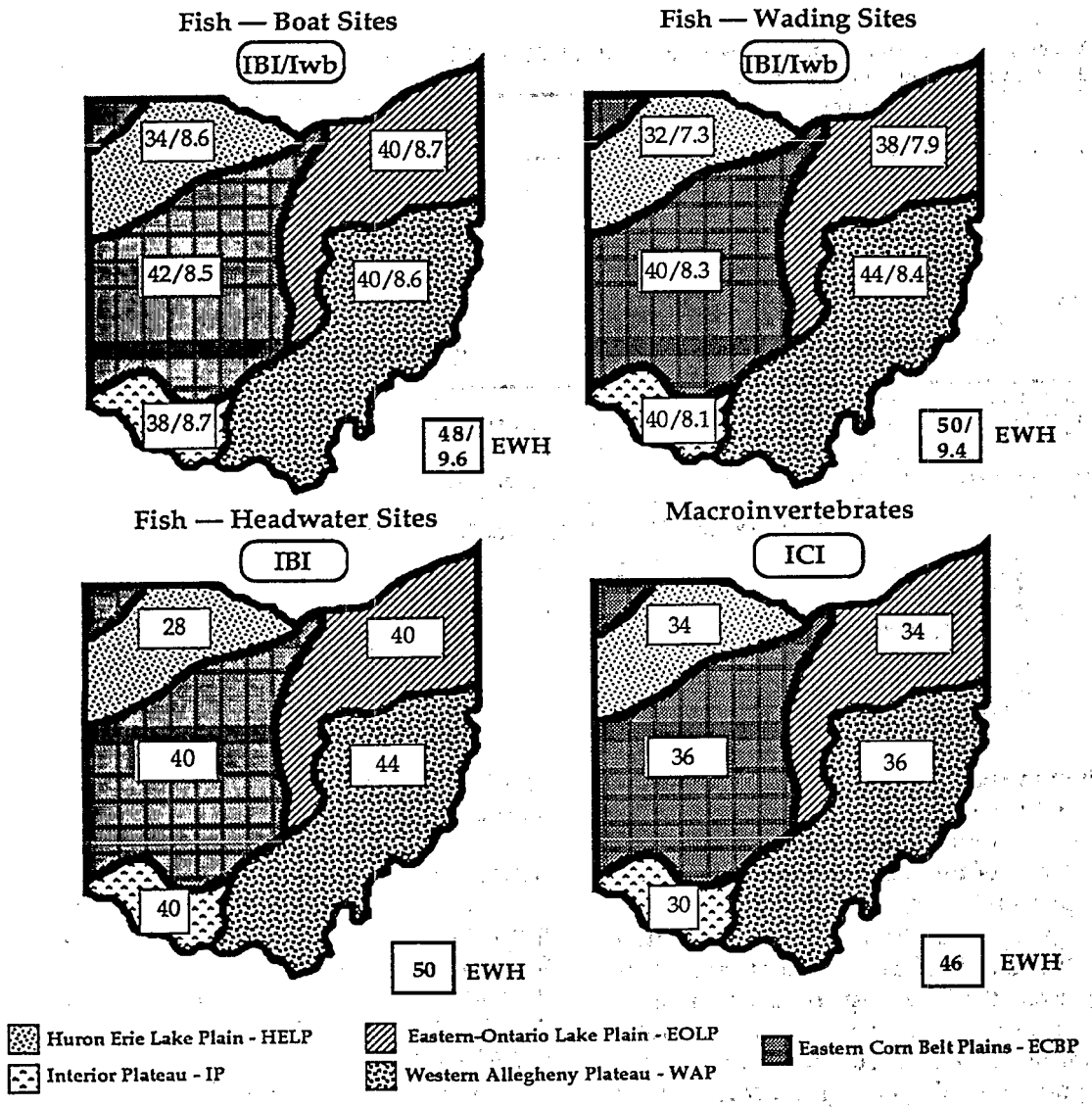


Figure 1.—Ohio biological criteria for the Warmwater Habitat (WWH) and Exceptional Warmwater Habitat (EWH) use designations arranged by biological index, site type for fish, and ecoregion. Index values on each map are the WWH biocriteria that vary by ecoregion as follows: IBI/MIwb for Boat Sites (upper left), IBI/MIwb for Wading Sites (upper right), IBI for Headwater Sites (lower left), and the ICI (lower right). The EWH criteria for each index and site type appear in the boxes located outside of each map.

sludge deposits, elevated ammonia-nitrogen, and continuous dissolved oxygen concentrations that were depressed below applicable water quality criteria. Extensive experience with this type of impact, the good correlation of the biological impairment with the dissolved oxygen profile, and the proximity of the source to the observed impairment made diagnosis relatively easy. Localized habitat alterations were not a predominant factor in the results.

Wills Creek

Table 2 shows results from Wills Creek in the eastern coal-bearing region of Ohio (W. Allegheny Plateau ecoregion). The upper watershed is impacted by runoff from surface mining, resulting in the extensive siltation of the substrates. The mainstem is additionally impacted by two municipal wastewater treatment plants and several small industries. The extensive siltation from the nonacidic mine runoff combined with the relatively low gradient results in an overlying physical impact that masks most of the influence of the point sources. While some localized upstream/downstream patterns are evident (e.g., ICI downstream Byesville), the overall pattern of nonattainment of the Warm Water Habitat (WWH) use is affected by the predominance of diffuse sources. Particularly important in diagnosing this situation was the Qualitative Habi-

Table 1.—Aquatic life use attainment status for the Warmwater Habitat (WWH) use designation in Blacklick Creek and Big Walnut Creek, July–October 1986.

RIVER MILE FISH/INVERT.	MODIFIED IBI	MODIFIED Iwb	ICI[a]	QHEI[b]	ATTAINMENT STATUS[c]	COMMENT
Blacklick Creek						
5.6/4.8	45	9.3	G	85	Full	Upstream Blacklick est. WWTP
4.7/4.7	33*	8.8	F	46	Partial	WWTP mixing zone
3.3/3.6	27*	6.7*	18*	52	Non	Downstream Blacklick est. WWTP
2.1[d]/2.1	32*	7.98*	26*	55	Non	Partially impounded
Big Walnut Creek						
15.8[d]/15.9	41	9.6	44	77	Full	Upstream Blacklick Creek
14.9[d]/—	28*	7.9*	—	63	(Non)	Downstream Blacklick Creek
—/12.8	—	—	46	—	(Full)	Downstream Blacklick Creek

[a]Narrative criteria used when ICI is not avialble (G = Good; F = Fair).
[b]All Qualitative Habitat Evaluation Index (QHEI) values are based on the most recent version (Rankin, 1989).
[c]Use attainment status based on one organism group is parenthetically expressed.
[d]Boat site for fish; all other fish sites are wading type.
*Significant departure from ecoregion biocriteria; "poor" and "very poor" results are underlined.

Ecoregion Biocriteria: E. Corn Belt Plains (ECBP)

Index—Site Type	WWH	EWH	MWH[1]
IBI—Wading	40	50	24
IBI—Boat	42	48	24
Mod. Iwb—Wading	8.3	9.4	6.2
Mod. Iwb—Boat	8.5	9.6	5.8
ICI	36	46	22

[1]Modified Warmwater Habitat for channel-modified areas.

tat Evaluation Index (QHEI) (Rankin, 1989) component scores which revealed the extensive disturbance of the Wills Creek substrates. Also important was a knowledge of the sources and the biological response in proximity to each.

The foregoing examples are only two of hundreds of evaluations that have been performed over a 12-year period. While biological data remain an indispensable item of information, chemical water quality, physical habitat, and source information are equally important in diagnosing probable causes and sources of impairment. Each tool, however, has its strengths and weaknesses. It is important to learn and recognize the relative virtues of each in order to assure a complete and accurate assessment.

Broad Scale Assessments

One way to gain additional insight into the problem of stream use nonattainment is to examine the water body for broad-scale patterns. Often a failure to look for such patterns can lead to an over-analysis of site-specific problems. This is akin to the adage of "not being able to view the forest because of the trees." Broad-scale concepts are very useful in site-specific problem analysis because they usually emanate from robust data bases where background "noise" is suppressed by the sheer volume of data. An example of this with the Ohio Environmental Protection Agency biological data is the distribution of sites that reflect "Poor" and "Very Poor" biological performance based on the IBI, modified Iwb, and ICI (Fig. 2). The first impression of the resulting distribution is the tendency for concentrations of sites yielding biological index scores commensurate with the "Poor" and "Very Poor" narrative descriptions to "cluster" in heavily industrialized areas of the state. This development would include industries such as steel making, rubber and plastics, petroleum refineries, glass making, and electroplating. In Ohio this pattern occurs in the northern half of the State.

Conversely, there seems to be an absence of clusters in the southern half of Ohio, particularly near Columbus and Dayton, two of the larger population centers in the state. Both cities lack the heavy industrial development common to the other areas. Pollution impacts in these areas are dominated by conventional sewage releases, both from wastewater treatment plants and combined sewers. Additional evidence that helps to establish a causal linkage to these observations is a map showing the distribution of sediment chemistry results that reflect highly elevated and extremely elevated concentrations of heavy metals (Fig. 2). A general correspondence to the clusters of "Poor" and "Very Poor" biological sites is evident. This information likely reinforces what most agency personnel have known, or at least suspected, for some time. The example illustrates the comparative usefulness of different biological and chemical assessment tools on a broad scale, particularly in an integrated application. While this type of assessment does not address

Table 2.—Aquatic life use attainment status for the Warmwater Habitat (WWH) use designation in the Wills Creek mainstem, July–September 1984.

RIVER MILE FISH/INVERT.	MODIFIED IBI	MODIFIED Iwb	MODIFIED ICI	QHEI[a]	WWH ATTAINMENT STATUS	COMMENT
75.9/75.8	33*	7.7*	30*	52	Non	Upstream all point sources
74.0/71.0	24*	5.8*	34[ns]	34	Non	Upstream Byesville WWTP
68.1/68.1	22*	5.3*	14*	41	Non	Downstream Byesville WWTP
66.5/66.7	29*	7.0*	16*	33	Non	
65.3/65.1	28*	6.4*	18*	38	Non	Downstream Natl. Cash Register
62.4/62.7	27*	6.9*	22*	48	Non	
61.8/ —	22*	5.7*	—	54	(Non)[b]	Downstream sewer line break
60.7/60.1	25*	7.7*	28*	52	Non	Downstream Cambridge WWTP
58.4/58.6	24*	6.3*	20*	37	Non	Downstream Crooked Creek
56.4/56.5	26*	6.6*	20*	42	Non	
53.5/53.5	29*	7.8*	34[ns]	55	Partial	
46.6/46.6	26*	6.2*	22*	42	Non	Downstream Salt Fork
37.7/ —	28*	6.5*	—	39	(Non)[b]	
27.0/ —	26*	5.8*	—	37	(Non)[b]	Downstream numerous mines

[a]All Qualitative Habitat Evaluation Index (QHEI) values are based on the most recent version (Rankin, 1989).
[b]Use attainment status based on one organism group is parenthetically expressed.
*Significant departure from ecoregion biocriteria; poor and very poor results are underlined.
[ns]Nonsignificant departure from ecoregion biocriteria (4 IBI or ICI units; 0.5 Iwb units).

Ecoregion Biocriteria: Western Allegheny Plateau (WAP)

Index—Site Type	WWH	EWH	MWH[1]
IBI–Boat	40	48	24
Mod. Iwb–Boat	8.6	9.6	5.5
ICI	36	46	30

[1]Modified Warmwater Habitat for mine-affected areas.

Source: Ohio Environ. Prot. Agency (1990b).

site-specific problems, it does provide the conceptual support necessary to operate a "case-specific" regulatory program, such as the issuance of National Point Discharge Elimination System (NPDES) permits.

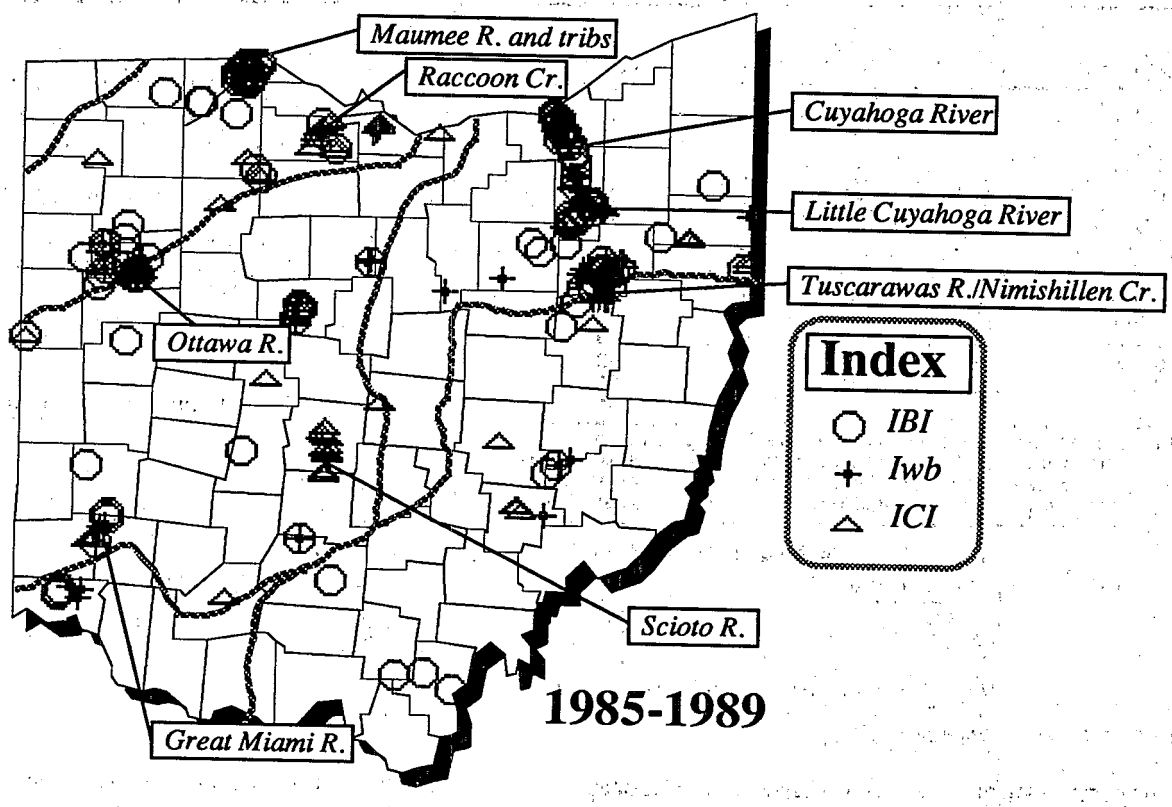
The strength of the biological data is the information it provides about whether or not an impairment exists and its severity. Often, this is assumed to be the limit of the usefulness of biological data. Prior to the development of many recent concepts and tools (i.e., regional reference sites, multimetric indices) this may have been true. However, much more of the biological information can now be utilized in multivariate approaches that begin to "sort out" and communicate important patterns of biological community response termed "biological response signatures."

Biological Response "Signatures"

The availability of a comprehensive, standardized ambient biological database from a variety of environmental settings has permitted certain patterns and characteristics of biological community response to be identified. A common criticism of ambient biological survey data has been its inability to determine the cause or source of an impaired condition. While this is probably valid for some of the traditional diversity indices (e.g., Shannon indices), number of species, biomass, and other single-dimension indices, it does not apply equally to the "new" generation multimetric indices such as the IBI and Invertebrate Community Index. When the response patterns of the various metrics and components of these indices were examined from areas where the predominant impairment causes and sources are well known, some consistent patterns emerged. Unique combinations of biological community characteristics that identify one impact type over others are referred to as "biological response signatures." These proved valuable in assigning causes and sources to the aquatic life use impairments analyzed in the 1990 305(b) report (Ohio Environ. Prot. Agency, 1990a).

A database including 25 similarly sized streams and rivers (drainage area range 90-450 square miles) from the Eastern Corn Belt Plains (ECBP) and Huron/Erie Lake Plain (HELP) ecoregions was arranged. Sampling generally took place between 1982 and 1989 and followed Ohio Environmental Protection Agency procedures (Ohio Environ. Prot. Agency, 1987, 1989a,b). General impact types were assigned to each sampling site as follows:

1. **Complex Municipal/Industrial:** This includes impacts from the complex combination and interactions of major municipal wastewater treatment plants and industrial point sources that comprise a significant



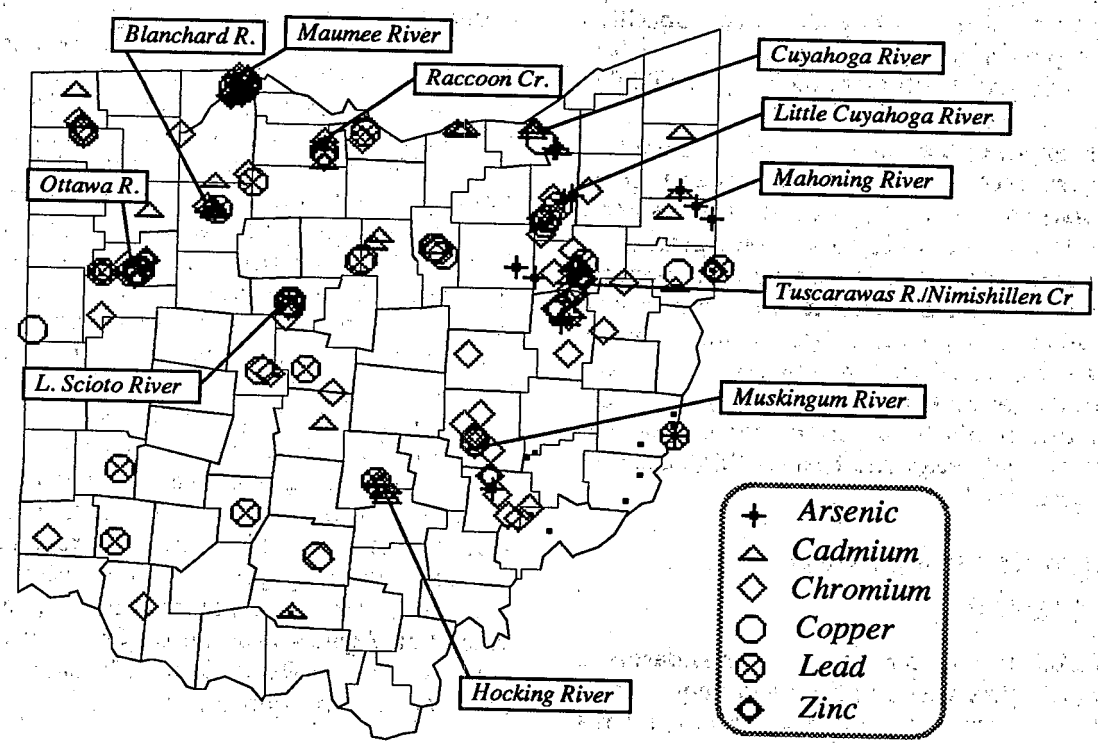


Figure 2.—Distribution of biological sampling sites with at least one biological index value reflecting "Poor" or "Very Poor" performance (upper) and sediment chemistry sites with highly elevated or extremely elevated heavy metal concentrations.

fraction of the summer base flow of the receiving stream and where one or more of the following have occurred: serious instream chemical water quality impairments involving toxics; recurrent whole effluent toxicity; fish kills; and severe sediment contamination involving toxics. This may include areas that have combined sewer overflows and/or urban areas located upstream from the point sources.

2. **Conventional Municipal/Industrial:** This includes impacts from municipal wastewater treatment plants that discharge conventional substances and where no serious or recurrent whole effluent toxicity is evident (these may or may not dominate stream flows). It may also include impacts from small industrial discharges that may be toxic, but that do not comprise a significant fraction of the summer base flows; other influences such as combined sewer overflows and urban runoff may be present upstream from the point sources.

3. **Combined Sewer Overflows/Urban:** Included are impacts from combined sewer overflows and urban runoff within cities and metropolitan areas that are in direct proximity to sampling sites. This includes both free-flowing and impounded areas upstream from the major wastewater treatment plant discharges. Minor point sources may also be present in some areas.

4. **Channelization:** Areas impacted by extensive, large-scale channel modification projects and where little or no habitat recovery has taken place comprise this impact type. Some minor point source influences may be present.

5. **Agricultural Nonpoint:** This includes areas that are principally impacted by runoff from the row crop agriculture that is the predominant land use in the ECBP and HELP ecoregions. Some minor point source and localized habitat influences may be present.

6. **Other:** Includes impacts not mentioned above — i.e., quarries, sand and gravel excavation, sanitary landfills, and flow alterations (immediate tailwater areas below dams).

One of these impact types was assigned to each of 225 sites sampled for fish and 111 sites sampled for macroinvertebrates. Assignments were based on the predominant impact that was directly influencing the site at the time the sampling took place. The assignments were based on the site-specific knowledge of the study area gained by the Ohio Environmental Protection Agency while conducting biological surveys of the 25 streams and rivers. The extent of spatial overlap between different impact types throughout this database is somewhat variable. However, the key objective of this analysis was to determine whether or not the feedback from the biological community can communicate about and characterize these differences.

Some preliminary results of this ongoing project are provided in Figures 3 and 4 and Table 3. The work thus far has concentrated on two- and three-dimensional analyses of IBI, MIwb, and ICI metrics and sub-components. Analysis of "smaller" community components (e.g., species level) is also being attempted. An example of one analysis is portrayed in Figure 3. Three components of the fish community data are included in a three-dimensional plot that illustrates the concept behind examining the combinant biological response characteristics of each impact type. The IBI, MIwb, and the frequency (percentage) of deformities, eroded fins, lesions, and tumors (DELT) on individual fish are different expressions of the relative health of the fish community at a given location. The Complex Toxic impact type (1) was compared on a three-dimensional basis to each of the five other impact types (2 through 6).

In each comparison the Complex Toxic impact type exhibited a fairly distinct pattern as compared to the other impact types. The amount of overlap was least with the Agricultural Nonpoint Source type (5) and greatest with the CSO/Urban impact type (3). The response characteristics of the Complex Toxic impact type generally include an IBI of less than 20-25, MIwb of less than 5.0-6.0, and DELT anomalies greater than 5-10 percent.

While other impact types may have one or two of these characteristics in common, very seldom do they have all three. One sample in the Agricultural Nonpoint Source type possessed all three of these characteristics and clustered with the Complex Toxic impact type. Upon further investigation it was learned that this particular site was downstream from an experimental "no-till" agricultural demonstration plot where pesticide usage was atypical. The resultant biological response confirmed that this particular impact fit the Complex Toxic impact type both biologically and culturally.

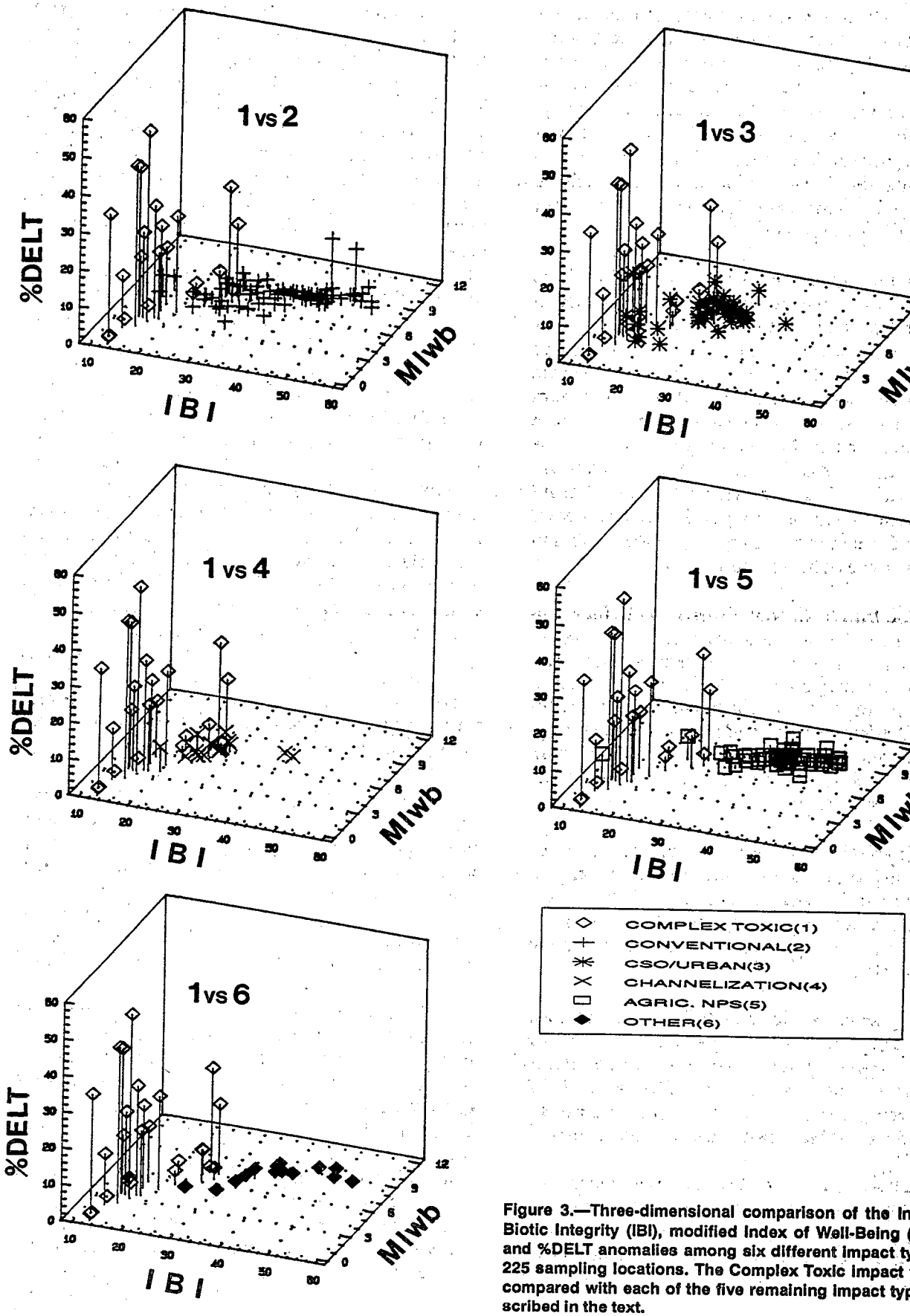


Figure 3.—Three-dimensional comparison of the Index of Biotic Integrity (IBI), modified Index of Well-Being (MIwb), and %DELT anomalies among six different impact types at 225 sampling locations. The Complex Toxic impact type is compared with each of the five remaining impact types described in the text.

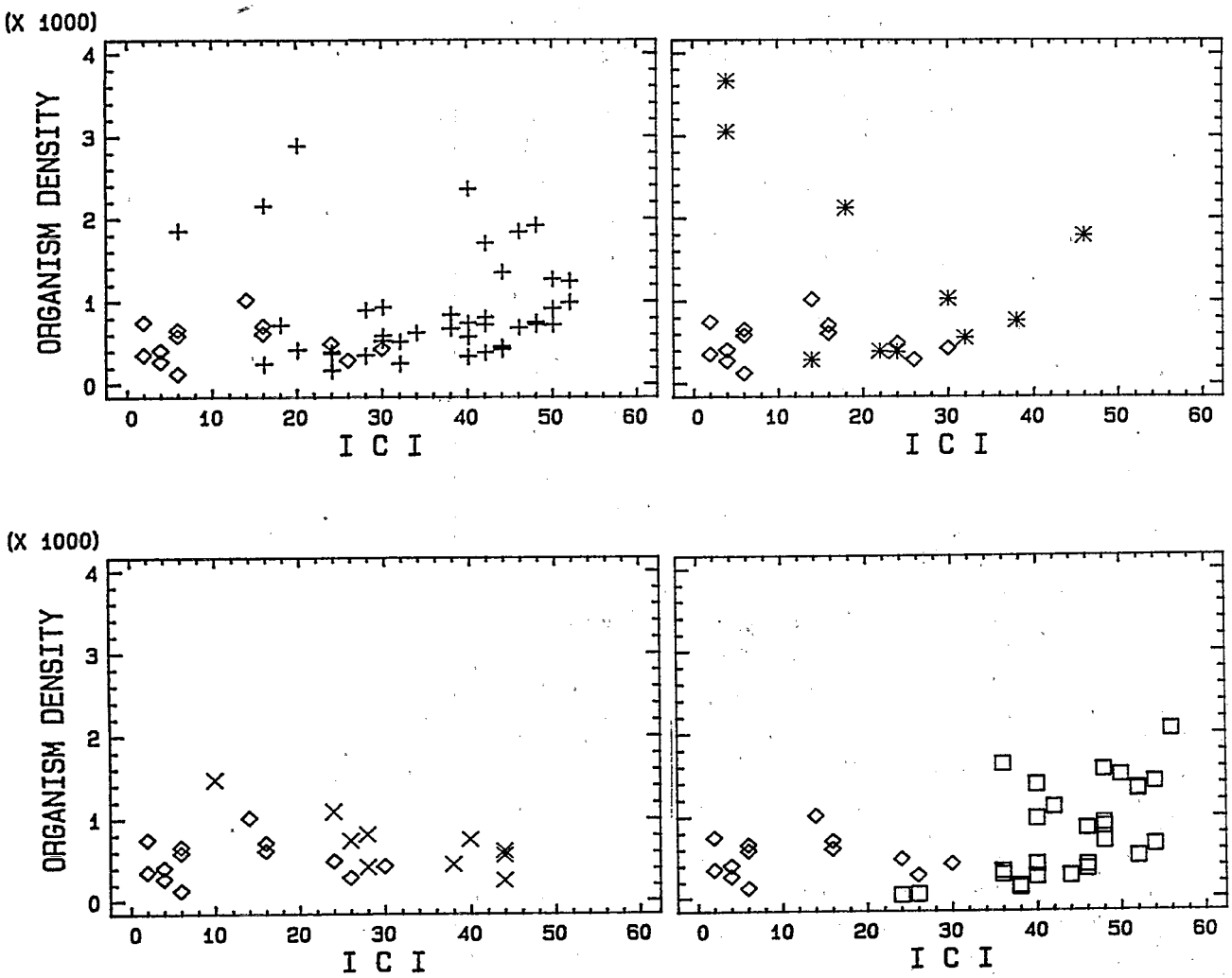


Figure 4.—Two-dimensional comparison of the Invertebrate Community Index (ICI) and organism density between the Complex Toxic (◇) and Conventional Municipal (+), CSO/Urban (*), Channelization (X), and Agricultural NPS (□) impact types at 106 sampling sites.

Some of the CSO/Urban impacted sites overlapped into the Complex type cluster in terms of the IBI and MIwb, but much less so in terms of percentage of fish exhibiting DELT anomalies. Some of these sites are located in areas with significant industrial sources that discharge into the municipal sewer system and that may have "mimicked" the Complex Toxic impact type. This example highlights the need to rely more on the biological response signature to characterize an environmental impact rather than the traditional process of cultural impact characterization based on chemical, physical, and process characteristics alone.

Table 3 shows the distribution of fish community index scores by impact type for the five narrative performance classes (Ohio Environ. Prot. Agency, 1987, 1990c). Sampling sites predominantly affected by the Complex Toxic impact type were most frequently in the "Very Poor" (55 percent) performance category, followed by the CSO/Urban impact type (23 percent). The highest narrative category reached by Complex Toxic impacted sites was "Fair" (18 percent). It should be noted that the Fair sites in this impact category were the farthest downstream from the major sources and represent the initial recovery along the longitudinal profile. The Conventional impact type had no sites in the "Very Poor" range and along with the Agricultural NPS and Other impact types, was the only type to have sites that attained the "Exceptional" performance level (equivalent to the EWH use). These three impact types also had the highest number of

Table 3.—Distribution of biological sampling results from 225 sites between the five narrative biological performance categories for the Index of Biotic Integrity and Modified Index of Well-Being for six major impact types.

IMPACT TYPE	VERY POOR	POOR	FAIR	GOOD	EXCEPTIONAL
1 Complex Toxic	55% (n=12)	27% (n=6)	18% (n=4)	0	0
2 Conventional Muni./Ind.	0	13% (n=10)	44% (n=35)	34% (n=27)	9% (n=7)
3 CSO/Urban	23% (n=9)	10% (n=4)	67% (n=27)	0	0
4 Channelization	7% (n=1)	33% (n=5)	60% (n=9)	0	0
Agricultural NPS	2% (n=1)	2% (n=1)	29% (n=15)	52% (n=27)	15% (n=8)
6 Other	6% (n=1)	18% (n=3)	23% (n=4)	47% (n=8)	6% (n=1)

Source: Ohio Environ. Prot. Agency (1990a).

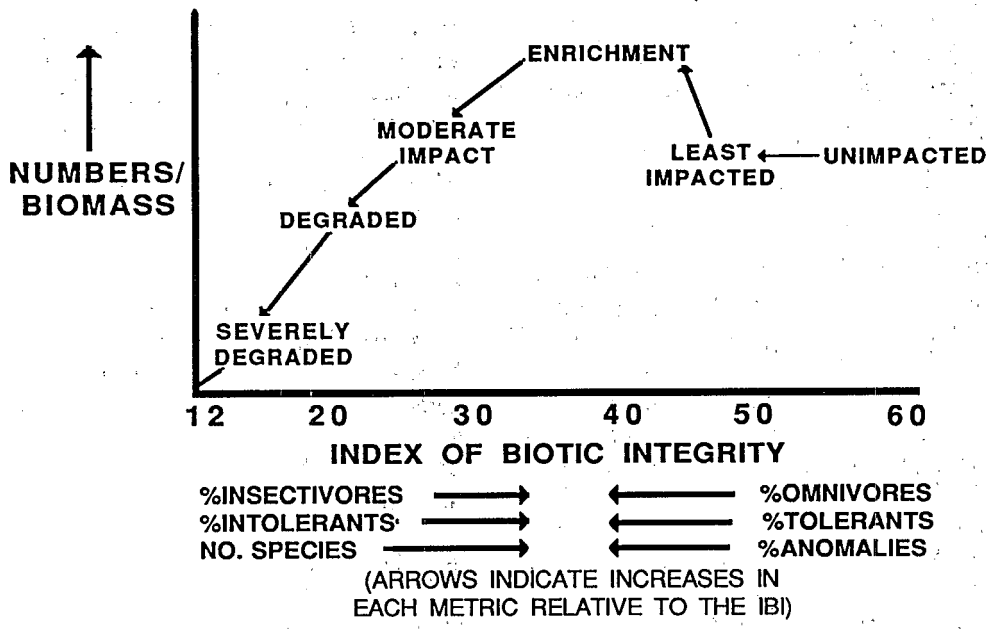
sites in the "Good" performance level (equivalent to the WWH use). The CSO/Urban and Channelization impact types, along with the Complex Toxic type, had no sites attaining the "Good" or "Exceptional" performance levels.

Using the macroinvertebrate community, a comparison of the relationship between the Invertebrate Community Index and organism density (number/square feet) demonstrates the need to access community information beyond the index result or the metrics that comprise the index. A comparison of the Complex Toxic impact type with the Conventional Municipal, CSO/Urban, Channelization, and Agricultural NPS impact types was made in a two-dimensional framework. In the comparison of the Complex Toxic and CSO/Urban impact types the ICI alone yields equally low results for each type of impact. Thus, this index alone was not able to discriminate the impacts. However, organism density, which is not a direct component of the ICI, yielded an improved separation of the two impact types (Fig. 4). This was also true in comparisons with the Conventional Municipal and Channelization impact types. The ICI alone separated most of the Agricultural NPS impacts. Thus, it is important to experiment with other aggregations of the community data that are not direct metrics of the indices used as the biological criteria for each organism group.

Although the Complex Toxic impact type separates well from the other impact types in this analysis, more overlap exists between the other five types. For example, the statistics in Table 3 are similar for the Conventional and Agricultural NPS impact types. Scatter plots also show a great deal of overlap. This is not surprising since the chemical and physical manifestations of each are functionally similar in the aquatic environment. Nevertheless, some differences may exist and are likely discernible by using more complex and iterative analyses than those demonstrated in Figures 3 and 4, and Table 3.

The Ohio Environmental Protection Agency is currently cooperating with Bolt, Beranek, & Newman, Inc., to evaluate techniques by which some of these more subtle differences might be defined using biological response signatures (Anderson et al. 1990). This involves the use of genetic algorithms employing artificial intelligence and machine learning techniques. One initial finding was the utility of one of the IBI metrics used for the headwaters site-type sensitive species. This metric combines the intolerant metric of the wading and boat site types with moderately intolerant species (Ohio Environ. Prot. Agency, 1987). This aggregation of the community data was by itself found to consistently indicate the **Complex Toxic** impact type with a reliability of 82 percent in stream and river sizes outside of its designed use in the Headwaters IBI.

Another way to describe the attributes of ambient biological data for characterizing different types of environmental impacts is with a conceptual model. Figure 5 shows a model of the response of a fish community to increasing stress from a "least impacted" to "severely degraded" condition. The comparison of numbers and/or biomass with the IBI shows this conceptual relationship. Beneath the graphic are narrative descriptions of biological com-



Conceptual Model of Community Response - Narrative Descriptions

Attributes	Severely Degraded (*Very Poor*)	Degraded (*Poor*)	Moderate Impact (*Fair*)	Enrichment (*Good*)	Least Impacted (*Exceptional*)
1) Community Condition Characteristics	No community organization - few or no species very low numbers only most tolerant, high % anomalies	Poorly organized community - few species, low numbers, tolerant species only, many anomalies	Reorganized community - tolerant species, intolerants in v. low numbers, omnivores predominate	Good community organization - good numbers of sensitive species, some intolerants	Highly organized community - insectivores, top carnivores, intolerants predominate, high diversity
2) Chemical Conditions	Acutely toxic chemical conditions *and/or*	Low D.O. with chronic toxicity *and/or*	Low D.O., nutrient enriched, no recurrent toxicity	Adequate D.O., no acute/chronic effects, elevated nutrients	No effects evident, background conditions, good D.O.
3) Physical Conditions	Total habitat loss, extremely contaminated sediments	Severe habitat degradation, severe sediment contamination	Modified stream channel, heavy siltation, canopy removal	Good habitat, no significant channel modifications	Excellent habitat, no modifications evident
4) Examples of Perturbations	Toxic discharges, dessication, acid mine drainage, severe thermal conditions	Municipal and industrial discharges, intermittent acute impacts	Municipal sewage, combined sewers, heavy agricultural use, non-acid mine drainage, moderate thermal increases	Minor sewage inputs, most agricultural non-point affected areas	No perturbations evident

Figure 5.—Conceptual model of the response of the fish community as portrayed by the Index of Biotic Integrity and other community metrics with narrative descriptions of impact types and corresponding narrative biological performance expectations.

munity characteristics, chemical conditions, physical conditions, and examples of environmental perturbations that are typical of biological community response across the five narrative performance classes. These are necessarily general and are not invariable. However, this model was developed from the Ohio Environmental Protection Agency's experience in analyzing biological, chemical, and physical data over a 12-year period and on a statewide basis. Thus, the model has a good foundation in the observation of actual environmental conditions and associated biological community responses.

Actual results of the IBI from four similarly sized streams and rivers were plotted together (Fig. 6) in an attempt to demonstrate the real application of these model concepts. These results demonstrate the utility of using the vertical scale of the IBI (or ICI, MIwb, etc.) to differentiate between different types of impacts. The left column lists the "gradient" of impact types associated with the vertical scale of the IBI with the actual impacts present in each of the four streams being listed in the right column. Riverine biological communities may experience spatially different impacts on a longitudinal upstream/downstream basis with the degree of departure and recovery dependent on the severity and type(s) of impacts being exerted on the biota. Walnut Creek and the Hocking River are examples (Fig. 6). Other streams are relatively unimpacted or have only moderate departures (Big Darby Creek) while others may be uniformly devastated (Rush Creek). These examples correspond to the narrative descriptions of community response and the attributes of the various impact types in Figure 5.

The point here is that the biota itself integrates differing types and degrees of environmental impacts on a spatial and temporal basis, providing feedback that is more accurate than can be achieved using cultural, surrogate, or process characterizations alone. Also, insight can be gained on what to expect as the predominant impacts in a particular segment change over time as a result of decreasing or increasing pollution levels. For example, a predominantly toxic impact should be expected to change to a conventional impact when the sources of toxicity are controlled. This may be evident on both a temporal and spatial scale. Often times impacts are "layered" in rivers, with the less severe impact types being masked by those that presently result in more severe degradation. As the more severe problems are reduced or eliminated the "lesser" problems may become evident in the results. An example of this is being observed in Ohio where the abatement of municipal and industrial point source problems is revealing nonpoint source impacts.

Summary

Definite patterns in biological community data exist and can be used in determining whether or not a water body is attaining its designated use and, if not, identifying the predominant causes of impairment. The Ohio Environmental Protection Agency has used this approach in producing the biennial Clean Water Act 305b report required by EPA, specifically the assignment of causes and sources of aquatic life use impairment (Ohio Envi-

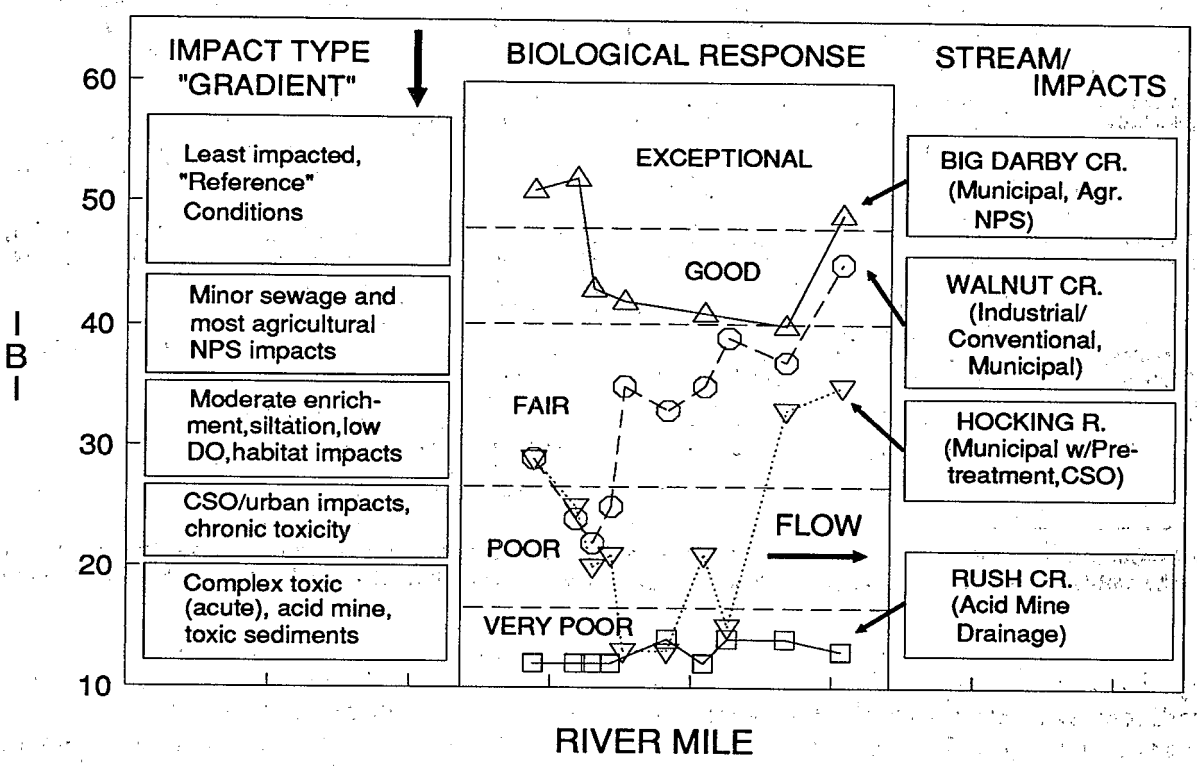


Figure 6.—Biological community response as portrayed by the Index of Biotic Integrity (IBI) in four similarly sized Ohio rivers with different types of point and nonpoint source impacts.

ron. Prot. Agency, 1990a). Other uses include supporting enforcement and litigation proceedings. For example, this type of assessment has been used to refute positions taken by the National Point Discharge Elimination System permit holders that the degradation measured was due to poor habitat or factors unrelated to their discharge. The biological response signatures can be particularly useful in demonstrating that the degradation is related to specific discharges, especially those involving the Complex Toxic impact type. While the legal requirements of the Clean Water Act may be viewed as sufficient to require entities to reduce pollutant loadings, the system of challenging these mandates requires the regulatory agency to defend the reasonableness of its regulatory actions. This type of ambient response data is particularly valuable in meeting that need.

Minimum requirements for using this type of data include having sufficient information to employ the use of multimetric evaluation mechanisms, a standardized approach to data collection, and consistent and responsive management of the database. Other factors that further increase the analytical power of the biological data include the use of multiple organism groups, an integrated approach to conducting ambient surface water assessments, and the inclusion of ancillary data such as biomass for fish.

It is important to make these and other data collection decisions early in the process. An example of the importance of these early decisions was the Ohio Environmental Protection Agency's decision to record external anomalies on fish, which 10 years later allowed the development of the %DELT metric of the IBI. This metric proved key in identifying the Complex Toxic impact type. At the time it was not known that this use would be possible; however, failure to include it as a quantitative measurement early in the process would have resulted in an unfortunate and irreplaceable loss of data. Thus, the ability to utilize biological data for diagnosis in Ohio was partly the result of decisions made more than 10 years ago, not only regarding which organism groups to sample, but about the types of information that should be recorded from each sampling effort. Frequently, biological monitoring programs are pressured to sacrifice data quantity and quality to meet regulatory and financial constraints. As seen here, such decisions can have far-reaching consequences over the long term.

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POSTERS

Using Machine Learning Techniques to Visualize and Refine Criteria for Biological Integrity

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ABSTRACT

Several techniques from the fields of artificial intelligence/machine learning and statistics were used to examine and analyze biological criteria (biocriteria) data. The goals of this research were: (1) to analyze biocriteria data, discovering patterns and relationships to aid water quality scientists in assessing environmental integrity of surface water sites; and (2) to develop, compare, and contrast computer techniques for aiding in the visualization and analysis of large databases. Specific classification techniques used were CART, ID3, and a Genetic Algorithm. An interactive data exploration tool was built based on ID3. The research produced several results: (1) Complex industrial sites can be identified with an 82 percent accuracy; (2) other sites are strongly corrupted by secondary impact, yielding an average accuracy of between 60 and 65 percent; (3) biological criteria have comparable accuracy to chemical, while combining both biological and chemical indicators produces slightly better results; (4) raw fish scores classify as well as composite indices; (5) ID3 is a valuable tool for identifying important attributes in a high dimensional attribute space; and (6) an attribute, "all intolerant species," presently used only for headwaters by the Ohio EPA, was identified for potential broad use.

Data

The biocriteria data used in this research are equivalent to 461 records and consist of measurements taken at water sites in Ohio. Each record holds the values of 495 attributes, including site information, biological and chemical measurements, and compound variables. Records include the primary and secondary pollution impact on that site (e.g., agricultural runoff) as determined by Ohio EPA water quality scientists.

The data were merged from several database sources into one file consisting of physical, biological (individual fish and insect species data), and chemical attributes, totaling 495 in all. These data were drawn primarily from one ecoregion of Ohio containing medium and large streams, and include reference sites. Each data record was classified by its

primary and secondary impact class as determined by Ohio EPA water quality scientists into eight categories. These are (1) Complex Municipal/Industrial; (2) Conventional Municipal/Industrial; (3) Combined Sewer Overflows; (4) Channelization; (5) Agricultural Nonpoint; (6) Impoundments; (7) Combined Sewer Overflows with Toxics; and (8) Other.

The data also include combined or derived attributes according to a tiered methodology. Fish and insect species are grouped together into subclasses by taxonomic and other means such as habitat. These subclasses are then used to build a scoring and normalizing system similar to the indicators used to measure the health of the economy. The Ohio EPA uses three: IBI (Index of Biological Integrity), IWB (Index of Well Being), and ICI (Invertebrate Community Index). IWB is based on structural attributes of the fish community, whereas the IBI also incorporates functional characteristics. ICI is based on the insect community. All of the derived attributes used to build these indices were also part of the data.

The data are very typical of real data sets. Characteristics include or imply:

- Noise, in terms of both measurement and impact type identification error;
- Unknown, missing, or by-case not-relevant data fields;
- A rich attribute structure (species taxonomy, tiered attributes);
- Distribution in several interlinked database files; and
- Multiple questions requiring more research.

Several questions were investigated with these biocriteria data:

- Can methods be developed for automating the determination of impact type(s) of a new site based on chemical, biological, and combined attributes?
- What attributes are more and less relevant when determining the impact type of a new site?
- Does the use of biological (or chemical) measurements alone provide superior classification results to that obtained using just chemical (or biological) data?
- How can chemical, biological, and combined attributes help discriminate the primary impact types and both the primary and secondary impact types taken in combination?
- Can single-species counts be used as a discriminator of impact type?
- Can the Index of Biological Integrity be used to generate a measure of impact severity? If so, can this measure of impact severity discriminate as part of the class label?
- Can the utility of some of the combined attributes for discriminating impact type be improved?
- Does an initial partitioning of the data, such as the separation of medium from large streams, help the discrimination task?

Technical Approach

Classification

The initial goal of this research was to correlate the human determination of impact type with the measured attributes so that future determinations would be automated. The data were analyzed with four classification algorithms.

For each technique the general methodology was to withhold a certain percentage of the data and train each specific algorithm with the remainder. By then classifying the withheld cases using each algorithm and matching the automated determination against the prior human determination, the effectiveness of each approach can be measured. In addition, running the systems on the biological and chemical attributes separately made it possible to compare the utility of each approach.

Tree Classifers: CART and ID3

ID3 is an algorithm for inductively synthesizing a binary decision tree for classification given a set of labeled training examples in the form of feature vectors (Quinlan, 1983; Pao, 1989). As in the game of "20 questions," the object is to find as few questions as possible that will correctly classify the data. Thus, ID3 determines the binary question that provides the most information about the identity of the data. Each question divides the dataset, S, into two groups, S_t and S_f, depending on whether the answer to the question is true or false for a particular datum. ID3 is then applied recursively to each group until the data cannot be partitioned further.

At each stage, ID3 asks the question that maximizes the information (most reduces the uncertainty) about the class membership of the data. The entropy, or uncertainty, existing before the question is asked is:

$$H(S) = -\sum_i P_i \log_2(P_i)$$

where P_i is the fraction of the elements in S belonging to class C_i.

After the binary question Q is applied, the data are divided into two groups, S_t and S_f, and the remaining entropy is:

$$H(S,Q) = P(S_t)H(S_t) + P(S_f)H(S_f)$$

where $P(S_t)$is the fraction of the elements of S for which the question, Q, is true. Similarly for $P(S_f)$.

The information gained by asking Q is then

$$I(Q) = H(S) - H(S,Q)$$

and the best question to ask is the one that maximizes I(Q).

When the training data have binary valued features, the feature, F, for which $H(S,F)$ is maximum, is chosen. When the values of the data features are continuous, as they are here, a feature, F, and a threshold, T, for which $H(S,F>T)$ is maximized, must be determined. For a given feature, the best threshold can be found by a linear search.

ID3 is problematic in that it overfits the data. C4, and CART (Breiman et al. 1984; Crawford, 1989) try to solve this problem by trimming tree limbs when they don't improve some heuristic measure of the goodness of the tree. MDL was used to prune the initial ID3-generated trees. The utility of CART with some of the initial data obtained from the Ohio EPA was also explored.

Genetic Algorithm/K Nearest Neighbors

Genetic algorithms are a learning paradigm based loosely on the model of biological evolution and first described in Holland (1975). Briefly, a population set of solutions to the problem is initialized and a reproduction/evaluation cycle is initiated. Reproduction includes operations such as crossover and random mutation of solutions. Reproduction rates for each member are determined by the evaluation function, which provides a measure of the ability to solve the problem at hand. Genetic algorithms have been applied successfully toward many tasks, including network layout and semiconductor design.

The nearest neighbors clustering algorithm is a well-known technique for categorization based on the distances between data points. In this study the Euclidean distance metric was used. A new classification algorithm was developed in which the genetic algorithm learns real valued weights for each attribute. By assuming higher weights to the attributes more attributes relevant for classification measures are adjusted so that like data points are determined to be in close proximity to each other. Assuming the weights have two significant digits, an exhaustive search would entail the examination of $100^{\text{number of attributes}}$ weight combinations, necessitating enormous computing capabilities. The genetic algorithm approach allows all areas in the space of possible solutions to be explored, albeit incompletely.

Neural Networks

Several machine-learning techniques can be used in conjunction with one another. For example, while neural networks are quite powerful, they can require considerable computer time to develop. Tree classifiers such as ID3 and CART are often used initially because they produce classifiers quickly. By extending the ID3 approach to use linear combinations of features, a neural net-like classifier can be quickly produced. Cluster analysis can also be used to develop gaussian features that provide a hidden layer of a neural network, with or without supervised training data. Neural networks (backpropagation and cascade correlation training) were applied initially to the EPA data, but the number of attributes and training set size led to training times that were much too long for initial exploration of the data.

Data Visualization

Any machine learning technique can be misapplied when treated as a black box. Displaying and exploring the data are important at each point in the analysis. Even classifiers, such as a decision tree, are quite useful as a data browsing technique in their own right. The classifier can be thought of as an apprentice to the domain expert; together they iteratively refine their understanding of the situation under investigation. Thus, the rules produced by a classifier are a compact description of the data;

in the process of understanding these rules, the expert may be led to refine the classification problem.

The ID3 tree classifier was used for interactive data exploration (see Fig. 1). A system was built that graphically presents the results of ID3, using the hypertext paradigm to probe nodes in the tree for information not currently displayed (e.g., number of examples covered by this node, ID numbers of the examples, attribute value for each example, etc.).

A graphic display for presenting the Genetic Algorithm/K Nearest Neighbors results was also constructed. Figure 2 displays the sets of weights learned during five training runs of the algorithm to discriminate agricultural nonpoint impacts (impact class 5) from all others. This display allows a person to quickly view results and discover patterns. The results in Figure 2 suggest that the number of darter species (row 1) is more important for the identification of agricultural nonpoint pollution than the number of intolerant species (row 4), as this species received a larger weight during each training run.

Results

Classification

These biological techniques showed that biological indicators were more sensitive than chemical measures in determining impact type of the data withheld for test. However, as a much greater percentage of chemical data had missing values, further experiments would be required to prove that biological analysis is in fact better than chemical. Second, no technique had an accuracy rate greater than 75 percent in the classification of withheld sites. The Complex Municipal/Industrial category had much better classification performance than the other categories.

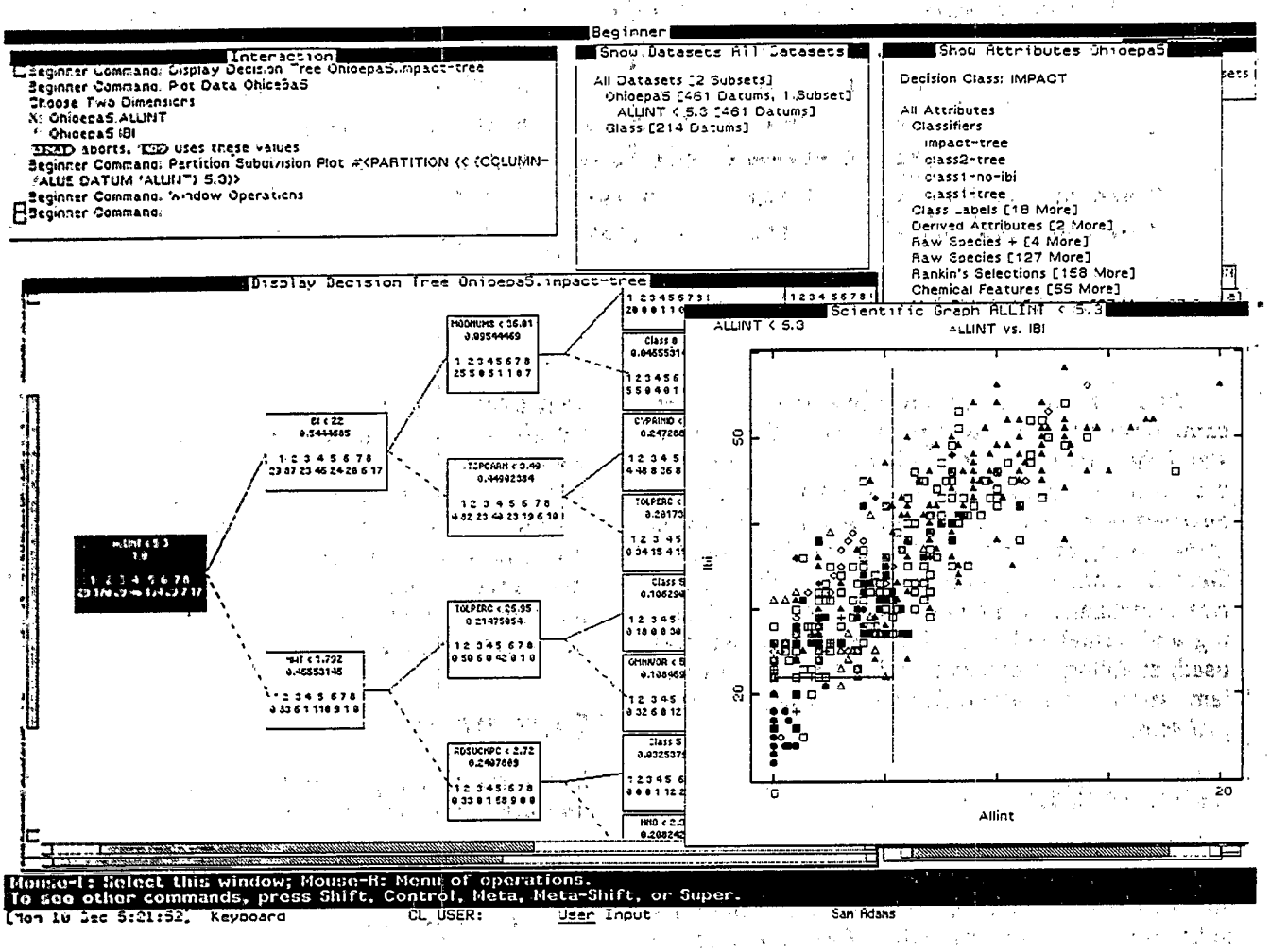

Figure 1—A screen image of the Data Exploration Interface, Beginner. Note: Beginner incorporates an ID3 tree classifier as a way of doing exploratory data analysis.

Training Mode: Single Class NM; Class: 5

	(1 2 3 4)	(1 2 3 5)	(1 2 4 5)	(1 3 4 5)	(2 3 4 5)
darter species	0.84	1.00	0.70	0.67	0.81
sunfish species	0.71	0.66	0.71	0.38	0.85
sucker species	0.40	0.78	0.33	0.28	0.21
intolerant species	0.37	0.18	0.34	0.46	0.37
top carnivores	0.85	0.53	0.73	1.00	0.82
omnivores	0.23	0.41	1.00	0.29	0.55
round bodied suckers	0.54	0.56	0.15	0.22	0.43
deformities	0.16	0.58	0.94	0.16	0.00
species minus exotics	0.34	0.22	0.92	0.32	0.28
sensitive species	1.00	0.95	0.98	0.71	0.62
sculpin & darter species	0.31	0.99	0.71	0.65	0.84
minnow species	0.73	0.80	0.70	0.68	0.17
insectivores	0.35	0.46	0.63	0.29	0.53
headwater species	0.03	0.19	0.94	0.78	0.20
pioneering species	0.47	0.49	0.28	0.32	0.57
% lith. spawners	0.19	0.27	0.13	0.04	0.40
# lith. spawners	0.56	0.03	0.05	0.77	0.06
K1	0.56	0.28	0.76	0.84	0.52
K2	0.94	0.63	0.64	0.46	0.82
K3	0.95	0.68	0.36	0.78	0.69
Training Error Rate	0.199	0.191	0.214	0.176	0.219
Testing Error Rate	0.304	0.348	0.344	0.269	0.355

Figure 2—Genetic Algorithm/K nearest neighbor results.

In the process of running this analysis, it became obvious that it was necessary to first understand the data more thoroughly, especially where it presented noisy, missing, or invalid values. It was also necessary to identify subproblems of predicting impact using biocriteria and chemical data from the Ohio EPA dataset. To accomplish this, the interactive visualization capabilities of our machine learning and visualization environment, Beginner, was used, enabling interactive exploration of the problem space and identification of tractable subproblems.

Visualization

By using the visualization tool, the domain expert was able to discover previously unknown relationships among the data. For example, preliminary results indicate that the Ohio EPA measure of intolerant species, INTOLS, would be more discriminating if it were broadened to include moderately tolerant species — as was done for another attribute, ALLINT. ALLINT, which was used in headwater cases by the Ohio EPA, was more useful than INTOLS as a discriminator of impact type. Complex Municipal/Industrial sites can be easily identified. Combined Sewer Overflow sites are difficult to separate from Agricultural Nonpoint sites using biological criteria alone, but relative degree of impact can be identified.

Summary

In summary, this project generated several results important in exploring complex databases:

- Differences in setup, run time, and results of machine learning algorithms were compared and contrasted.
- Many issues associated with large databases were explored. For example, the handling of

missing values and data records that can be classified into multiple categories was investigated.

- A data exploration/visualization tool that successfully developed, confirmed, and disproved hypotheses in a time- effective manner was constructed.

This project also generated important results in the use of criteria for biological integrity:

- an attribute currently being used in evaluating headwaters, ALLINT, is a useful discriminating attribute for all rivers;
- individual fish species that are ubiquitous in Ohio rivers can serve as indicators of biological integrity; and
- the presence of some fish species helps in determining a river's change in biological integrity. For example, low values of the tiered attribute, HEADWTR (headwater species), was used in discriminating large moderately impacted municipal/industrial-class streams, possibly indicating recovery.

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Poeciliopsis: A Fish Model for Evaluating Genetically Variable Responses to Environmental Hazards

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Recent studies of responses to cellular stress (such as the heat shock response) combined with earlier studies of inducible detoxification systems (such as the cytochromes P450) have shown that organisms in general have sensitive genetic systems for monitoring environmental stressors. These methods for measuring stress use an organism's own stress response induction pathways as onboard biosensors and induction products in the form of stress mRNA and proteins as indicators of environmental stress.

This new approach proposes a new indicator, cellular stress responses, as early warning systems to signal the need for remedial action before severe ecosystem disturbances result in loss of species. The practice of relying on massive fishkills as indicators of environmental problems does not take into account the numbers of weakened survivors that later quietly die of secondary infectious diseases or cancer. As environmentalists become more prevention-oriented, the monitoring of stress in feral animals, as well as establishing species-specific risk levels, may become the preferred goals. Several applications of this approach to environmental problems are presented.

One example relates to the recent concern that over the next decade global warming of roughly 2°C will be experienced. It is thought that this warming could be progressive if remedial action is not taken over the next decade. Climatic shifts of this sort contain the ingredients of mass extinctions. The effects depend on the degree of genetic variation available in populations. This experiment used six species of the Sonoran topminnow, *Poeciliopsis* and eight of its all-female hybrid clones as models to evaluate genetic deployment of resistance to heat stress in natural populations. It was established that thermal resistance can be conferred by preheating the fish to just below killing temperatures (37-38°C) for one hour, after which survival at the normal killing temperature (39-41°C, depending on the biotype) is considerably enhanced. When liver tissues from preheated fish are examined, using two-dimensional polyacrylamide gel electrophoresis, they contain stress proteins that are not present in fish at normal temperatures.

In addition, there is extensive biochemical diversity in the isoforms of two major families of heat shock proteins (hsp70 and hsp30 families), suggesting that genetic variation in these proteins may contribute to differences in thermal resistance among the *Poeciliopsis* biotypes. The hsp70 and hsp30 proteins are thought to repair thermal damage and to protect cells from lethal damage, respectively. Recent studies have confirmed earlier hypotheses that the heat stock response is keyed to protein damage and appears directed toward restoring protein homeostasis in cells subjected to a variety of stressors in addition to heat, including heavy metal ions, arsenicals, amino acid analogues, and tissue trauma.

Differences in susceptibility to chemically induced hepatocarcinogenesis, both in tumor incidence and in tumor type, are found within species as well as between species of *Poeciliopsis*. This diversity of response among genotypes maintained in the laboratory aquarium facility allows examination of physical, chemical, and genetic factors that may contribute to differences in tumor induction among genotypes, The assemblage of biotypes in the colony includes inbred and outcrossed stocks, as well as unique all-female species that reproduce clonally, thus allowing multiple replicates of wild genotypes to be held constant while the environment is manipulated. These fish are being used to examine

whether carcinogens become effective at lower-than-threshold doses when hepatocyte proliferation is initiated with independent stimuli such as heat stress and chemical toxicants. Using dimethylbenz[a]anthracene (DMBA) as a toxicant in cell proliferation studies, it has been established that the number of days after treatment to maximum levels of mitosis (2-12 days) is highly predictable and is influenced by the time of exposure (10-22 hours) to a toxic concentration (5ppm) of DMBA.

Exposing *Poeciliopsis* to sublethal temperatures for 30 to 60 minutes results in the death of embryos in pregnant females and in liver cell damage to adults. Hepatocyte proliferation is thus stimulated, which peaks 2 to 3 days after the imposition of heat stress. Subsequent studies will determine if prior initiation of cell proliferation will enable tumors to be induced at lower concentrations than in fish that have not been exposed to heat. Since fish seeking food enter water that is hot enough to risk their lives, presumably it is hot enough to cause cell damage and initiate unscheduled cell proliferation.

Many compounds do not become toxicants or carcinogens until they are metabolically activated by an oxidative enzyme, a cytochrome P450. Using the *Poeciliopsis* hepatoma cell line, it was demonstrated that cytochrome P450 activity can be induced in cell culture. It has thus been possible to carry out dose-response studies of hepatotoxicity and modulation of P-450 inducers and inhibitors (benzoflavone) on the effects of DMBA and benzo[a]pyrene (BaP). Preliminary studies suggest that the toxic levels of DMBA and BaP for cells in culture are generally comparable to those for live fish.

Working with nitrosodiethylamine (NDEA), it has been determined that among *Poeciliopsis* genotypes NDEA deethylase activity (liver microsomal cytochrome P450pj) varies both in maximal activity and in optimal temperature. Metabolic responses after exposure to different concentrations of NDEA will now be compared over a range of temperatures, examining production of phase 1 metabolites and the formation of DNA adducts.

This system enhances the understanding of fish as monitors of domestic water supplies; it also provides a means to assess the variation in response to chemical and thermal stress that is stored in the gene pools of wild populations. Such variation may play a major role in how manmade forces of selection will shape future populations.

ACKNOWLEDGMENTS: Members of the Marine/Freshwater Biomedical Sciences Center who have contributed to this research are: J.F. Crivello, L.A.E. Kaplan, P.J. diIorio, M.E. Schultz, J.J. Stegeman, and C.N. White.

Assessing Biological Integrity Using EPA Rapid Bioassessment Protocol II — *The Maryland Experience*

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The Maryland Department of the Environment, Water Quality Monitoring Division, has begun using the EPA Rapid Bioassessment Protocol II as part of a statewide water quality monitoring network, and in selected special studies. The Protocol II, with 100+ organism subsamples identified to family level, was considered an efficient method for obtaining quality data from a large number of streams.

Over 200 rapid assessment samples were completed during the 1990 field season. The streams sampled were located in a number of different ecoregions. The results from two ecoregions, the Potomac drainage from the Allegany Plateau and the coastal watersheds of the Choptank and Chester Rivers, were chosen to illustrate our experience (the Maryland experience).

Stations were chosen from 1/62,500-scale county maps. The most downstream third-order reach with a road crossing was the first choice. If this location was inaccessible or lacked proper habitat, the next closest road crossing was used. All but minor impacts to the upper watershed of a stream were assured to be reflected at the third-order sampling location.

The sampling methods were based on the Rapid Bioassessment Protocol II described by U.S. EPA (Plafkin et al. 1989). A 1 m^2 kick seine sample was collected from the best available habitat, and a random 100+ organism subsample obtained. The subsample was identified to the family level in the field, and the sample was archived for future reference. No Course Particulate Organic Matter samples were collected because of the varied availability of this substrate. The habitat was assessed at each station with habitat characteristics customized for each ecoregion.

Reference streams were chosen from each ecoregion for both biological and habitat indices. The choice of a reference stream was an intuitive decision taking into consideration all biotic, habitat, and water quality factors. The biological reference stream was not necessarily the habitat reference stream.

The information from the family-level identification was processed through the various metrics described for Protocol II and a biological score was obtained. The biological and habitat scores were then taken as a percentage of their respective references. The results are plotted in Figures 1 and 2.

The divisions of unimpaired, moderately impaired, supporting, etc., are based on the characterizations of poor, fair/good, and excellent that are applied to the raw biological and habitat scores. Streams that fell in the lower right-hand portion of the graph tended to have water quality problems such as acid mine drainage, STP effluents, and agricultural runoff. The macroinvertebrate community was severely impacted.

For purposes of initial characterization and monitoring, those streams that fell in the severely impaired classification were candidates for a more intensive study to better define the source of the impact. This study would use the Protocol III or other quantitative methods.

The indices used in the assessment calculations were described by Plafkin et al. (1989). Some of these, particularly the ratios, did not correlate well with the others except in cases of impacts. In an effort to overcome this shortcoming, a number of alternate indices are being considered as substitutes, such as Chironomidae/Total Diptera, Invertebrates/Total Trichoptera, and Non-insect Invertebrates/Total Sample. If these or other alternates

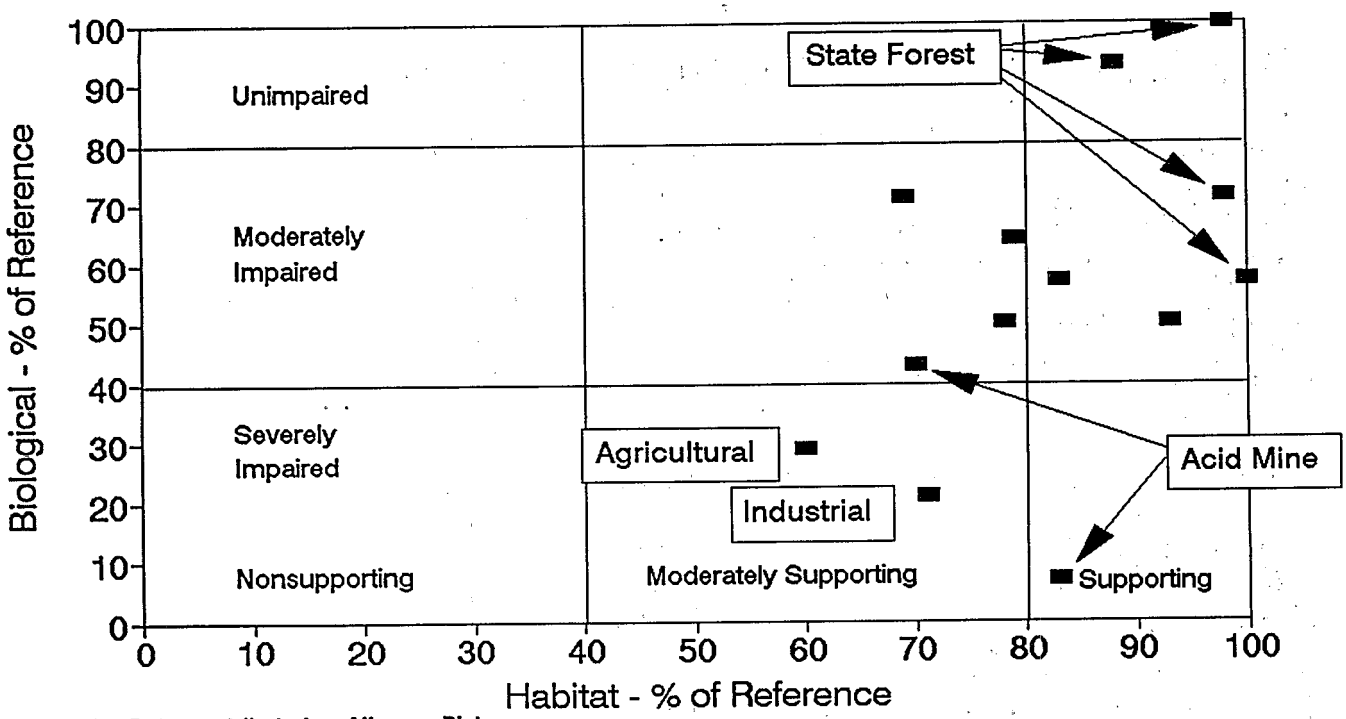

Figure 1.—Potomac tributaries–Allegany Plateau.

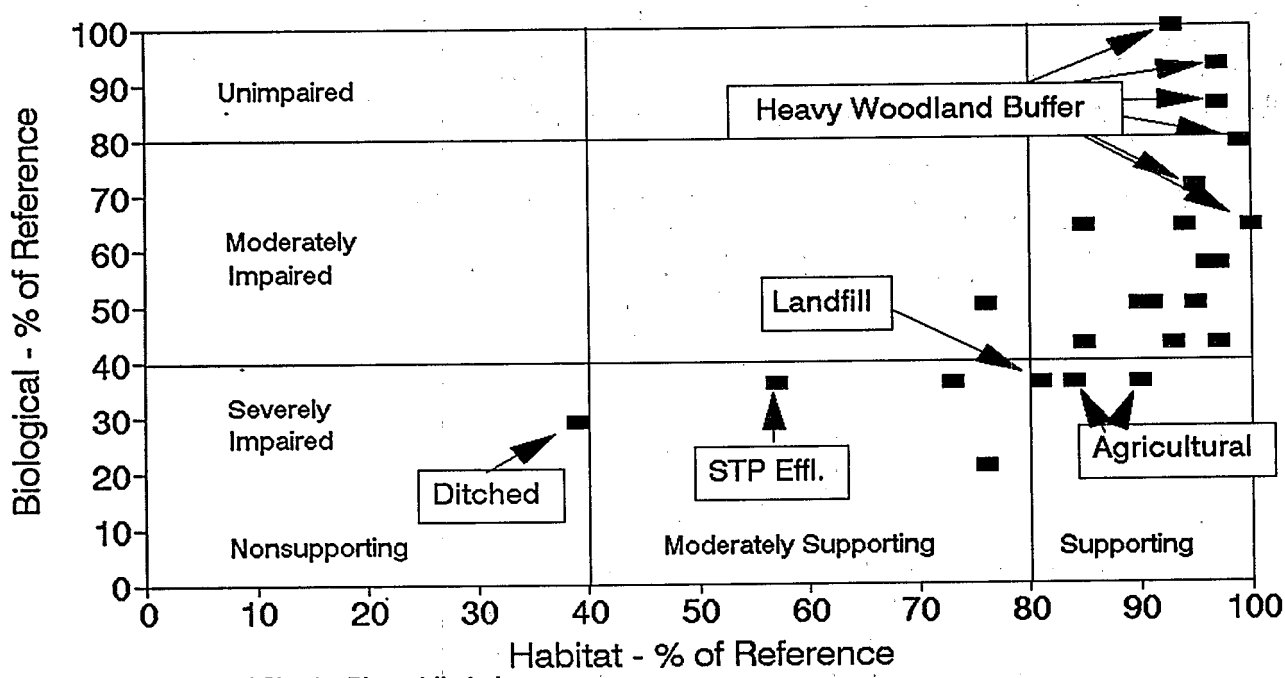

Figure 2.—Choptank and Chester Rivers tributaries.

provide better correlation and information about the sample, they will be included in all past and future assessment calculations.

We feel that the U.S. EPA Rapid Bioassessment Protocol II is an effective tool for initial characterization and monitoring of streams in Maryland. It has allowed broader coverage of state waters with minimal additional time and expense, and has contributed toward the U.S. EPA goal of "fishable/swimmable."

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The Use of the Qualitative Habitat Evaluation Index for Use Attainability Studies in Streams and Rivers in Ohio

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The Qualitative Habitat Evaluation Index (QHEI) has been developed to help distinguish the influence of habitat effects on fish communities in Ohio streams. The index is a composite of six habitat variables: substrate, instream cover, riparian characteristics, channel characteristics, pool and riffle quality, and gradient and drainage area. The index relies on visual estimates of several characteristics of each habitat variable and can be completed in less than an hour for a 200-500 meter stream segment. Components of each variable have been assigned scores based on observed or predicted relationships with fish species diversity and/or measures of community integrity. The QHEI was significantly correlated with the Index of Biotic Integrity in Ohio streams and rivers, however the nature of the relationship varied by ecoregion. Ecoregion-level, reach-level, and subbasin-level habitat quality factors appear to act a "covariates" that likely limit the site-specific predictability of any habitat indices that fail to consider them. The use of the QHEI for use attainability analyses includes the compilation QHEI subcomponents by the aquatic life use they are most strongly associated with (modified warmwater, warmwater, and exceptional warmwater habitat) and the ratios of these subcomponents to one another in a given stream reach.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

The Use of the Amphipod *Leptocheirus Plumulosus* to Determine Sediment Toxicity in Chesapeake Bay: Development and Field Applications

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Ideal species for testing the toxicity of estuarine sediments exhibit sensitivity to sediment contaminants and are physiologically adapted to extreme variability in salinity and sediment type. We propose the use of the amphipod *Leptocheirus plumulosus* to test the toxicity of sediments in Chesapeake Bay and other east coast estuaries. This species is an ecologically important inhabitant of both oligohaline and mesohaline sections of Chesapeake Bay, and is found infaunally in sediments ranging from fine sand to very fine mud. No significant differences were observed in amphipod survival among four salinity treatments (5, 15, 20, and 32 ppt). Additionally, no significant differences were observed in amphipod survival among four salinity treatments varying in particle size and organic content. Acute 96-hour LC-50 values for aqueous cadmium at a salinity of 6 ppt were 0.26 mg cd/L and 0.19 mg cd/L for *L. plumulosus* and *Hyalella aztesa*, a common freshwater sediment test organism, respectively. A field survey was conducted in which *Leptocheirus plumulosus* were exposed to sediments from a variety of sites within Chesapeake Bay. The sites ranged from highly industrialized harbors to embayments containing commercial and community marinas. Ambiguity between qualitative benthic analysis and amphipod survivorship at a portion of the test sites highlight the need to implement toxicity tests utilizing sublethal endpoints. Laboratory experiments conducted to this end indicate that significant growth of juvenile *L. plumulosus* occurs under laboratory conditions, and that morphological features allowing for the differentiation of male and female amphipods appear after 20 days.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Compliance Monitoring of the Aquatic Biota in Vermont

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ABSTRACT

In 1986, Vermont Water Quality Statutes were amended to require all land-based wastewater disposal systems with a capacity of greater than 6,500 gallons per day, including spray irrigation and community sub-surface systems, to obtain an Indirect Discharge Permit (IDR) from the Department of Environmental Conservation (DEC). This statutory amendment established a narrative compliance criterion of "**no significant alteration of the aquatic biota**" in surface waters adjacent to such systems (Vt. Dep. Environ. Conserv. 1986). The DEC was charged with developing the rules and regulations that would implement this criterion (Vt. Dep. Environ. Conserv. 1990). The legislative intent of the regulation was primarily to protect fragile, high-elevation streams that were at risk from impacts that altered the basic biological and aesthetic character of surface waters adjacent to high-volume waste disposal systems installed to service recreational development. Under this interpretation, discharges that caused "benign" alterations to biological integrity (e.g., benign enrichment), but impaired other values and uses (e.g., proliferation of algal growth), would be considered significant under this criterion. The DEC was therefore required to develop numeric biological criteria that would evaluate alterations of the aquatic biota in terms of both biological integrity and impact to nonbiological values and uses.

The Vermont Program

A protocol document for making determinations of significant alteration in the context described in the Abstract was prepared (Vt. Dep. Environ. Conserv. 1987). The document addressed the following major considerations: (1) target monitoring community; (2) sampling strategy and site selection; (3) sampling and processing methods; and (4) data analysis and criteria development. For each of these factors, a series of goals and objectives was specified and methodologies that best met those goals and objectives were determined.

Overall, the Department of Environmental Conservation felt that aquatic macroinvertebrate sampling was the most cost-effective means of establishing a database from which compliance decisions could be made. Macroinvertebrates were chosen as the target community for a variety of reasons, including sensitivity to perturbation, ability to integrate impacts over time and across trophic levels, and the high informational content of macroinvertebrate samples. The existence of well-established sampling and analytical methods, and in-house data base and expertise were also important considerations.

A paired site (control/impact) sampling protocol with on-site controls was selected as the primary sampling strategy. The objective of site selection was to isolate the stream reach potentially impacted by the discharge to minimize the effect of non-discharge-related perturbations.

The sampling methods were intended to reduce variability within and between control and impact sites caused by sampling error and habitat heterogeneity. A data quality objective (DQO) was estab-

lished consisting of population abundance estimate with a percent standard error of less than 20 percent. Available data indicated that these goals could be consistently met or with the use of rock-filled basket artificial substrates at a replication level of 5. A minimum density of 300 animals per replicate would be required to meet the DQO of 20 percent. Sample processing following standard preservation and in-lab sorting under magnification procedures were specified, with special protocols for subsampling if necessary. The protocol specified taxonomy to the lowest possible level using standard reference keys for individual orders of animals. Sample archiving and reference collections were also required.

Because compliance monitoring was to be conducted by the permittee or its agent, a relatively high level of DEC oversight would be necessary to ensure high quality data. Prior to initiating monitoring, the permittee would be required to submit a detailed QA/QC (quality assurance/quality control) plan to the Department of Environmental Conservation for approval. In addition, the Department personnel were to conduct joint site visits and evaluations, process split samples, examine reference collections, review all data submitted, and generally maintain close contact with all agents generating data.

A great variety of metrics can be used to describe the functional and structural characteristics of macroinvertebrate communities. For the purposes of determining significant alterations, the Department of Environmental Conservation chose four metrics for comparing alterations between control and impact sites.

1. The **Pinkham-Pearson Coefficient of Similarity** was selected as a screening metric to make an initial evaluation of the degree of community structure similarity between control and impact sites.

2. A modification of the **Hilsenhoff Biotic Index** was selected as an evaluation metric primarily because of its sensitivity to alterations in nutrient dynamics, the major anticipated impact.

3. **Ephemeroptera (mayfly), Plecoptera (stonefly), Trichoptera (caddisfly) taxa richness (EPT)** was selected as an evaluation metric because of the anticipated prevalence of these orders in target streams and their sensitivity as an indicator of high quality diversity.

4. Finally, **relative abundance** was selected as an evaluation metric because of the general sensitivity of abundance to toxics, trophic alterations, and overall habitat alterations.

The relatively extensive macroinvertebrate database maintained by the Department of Environmental Conservation was evaluated by Department staff to determine the amount of change in the selected metrics that would be indicative of significant alteration. With the exception of relative abundance, these change criteria were determined independently of statistics and were selected to represent alterations of biological significance. In the case of relative abundance, the biological and statistical significances ($p < .05$, Mann-Whitney U-Test) were thought to be equivalent. Exceeding the allowable change criterion in any one metric would result in a determination of significant alteration. Because of excessive data variability (failure to meet DQOs), the change criteria could be exceeded without producing a statistically significant change. Therefore, a confirmation of statistical significance would be required ($p < .05$, Mann-Whitney U-Test) prior to a significant alteration determination.

Results

To date, more than 25 Indirect Discharge Permits requiring biological monitoring have been issued by the Department of Environmental Conservation. Monitoring frequency is determined primarily by the size of the system and ranges from twice per year (winter and late summer) to once during the five-year life of the permit. The regulated community has been very responsive and cooperative in implementing compliance monitoring. The Department of Environmental Conservation and monitoring personnel have maintained close communi- cations and have worked together to resolve problems as they arose. Communication is facilitated by the small size of the state of Vermont and the relatively small number of consultants involved in monitoring activities.

In general, performance expectations have been met. Macroinvertebrates have proven to be an excellent monitoring community providing data adequate for making informed decisions. Consulting biologists have demonstrated considerable expertise in conducting monitoring activities and processing and analyzing samples.

At the same time, sampling strategy and sampling methods have not always attained the standards set by the regulations. In some cases, appropriate paired sites on the same stream are not

available because of physical limitations or potential degradation of control sites by nonregulated perturbations. Attempts to locate control sites in adjacent watersheds have met with mixed success. By far the greatest problem has been the physical displacement of rock baskets during exposure because of extreme hydrologic events. This problem has been minimized by close observation of weather conditions and substrates during exposure. Another persistent problem has been low productivity of receiving waters, resulting in less than the minimum number of organisms per replicate required to meet DQOs. As a consequence, the data have not been adequate for making regulatory decisions in some cases. Selection of criteria indicating significant alteration of the aquatic biota has proven to be appropriate in most cases, although a review to evaluate the need for modifications is underway.

The majority of sites being monitored have demonstrated compliance with the criteria. In cases where alteration of biota has been found, enforcement response has ranged from increased monitoring intensity to major modifications to treatment systems.

The use of numeric biological criteria for regulatory purposes is a potentially contentious process. It is critical that final decisions regarding noncompliance and subsequent enforcement response not be executed in a technical vacuum, but rather with full consideration for interpretations of the monitoring data using the best professional judgment of both regulating and consulting biologists.

In addition to providing compliance information, biological data collected through the Vermont program have provided additional benefits. General knowledge of stream ecology and the response of stream biota to low levels of pollution has been greatly expanded. Chemical monitoring permit requirements provide data with which to evaluate dose/response observations in the receiving water. The general awareness of aquatic biota on the part of regulators and the regulated community has improved, to the ultimate benefit of pragmatic water quality management. In some cases, data generated through this program have detected water quality degradation in receiving streams caused by sources unrelated to the discharge being monitored, such as failed erosion control systems.

In summary, biological compliance monitoring in Vermont has resulted in the production of high-quality data describing actual in-stream impacts from indirect discharges. These data have been successfully used to make compliance decisions that are acceptable to both regulators and the regulated community. This program has demonstrated that biological monitoring, when applied in a program with clear goals and objectives, a high degree of QA/QC, numerical standards, and cooperation between regulators and the regulated community can be a valuable and extremely pragmatic water quality management tool.

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A Method for Rapid Bioassessment of Streams in New Jersey Using Benthic Macroinvertebrates

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A family level rapid bioassessment method (RBM) using benthic macroinvertebrate communities was developed and field tested for water quality evaluations in New Jersey streams. The method is a regional modification of EPA's family level rapid bioassessment protocol developed to screen and prioritize sites having impaired water quality. Depending on geographic location, macroinvertebrates are sampled from riffle areas or multiple instream habitats using kick net procedures. The RBM was applied to a set of approximately 200 sites over a two year period. The community analysis used to determine biological condition consist of five biometrics: 1) total taxa richness, 2) Ephemeroptera, Plecoptera and Trichoptera (EPT) richness, 3) percent dominance, 4) percent EPT and 5) modified biotic index. Biological criteria were established for three categories of water quality (non-impacted, moderately impacted and severely impacted), and the natural variability associated with individual biometrics was examined. Replicate sample comparisons made in several unimpacted reference streams did not result in assignment of differing water quality categories, suggesting that variability associated with individual biometrics was not sufficient enough to cause inaccurate water quality assessment. The rapid bioassessment appears to provide an acceptable approach to accurately screen for water quality impairment.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Development of Biological Impairment Criteria for Streams in New York State

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ABSTRACT

Biological criteria were recently developed for measuring significant water quality impairment in flowing waters of New York state (Bode et al. 1990). The criteria established are based on sampling benthic macroinvertebrate communities; they measure impairment as a quantitative change from conditions upstream of a given discharge. The sampling methods used are the traveling kick method for stream segments with wadeable riffles, and the multiple-plate artificial substrate sampler for stream segments without wadeable riffles. Replication in sampling is necessary to insure reliability of data. The parameters on which the criteria are based are biotic index, EPT value, species richness, species dominance, and percent model affinity. Because the criteria are directed toward enforcement rather than detection, they are numerical rather than narrative, and site-specific, rather than regional. Site-specific criteria have advantages over regional criteria in accounting for natural variability by comparing results to an upstream control site; this approach is also able to target the cause of impairment to specific discharges. Habitat comparability criteria were established to ensure high habitat similarity between the upstream and downstream sites. The parameters measured are current speed, substrate particle size, substrate embeddedness, and canopy. The proposed criteria were drawn from data sets collected from flowing waters in New York state over a 17-year period (1972-89). Preliminary criteria were based on changes between levels of an existing four-tiered classification of water quality used in New York state. The criteria were then tested over a 2-year period and modified as necessary. Issues tested include sensitivity and accuracy of the criteria, adequacy of the 2-minute/5-m kick sample, replicate variability, adequacy of the habitat criteria, and seasonal variability. Data from sites designated as having significant biological impairment were corroborated with available chemical data to confirm possible impairment.

Specifications of Biological Impairment Criteria

Sampling Methods

Two sampling methods are used, dependent on the availability of wadeable riffles. For streams with wadeable riffles in the desired reach, the traveling kick method is used, taking three 2-minute/5 m samples. One hundred organisms are subsampled from each sample. For streams without available wadeable riffles, multiple-plate artificial substrate samplers are used, with three 5-week exposure periods. Multiple-plate samplers have been used in New York state since 1972, with the modifications of using three hardboard plates, each 6 inches square, suspended 1 m below the water surface.

Index Levels

Significant biological impairment is indicated for kick samples when one or more of the levels in indices a–e is exceeded and the change is also shown to be statistically significant at the level of P = .05. Significant biological impairment is indicated for multiplate samples when one or more of the levels in indices a–d is exceeded and the change is also

shown to be statistically significant at the level of P = .05.

a. **Biotic index.** The biotic index is calculated by multiplying the number of individuals of each species by its assigned tolerance value, summing these products, and dividing by the total number of individuals. Tolerance values have been assigned on a scale of 0 to 10. The criterion for this parameter is + 1.5; an increase of 1. or more exceeds the allowable amount of change.

b. **EPT value.** The total number of species in the orders Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies) found in the sample or subsample. The criterion for this parameter is –4; a decrease of 4 or more exceeds the allowable amount of change.

c. **Species richness.** The total number of species found in the sample or subsample. The criterion for this parameter is –8; a decrease of 8 or more species exceeds the allowable amount of change.

d. **Species dominance.** This is the percent contribution of individuals of the most numerous species or taxon in the sample. The criterion for this parameter is + 15; an increase of 15 or more exceeds the allowable amount of change.

e. **Percent model affinity.** This new index is a measure of similarity to a model non-impacted community based on percent abundance in seven major groups (Novak and Bode, in prep.). Percentage similarity is used to measure similarity to a community of 40 percent Ephemeroptera, 5 percent Plecoptera, 10 percent Trichoptera, 10 percent Coleoptera, 20 percent Chironomidae, 5 percent Oligochaeta, and 10 percent Other. The criterion for this parameter is –20; a decrease of 20 or more exceeds the allowable amount of change.

Procedures for Application of Biological Impairment Criteria

1. Choose appropriate sampling method (kick sampling or multiplate sampling) by determining availability of wadeable riffles.

2. Select an upstream site and downstream site that meet the habitat criteria for site comparability for current speed, substrate particle size, substrate embeddedness, and canopy.

3. Conduct sampling at the upstream and downstream site using kick sampling in streams with wadeable riffles and multiplate sampling in all other streams. For kick sampling, four replicates are collected at each site; for multiplate sampling, three 5-week exposures are conducted.

4. Conduct laboratory sorting and identification of samples, using the level of taxonomy required for each group.

5. For kick samples, use percentage similarity to calculate similarity between three of the replicates at each site. If similarity if less than 50 for any replicate pairing, resubsample 100 organisms from the replicate with the lowest average similarity. If similarity is still less than 50 for the replicate pairing, subsample a fourth replicate from the site. If 50 percent similarity cannot be achieved with these replicates or subsamples, resampling is necessary.

6. Calculate parameters a–e for kick samples and parameters a–d for multiplate samples. Compute the average for the three samples from each site.
   - Biotic index
   - EPT value
   - Species richness
   - Species dominance
   - Percent model affinity

7. Compare values from the downstream site to those from upstream site (Fig. 1). For kick samples, violation of one or more criteria for parameters a–e indicates provisional impairment. For multiplate samples, violation of one or more criteria for parameters a–d indicates provisional impairment.
   - Biotic index: + 1.5 (0–10 scale)
   - EPT value: –4
   - Species richness: –8
   - Species dominance: + 15
   - Percent model affinity: –20

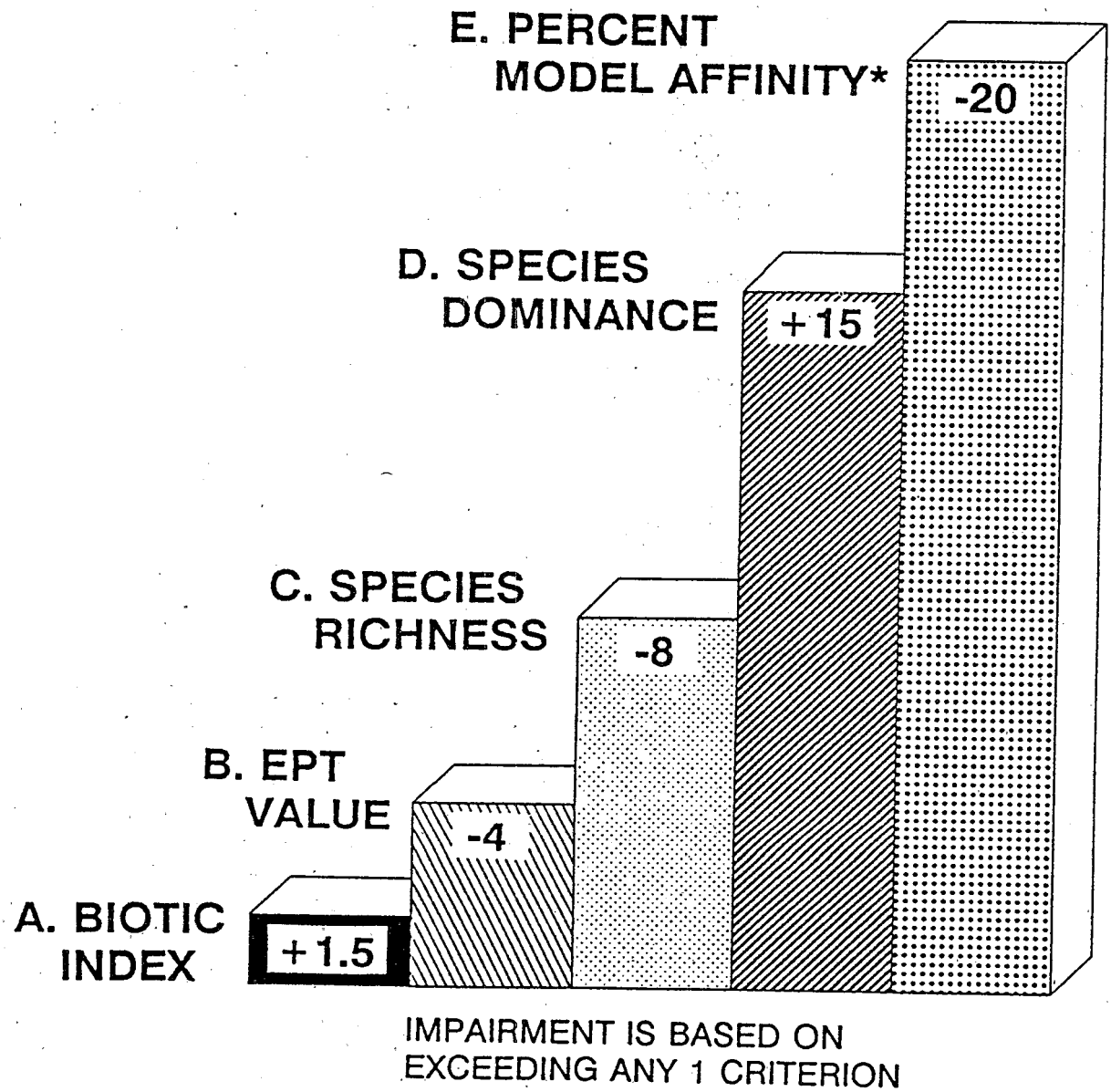

Figure 1.—Biological impairment criteria for flowing waters in New York state. *Percent model affinity is not used with multiplate samples.

8. For sites with provisional impairment, perform the Student's T-test to determine if results are statistically significant at the level P = .05. If results are significant, biological impairment is indicated.

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Development of Sediment Criteria for the Protection and Propagation of Salmonid Fishes

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ABSTRACT

Salmonid spawning and rearing are protected beneficial uses of waters in most western states. Nonpoint source activities causing accelerated sedimentation to streams can adversely affect salmonid growth and survival. Water quality criteria, proposed for inclusion in Idaho's water quality standards, have therefore been focusing on protection of developing embryos and young fish from the detrimental effects of sediment. The approach is supported in the literature and has been verified by field testing in Idaho.

Introduction

The feedback loop concept of nonpoint source pollution control has been incorporated into Idaho water quality standards (Ida. Dep. Health and Welfare, 1990). This concept requires development of in-stream criteria to protect the beneficial uses of the State's waters. Feedback from in-stream monitoring is compared to the in-stream criteria to determine whether or not best management practices (BMPs) applied to nonpoint source activities are effectively protecting the beneficial uses.

Fine sediment pollution that impairs habitat for rearing and reproducing salmonid fishes has been reported in 90 percent of all impacted stream segments in Idaho (Ida. Dep. Health and Welfare, 1988). This condition prompted water quality experts to initiate an extensive review of the literature covering sediment effects to fish. Based on this review (Chapman and McLeod, 1987) and the advice of 50 local and regional technical experts, sediment criteria were proposed for inclusion in the State water quality standards (Harvey, 1989).

One criterion is designed to protect incubating salmonid eggs from the detrimental effects of fine sediment on critical dissolved oxygen delivery through the substrate. Another prevents increases in sediment accumulation in cobble rearing spaces critical to over-winter survival of young salmonids.

Salmonid Embryo Survival in the Spawning Redd

Methods

Chapman and McLeod (1987) concluded that for incubating salmonid embryos, survival to emergence is inversely related to the proportion of fine sediment increases in the incubation environment.

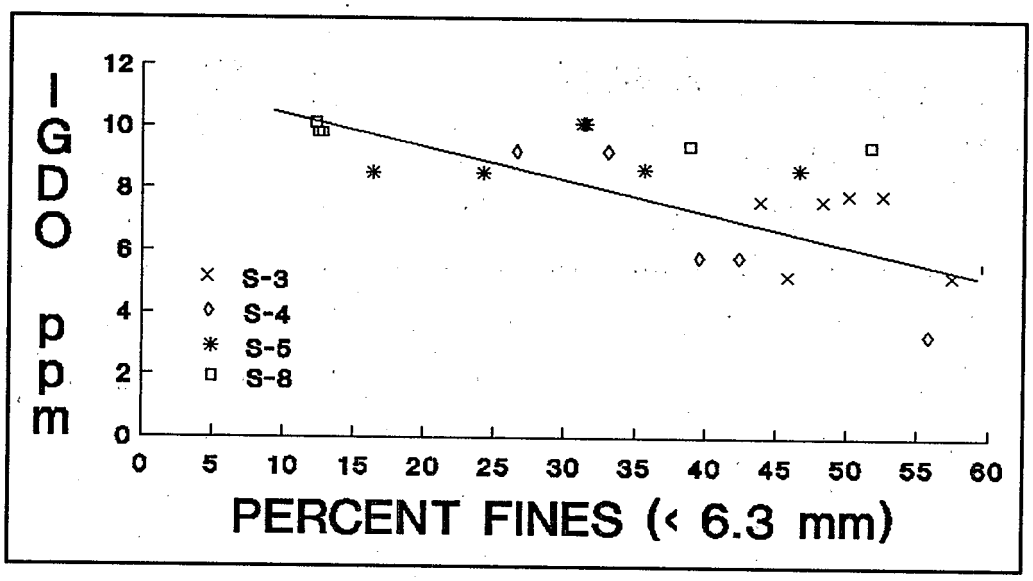

Figure 1.—Effect of fine sediment on dissolved oxygen in the intragravel incubation environment measured on Rock Creek, Idaho.

Also, survival of salmonid embryos is positively correlated with apparent velocity and permeability. Dissolved oxygen affects both emergence success and timing.

A methodology for monitoring sediment impact on incubation of salmonid embryos *in-situ* has been developed. The technique measures intragravel dissolved oxygen, percent intragravel fine sediment, and percent survival of embryos to emergence in artificial egg pockets. Monitoring in natural egg pockets has proven ineffective and destructive to the beneficial use. The artificial redd technique permits measurement of the fine sediment infiltrating egg pockets and the dissolved oxygen concentration surrounding the incubating embryos. These values are compared with egg survival and alevin escapement from the artificial egg pockets.

Results

Testing in Idaho has shown the technique to be useful in varying seasons and stream conditions. The validation work also verified that fine sediment impairs permeability within egg pockets resulting in dissolved oxygen depression sufficient to suffocate incubating eggs (Maret et al. in prep.). Figure 1 shows that as fine sediment approaches 40 percent, dissolved oxygen within the intragravel environment decreases to levels impairing growth and survival of incubating embryos. Tests conducted in substrates with coarser sediments showed little or no dissolved oxygen depression, but mortalities by entrapment were observed among developed alevins trying to escape heavily sedimented egg pockets (Burton et al. 1990). Excessive fine sediment may also affect growth and condition of surviving embryos as indicated by conclusions from the Rock Creek Rural Clean Water Program study (Maret et al. in prep.).

The proposed salmonid spawning criterion is based on dissolved oxygen concentrations within the incubation environment. Attempts to establish a permeability criterion indicated that it would be technically unfeasible. In practice, intergravel dissolved oxygen should be a good surrogate for gravel permeability. No standard methodology currently exists to quantify escapement success. An interim standard is being developed for use until functional relationships between percent fine sediment and alevin survival to emergence have been established.

The proposed criterion is:

- Nonpoint source activities shall not cause intragravel dissolved oxygen in spawning gravels to decline, below a weekly average of 6 milligrams per liter.

Salmonid Survival in the Intercobble Environment

Methods

The interstitial space found in streambed cobble habitats is important to survival of juvenile salmonids. These fish use the interstitial space primarily for feeding and refuge cover, especially in winter. When this habitat has been replaced by the intru-

sion of fine sediment, salmonids must find other suitable habitat either by migrating from the stream reach or within the same stream. Chapman and McLeod (1987) found in their literature review that real and detectable relationships exist between land-disturbing activities and increased fine sediment in the aquatic environment. The weight of evidence indicates that areas with high embeddedness tend to have lower densities of salmonids and additional sediment that reduces living space, increases mortality.

A protocol for measuring embeddedness has been developed by Burns and Edwards (1985). The method was further refined for sampling design and statistical treatments by Skille and King (1989). Using this approach, cobbles within a specified size range are drawn from a 60 cm diameter sampling plot on the bed of the stream. Each cobble is measured for depth embedded in fine sediment. Areas within the plot completely covered by fine sediment are weighted as fully embedded. The mean of all measurements on the plot is counted as one sample. A number of samples (or plot measurements) are collected from random locations in the stream over a stream reach equal to 20 times the channel width. The number of samples (plots) ranges from 10 to 50, depending on the sample variability of the stream. As variability increases, more samples are required. The standard sample size is equal to the number needed to predict the mean cobble embeddedness within a 95 percent confidence interval on the t statistic.

Results

Using this technique for measuring cobble embeddedness has allowed quantification of intercobble habitat degradation resulting from excessive sedimentation to the stream. Embeddedness measurements have always demonstrated a high inverse correlation to living space used by the fish. In addition, streams impacted by nonpoint source activities show significantly higher embeddedness as compared with minimally impacted control sites.

The proposed criterion is:

- No statistically demonstrable increase, at the 95 percent confidence interval, in natural baseline percent embeddedness as the result of nonpoint source activities shall be permissible in salmonid rearing habitats. Impacts of sedimentation on interstitial space habitats important to salmonid rearing will be assessed by measurement of cobble and rubble percent embeddedness. Baseline percent embeddedness will be determined by a quantitative technique in-stream reaches with similar geomorphology and stream power which are unaffected by nonpoint source sedimentation. A percent embeddedness value will consist of a mean at the 95 percent precision level of the t statistic.

Summary

Excessive fine sediment impairs salmonid growth and recruitment. The effect within the incubation environment is to reduce intragravel flow velocity and therefore the delivery of oxygen to developing embryos. Fine sediment intrusion in the top layers of the egg pocket may also restrict emergence after development of the embryos.

Intercobble space is a critical habitat for juvenile salmonids. Replacement by fine sediment severely degrades this environmental requisite.

Quantitative methods for estimating and monitoring sediment effects on salmonids have been specified. As a result, biocriteria have been developed and proposed for inclusion in Idaho's water quality standards.

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Biomonitoring Methods in the Tennessee Valley

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ABSTRACT

The Tennessee Valley Authority is expanding its water monitoring program to include biomonitoring methods. Methods being compared include EPA's Rapid Bioassessment Protocols for both fish and macroinvertebrates, Karr's IBI as modified by Saylor, extensive quantitative and qualitative macroinvertebrate sampling, the EPT evaluation of the insect community used by North Carolina Department of Environmental Management, periphyton sampling, and both laboratory and ambient toxicity tests where appropriate. This effort will require the adaptation of techniques and analyses to reflect conditions in the chemistry, and stream habitat quality will also be assessed. These comparisons will be performed on a set of rivers that have been impacted by agricultural use, heavy metals, or xenobiotics. Results will be used to determine the efficiency of each method and its utility in evaluating specific impacts. From these data an organized, cost-effective, and adaptable approach to water quality assessment will be developed for the Tennessee Valley.

Introduction

The Tennessee Valley Authority (TVA) performs biological surveys on a wide variety of streams and rivers in the Tennessee River basin. The sampling strategy often is designed to complement chemical and physical monitoring, and typically requires collecting both fish and macroinvertebrates. Recent efforts have included analyses of community-level changes relative to pollution (e.g., IBI), toxicity tests, and measurements of various contaminants in fish tissue. Even with intensive field work from March to September, water quality can be assessed on only a fraction of the watershed.

To improve and expand this monitoring effort with a fixed or decreasing budget and limited manpower, efficiency must be increased. The TVA is conducting a comparison of biomonitoring methods in Spring 1991.

TVA has employed a variety of techniques to assess the water quality of streams and rivers, including chemical, physical, and biological surveys (fish, macroinvertebrates), and toxicity tests. While new methods appear in the literature every few years and others are modified, the appropriateness of the methods TVA is currently using or the utility of newer methods, such as the rapid procedures being promoted by the U.S. Environmental Protection Agency (1989) have never been evaluated.

Currently, Aquatic Biology staff evaluate water quality from analysis of the fish community using several standard procedures: the Index of Biotic Integrity (IBI) developed by Karr (1981); an assessment of the macroinvertebrate community with EPT taxa (Ephemeroptera, Plecoptera, Trichoptera), total taxa, and percent composition; and chemical analyses of water samples. These methods are labor intensive, expensive, and require months to produce results. Such a delay is often unacceptable.

With the expansion of TVA biomonitoring efforts, it is more important than ever to improve field and analytical efficiency. The recently published

EPA manual on rapid bioassessment procedures includes faster, more field-oriented methods to determine the health of the fish and macroinvertebrate communities (U.S. Environ. Prot. Agency, 1989). These methods reduce sampling and processing time, and permit faster determination of water quality. However, little information is available on the validity of these rapid methods in comparison with traditional methods or their utility under a wide variety of circumstances. In addition, the project will determine the feasibility and validity of *in situ* toxicity tests, which may be less expensive than laboratory toxicity tests. Finally, the project will assess the effectiveness of algal indices of water quality.

The comparison study will be conducted in Spring 1991, on four rivers (Middle Fork Holston, Pigeon, Big Sandy, and Oostaunala Creek). These rivers vary in size and have different water quality problems. The traditional IBI and macroinvertebrate assessments will be coupled with rapid methods proposed by EPA, *in situ* toxicity tests with fathead minnows and *Ceriodaphnia dubia*, algal identification, and analyses of water chemistry. The project hopes to determine which methods are fastest, least expensive, and most accurate for characterizing water quality in various sized streams with different water quality problems.

Summary

Materials and Methods

- Biological and chemical samples taken in four rivers (see Fig. 1).
- Analytical methods
  - IBI
  - Complete macroinvertebrate sampling
  - Rapid bioassessment methods
  - *In situ* toxicity tests
  - Water chemistry analyses

Results

- Comparability of various biomonitoring methods in identifying water quality problems
- Ability of various methods to discriminate between point and nonpoint source pollution
- Sensitivity of methods in streams and watersheds of different sizes
- Cost/benefit analysis of methods
- List of suggested methods, based on budget, known problems, stream conditions.

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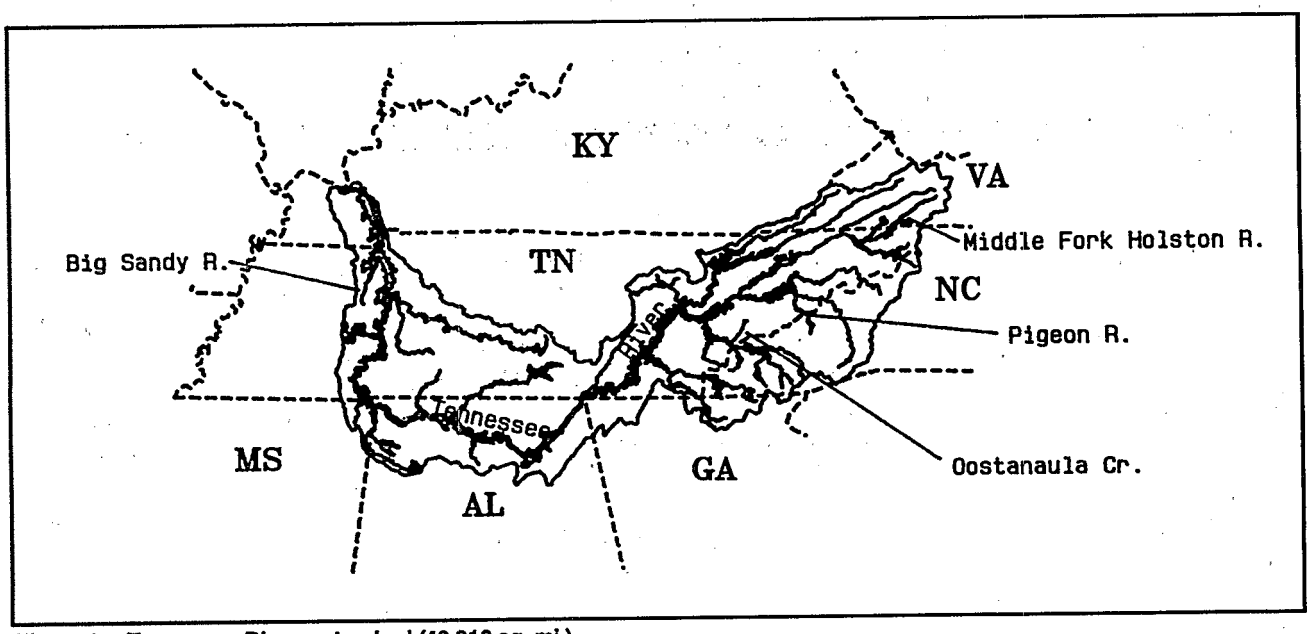

Figure 1.—Tennessee River watershed (40,910 sq. mi.).

Development of Biological Criteria for Use in Maine's Water Quality Classification Program

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ABSTRACT

The Maine Department of Environmental Protection has begun the process of incorporating the use of biocriteria into its water assessment program. The Department established narrative standards that described uses and characteristics of each class of water within its classification system. Macroinvertebrate data were used as the basis of a three-stage protocol to assign water classifications to specific waterbodies and target those that could benefit from remediation. The first stage classifies samples into one of four water quality groups according to macroinvertebrate characteristics, based on a linear discriminant model. This model produces a 74 to 88 percent probability of correctly placing a biological community sample into its appropriate classification. The second stage refines the prediction by applying class-specific criteria to the samples to assess attainment of the unique standards of a given classification. The third stage utilizes the expertise of biologists to adjust the scores obtained in stages one and two. The result is a rapid and accurate decisionmaking tool that utilizes both statistical probability and human judgment to assess aquatic life in waterbodies in the state of Maine.

Introduction

The State of Maine began its development of biocriteria by establishing narrative standards for each of the classes within its water classification system. The purpose of these narrative standards was to identify specific conditions of the biological community that supported the uses and characteristics of each class of water, rather than to merely establish a ranking from "good" to "poor." These standards and accompanying statutory definitions identify specific attributes of the biological community for evaluation (Courtemanch and Davies, 1987).

The benthic macroinvertebrate community was chosen as a practical community component for sampling and evaluation. Each class is distinct from the others and thus requires a different set of metrics or different criteria values for the metrics. Maine is now at the stage of proposing numeric criteria to interpret and evaluate the narrative standards.

The original proposal of a hierarchical test design (Courtemanch and Davies, 1987) has been expanded to include a three-stage test. The first stage includes general tests of the macroinvertebrate data, followed by the application of the data to a linear

discriminant model that determines the probability for placement in any class. The second stage tests specific attributes of the assigned class, using both data compared to a reference site when available, and unreferenced data. The final stage provides for the use of professional judgement to adjust the scores of the first two stages, taking into account both ambiguous findings resulting from data collection and processing, and unique habitat conditions that influence community development.

First-Stage Criteria

To develop the first-stage model, an initial classification assignment reflecting the narrative standards had to be constructed. Three agency biologists, familiar with macroinvertebrate data interpretation, independently and blindly (site identity unknown) evaluated data from 145 samples and assigned one of four classes (A, B, C, or nonattainment of the lowest class.) Next, results of this evaluation were compared. There was unanimous agreement in the assignment for 114 (79 percent) of the samples. The remaining samples were then reevaluated collectively by the biologists with site information revealed and a consensus classification was assigned. A subset of the data was also provided blindly to two nonagency biologists from a technical review committee overseeing the criteria development process. Concurrence was found for 86 percent of the samples between the agency biologist assignments and nonagency biologists. It was concluded that there was substantial agreement in the biological interpretation of the statutory language.

Following classification of the test data set, it was possible to develop a model that best simulated the biologists' decisions. To do this, all the criteria that any of the agency biologists had used in making their determinations were quantified. The outcome was 31 variables (Table 1). No reference comparative variables (e.g., percent change, similarity) were included at this stage.

Factor analysis and stepwise discriminant analysis reduced this number to nine quantitative variables (Table 1). These nine discriminating variables were used to build a linear discriminant model (Green and Vascotto, 1978). A jackknife procedure (Mosteller and Tukey, 1977) was used to assure the stability of the model using four runs, each with 25 percent of the samples removed. The model assigns a classification based on the highest probability for membership in one of the four classes.

Table 1.—Metrics used in initial biologist classification. Underlined metrics are those selected for the discriminant model.

1. Total abundance (log transformed)
2. Generic richness
3. Ephemeroptera abundance (log transformed)
4. Plecoptera abundance (log transformed)
5. Ephemeroptera abundance / Total abundance
6. Plecoptera abundance / Total abundance
7. Ephemeroptera richness
8. Ephemeroptera richness / Generic richness
9. Plecoptera richness
10. Plecoptera richness / Generic richness
11. Ephemeroptera + Plecoptera + Trichoptera (EPT) richness
12. EP richness / Generic richness
13. EPT richness / Diptera richness
14. Non-EPT richness / Generic richness
15. Oligochaete abundance / Total abundance
16. Hirudinea abundance / Total abundance
17. Gastropoda abundance / Total abundance
18. Chironomidae abundance / Total abundance (log transformed)
19. Diptera richness / Generic richness
20. Tanypodinae abundance / Total abundance
21. Tribelos abundance / Total abundance
22. Chironomus abundance / Total abundance
23. Hydropsyche abundance
24. Hydropsyche abundance / Total abundance
25. Glossosoma abundance / Total abundance
26. Brachycentrus abundance / Total abundance
27. Percentage of predator abundance
28. Ratio of collector-filterers + collector-gatherers to predators + shredders
29. Number of functional feeding groups represented
30. Hilsenhoff Biotic Index
31. Shannon-Wiener diversity index

Table 2 compares the classification assignment to the model-predicted classification. In making a decision on attainment, the model must correctly predict class assignment; however, errors where the model predicts a higher class are tolerable since attainment decisions are based on minimum conditions. Conversely, the model can err by predicting that a community is a lower class than assigned (right of the matrix line). The frequency of this type of error was judged to be acceptable because the potential remains for the error to be corrected in the second and/or third stages of the protocol. The classification probabilities are used as the raw score for the first-stage test decision.

Table 2.—Percentage of concurrence between assigned and model predicted classification.

	Model predicted class			
Assigned class	A	B	C	NA
A	74	26	0	0
B	13	76	11	0
C	0	25	75	0
NA	0	0	12	88

Second-Stage Criteria

Second-stage analysis is separated into that for samples with high water quality reference sites (compatible with Class A standards), and those without reference sites. While the first-stage analysis provides a prediction and probability based on variables or discriminators common to all classes, the second stage provides further testing of the data using unique tests within each class. These tests are chosen to address aspects of the standards that are specific for each class, and which have not been used to build the linear discriminant model.

Second-stage criteria first assess the presence and abundance of indicator taxa. Indicator taxa were selected based on: (1) significant occurrence in the entire data set (occurring at ≥10 percent of all sites), thus being sufficiently common to be a good indicator; (2) dominant abundance in the specific class (≥60 percent of total abundance in the dataset occurring in the specified class); and (3) significant occurrence within the specified class (occurring at ≥25 percent of the sites in the specified class). Some of the strongest indicator taxa include:

- **Class A:** *Leucrocuta, Paragnetina* (Plecoptera); *Serratella, Eurylophella* (Ephemeroptera); *Brachycentrus, Psilotreta* (Trichoptera)
- **Class B:** *Baetis* and *Tricorythodes* (Ephemeroptera); *Chimarra, Neureclipsis* and *Lepidostoma* (Trichoptera); *Simulium* (Diptera)
- **Class C:** *Dicrotendipes* and *Conchapelopia* (Diptera).

An additional approach used in the second stage of analysis draws on information available from comparisons of the test site to a clean water "upstream" reference site. Reference site criteria were set to ensure closely matched habitat conditions (reference sites usually located upstream on the same waterbody), and very high water quality (reference sites must attain Class A standards). Comparative indices are computed for these sites and the results are used to strengthen confidence in the likelihood of a site belonging to a given classification (Fig. 1).

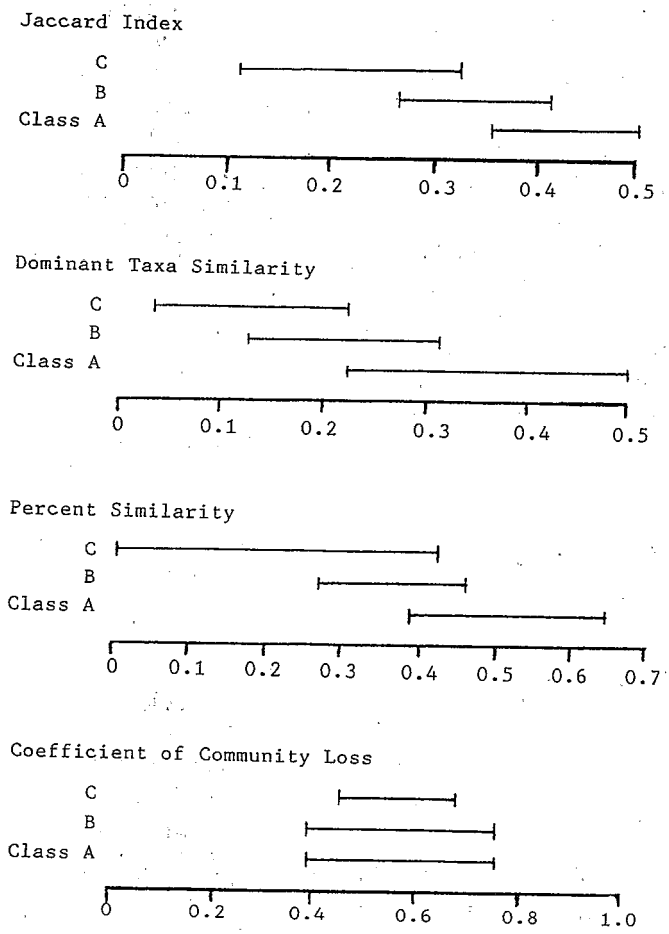

Figure 1.—Confidence intervals (95 percent) for selected comparative metrics used in second stage.

Third-Stage Criteria

The third-stage analysis does not set criteria, but rather provides a mechanism for adjusting the scores in stages one and two. This process relies on professional biological judgement, as well as documented evidence of conditions that can result in atypical findings. Examples of conditions that could trigger adjustment mechanisms are unusual habitats, natural or human-induced disturbance of the sample site, or known or suspected problems with sample collection or analysis. Following are some examples of unusual habitats:

- **Lake outlets/regulated flows:** Influence variables including total abundance, variables based on relative abundance, Diptera abundance, hydropsyche abundance, percent collector-filterers.

- **Substrate character** (especially soft-bottom habitats): Influences the biological pool available to colonize the artificial substrates, thus affecting richness-related measures.

- **Low velocity:** Tend to suppress total abundance and filtering activity.

- **Tidal movement:** Alters community composition compared to unidirectional flow, loss of filtering activity.

- **Anomalous samples:** Unusual samples, particularly where disturbance is suspected (e.g., spates, flow control, vandalism), may be discarded.

Summary

The State of Maine has combined statistically derived predictions of classification attainment with criteria based on class-specific ecological attributes and professional biological judgement to develop a water quality classification attainment decision protocol based on samples of benthic macroinvertebrates. The protocol can be used for sites with or without an associated high quality reference site. The result is a statistically defensible and reproducible decisionmaking tool that also allows for the exercise of professional biological judgement.

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The Relationship of Chironomidae (Diptera) Community Structure to Chlordane Levels in Sediments of Streams Near St. Louis, Missouri

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ABSTRACT

Chironomidae pupal exuviae were collected along tributaries of the Meramec River near St. Louis, Missouri, at sites with varying levels of technical chlordane in the sediments to test the efficiency of this collection technique as a method for detecting stream sites impacted with moderate to high levels of chlordane. Exuviae were identified to lowest possible taxonomic level and analyzed relative to chlordane levels found at the collection sites. Sites were placed into high and low chlordane categories and comparisons were made between them using the Kruskal-Wallis test. Of the 18 total sites sampled, 13 were used in the analysis. Five of the sites were excluded because of extraneous factors that may have altered the faunal composition (e.g., sewage treatment plants, other pollutants, spring influence, disruption of the site). Comparisons of mean community index values between high and low chlordane categories and mean percentage abundance of taxonomic groups, individual taxon, and functional feeding groups between high and low chlordane categories revealed many significant differences. Overall, numbers of taxa and pollution-sensitive taxa were reduced in the high chlordane category. The high chlordane sites were dominated by detritus-feeding taxa (mostly Chironomini), pollution-tolerant algavores (mostly *Cricotopus* species), and pollution-tolerant predators within the genus *Procladius*. It was concluded from these results that stream sites containing sediments with high chlordane levels affect the chironomid community structure in a predictable pattern and that the collection of pupal exuviae was an effective technique for sampling the chironomid community. This sampling protocol could be used for preliminary surveys of sites suspected of chlordane contamination or long-term monitoring of impacted sites, thus decreasing time required in the field and the lab.

Background

When adult Chironomidae (nonbiting midges) emerge from their aquatic environment, they leave behind a pupal exuvia that floats for a period of up to two days and tends to accumulate with other flotsam at catch points along the course of a stream or river (Wiederholm, 1986). Collecting and identifying chironomid pupal exuviae has shown to be an effective method for surveying lotic systems and for studying the impacts of organic sewage enrichment and heavy metals on those systems (Ferrington, 1987; Wilson, 1988). This project was undertaken to examine the applicability of this sampling methodology for detecting stream sites contaminated with relatively high levels of chlordane as compared to other local streams. If distinct differences or predictable patterns could be found

in the chironomid communities based on the relative chlordane levels in the sediments at the sites, this method of biological monitoring could prove to be useful as a preliminary survey technique for detecting high levels of chlordane at sites or as a long-term monitoring technique to observe recovery at sites.

Chironomids are extremely valuable as a study organism for this project because of their high species diversity, common abundance in most freshwater systems, and generally low mobility as mature larvae (Ashe et al. 1987). Presently, many chlordane monitoring programs are based on tissue analysis of fish (carp), which are highly mobile and not continually exposed to the sediments where chlordane tends to accumulate. Therefore, studying only carp or other fish increases the probability of wrongly assessing a site as nonimpacted. Many species of Chironomidae spend most of their larval life burrowing and feeding on sediments. Other species may live on top of the sediments or feed on suspended particles of organics (Oliver, 1971). Chironomid larvae occupy numerous micro-habitats, thus relaying increased amounts of information about the dynamics of a freshwater system to the researcher.

Methodology

On May 10 and 11, 1988, chironomid pupal exuviae were collected from tributaries along the Meramec River, approximately 40 miles southwest of St. Louis, Missouri. The collections coincided with a sediment sampling survey undertaken by the Missouri Department of Conservation to isolate possible sources of chlordane in the tributaries.

The pupal exuviae samples were collected using a standard protocol in which each site is sampled by dipping a white pan into catch point areas along the stream edge where flotsam has accumulated (Ferrington, 1987). "Dips" from all possible catch points at a site are poured through a 125 micron sieve for 10 minutes. The debris retained in the sieve is then washed out into the pan using 80 percent ethanol and poured into a labelled jar. The samples within the jars are sorted in the lab by hand-picking the exuviae under a dissecting microscope. Common and conspicuous exuviae may be identified unmounted; however, rare and minute taxa must be mounted on slides and identified using a compound microscope. At least two representatives of each taxon are permanently mounted for positive identification and voucher material. All identifications and abundances are recorded on data sheets for future analysis.

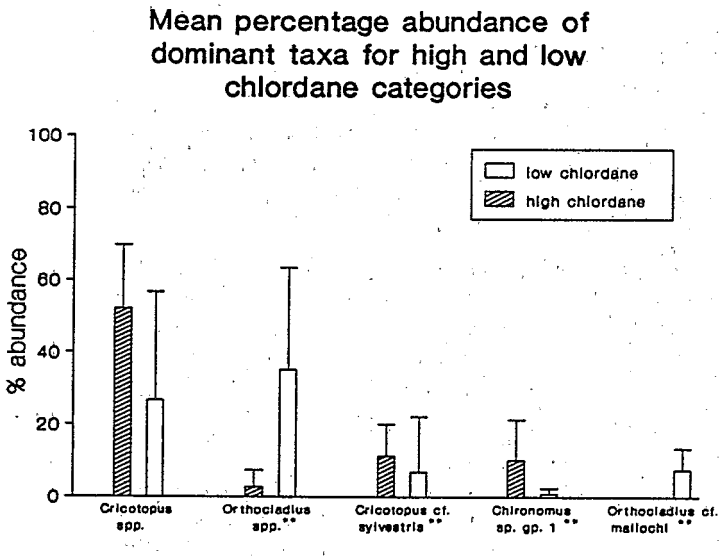
Discussion

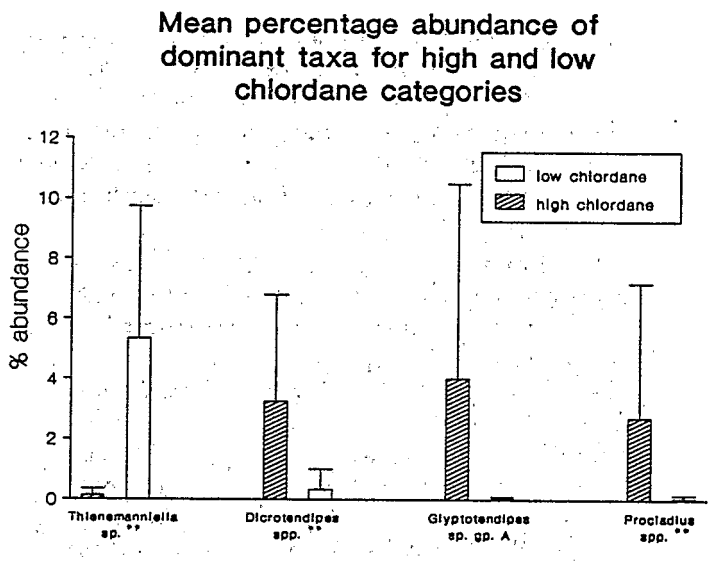
Correlations, cluster analysis, and Kruskal-Wallis tests were initially applied to the data. Only the results of the Kruskal-Wallis tests are presented, as they reflect the overall patterns found in all the other analyses (Fig. 1).

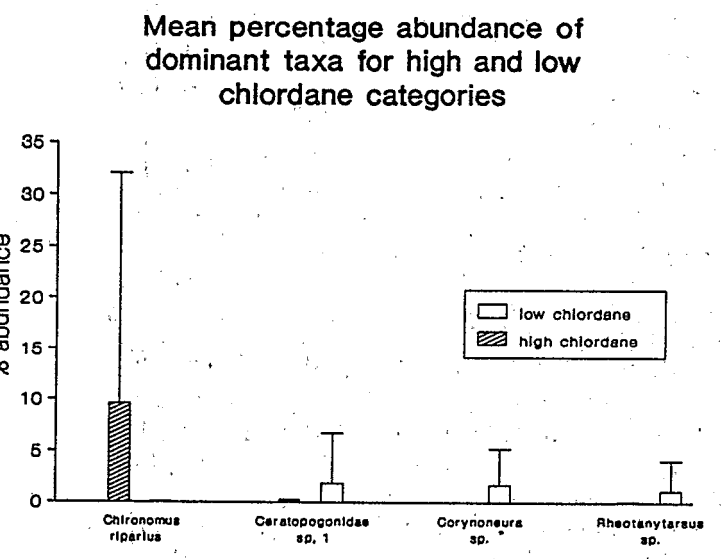
The chironomid fauna at sites with high technical chlordane in the sediments were dominated by pollution-tolerant taxa such as *Chironomus* species, *Dicrotendipes* spp., *Glyptotendipes* sp. gp. A, *Cricotopus* species, and *Procladius* spp. Most of the Chironomini species are detritivores that burrow and feed on sediments in streams. These taxa should have been exposed to the highest levels of chlordane. Species within the genus *Procladius* are predators that usually occur on top of the sediments in slower-moving water (Beck, 1978).

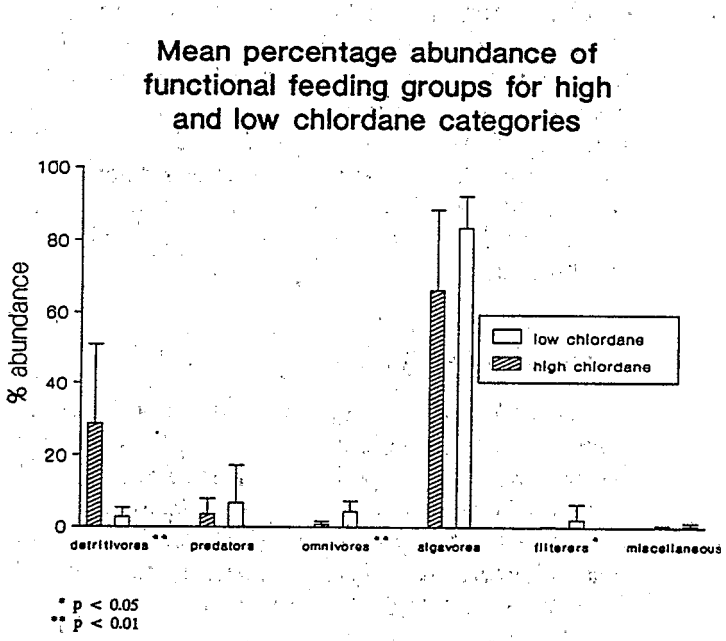
High chlordane sites also showed less omnivores and filterers. These two functional feeding groups, although not found burrowing in sediments, are still exposed to them. Omnivore is a general category that describes taxa feeding on a variety of foodstuffs (algae, sediments, animals) and that tend to move between microhabitats. Filterers may build tubes on the surface of sediments, rocks, or plants and feed on suspended organic material. Their exposure to contaminated sediments while possibly not as high as detritivores, may have been enough to decrease their abundance in highly contaminated sites. Taxa within the omnivore and filterer groups tend to be less tolerant of stressful conditions than taxa within the detritivore group (Beck, 1978).

Analysis of algavores revealed a large shift from high chlordane sites to low chlordane sites. High chlordane sites contained mostly *Cricotopus* species, whereas low chlordane sites had the largest abundance being among *Orthocladius* species. *Cricotopus* species were present at low chlordane sites, but in fewer numbers than at high chlordane sites. The most obvious difference between high and low chlordane sites was the presence of more algae-feeding taxa besides *Cricotopus* and *Orthocladius* species at low chlordane sites. These taxa, such as *Tvetenia* cf. *calvescens*, *Thienemanniella* sp., *Eukiefferiella* spp., *Para-metriocnemus* spp., and *Rheocricotopus* sp., are usually listed as occurring in relatively "good" water quality. Algavores usually restrict feeding to filamentous mats of algae or single-celled algae on the surfaces of rocks, plants, and the benthic substrate. This functional feeding group is probably least exposed to chlordane in the sediments; however, they may be affected more by chlordane in the water column than other groups. Because chlordane usually enters the stream system









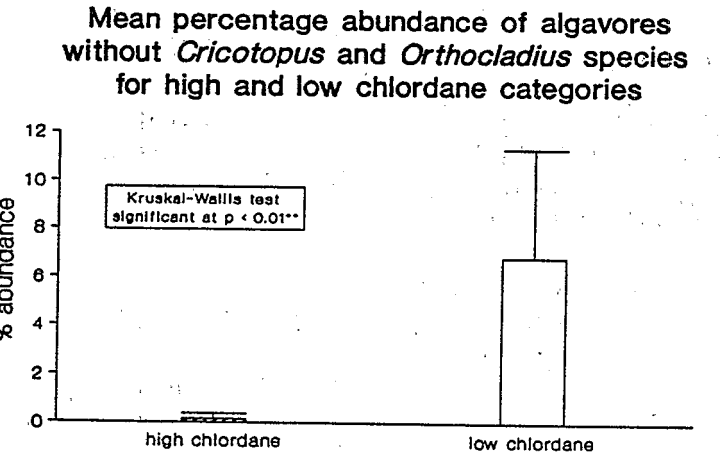


Figure 1.—Kruskal–Wallis tests between high and low chlordane categories.

during runoff events, decreased presence of algavores may reflect recent "slugs" of chlordane in the water column.

Summary

Collections of Chironomidae pupal exuviae have been shown to be useful in assessing impacts of organic sewage enrichment and heavy metals on stream systems by past researchers. The technique has also been advantageous over larval collections for several reasons: (1) larval collections usually miss very small larvae; (2) larval collections tend to miss some of the microhabitats in which larvae occur; (3) processing in the field and lab is usually much faster using the exuviae method; and (4) exuviae represent only one age class, whereas larvae

may be collected from many age classes, thus compounding difficulties in their identification.

In this study, collections of pupal exuviae were successful in revealing differences between stream sites impacted with high and low levels of technical chlordane. High chlordane sites contained less taxa than low chlordane sites, and the taxa present at high chlordane sites tended to be pollution-tolerant. These results suggest that the collection of chironomid pupal exuviae may be useful as a preliminary survey tool or long-term monitoring technique in lotic systems impacted with chlordane. Factors that could influence faunal distribution in lotic systems besides a target pollutant, such as chlordane, must be taken into consideration. For example, it may be impossible to distinguish the effects of a sewage treatment plant and chlordane on the same stream system.

At present, no indicator species can be identified to detect the presence of chlordane. As this is the first project exploring the relationship of Chironomidae communities to chlordane in the sediments in which they occur, the results should be analyzed for general patterns, perhaps identifying indicator communities instead of individual taxon. Analysis of ecological factors, such as feeding habits of larvae, may prove to be most useful for applying this technique to other ecoregional areas because taxa in similar order streams may shift between ecoregions, while the functional feeding groups should remain stable between ecoregions.

ACKNOWLEDGMENTS: This project was funded in part by the U.S. Environmental Protection Agency through the National Network for Environmental Management Studies fellowship program (Project Control Number U-913134-01-0). Work space and technical assistance were provided by the Water Quality and Freshwater Ecology section of the Kansas Biological Survey at the University of Kansas.

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Regional Standardization of Taxonomy

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ABSTRACT

The Clean Water Act requires the implementation of aquatic habitat monitoring, and numerous surveys have been conducted to characterize those biological communities. A fundamental component of these surveys is taxonomic data that list the taxa and their abundance. Typically, several surveys are conducted by both public and private organizations within a given geographic region. A problem in realizing the full potential of these data is the lack of taxonomic consistency among surveys. The Southern California Association of Marine Invertebrate Taxonomists (SCAMIT) was formed in 1982 to provide regional standardization among benthic marine surveys in the southern California bight. SCAMIT schedules a yearly agenda of taxonomic topics, including regular exchange of specimens that have been noted as inconsistently identified or are new to science. National and regional taxonomic experts lead workshops presenting innovative identification techniques, new taxonomic keys, and review of voucher collections. A central voucher collection is maintained, consisting of specimens exchanged and reviewed at meetings. The results of these meetings and workshops are distributed in a monthly newsletter. Several aspects of biological criteria development can benefit from regional standardization of taxonomy. The biological survey design should include a component for regular calibration of taxonomic data. Selection and assessment of regional reference sites should utilize regionally standardized data. Selection of aquatic community components for detailed analysis, whether for statistical manipulation or toxicity testing, should be supported by the survey's taxonomic data. Biological indices, commonly used as regulatory tools to manage complex environmental impact issues, are dependent on quality and comparability of the underlying taxonomic data. SCAMIT's activities have greatly enhanced taxonomic quality control and standardization within and among benthic marine data bases in southern California. Implementation of taxonomic standardization in other regions should serve to improve national biological criteria for surface water programs.

"What's the use of their having names," the Gnat said, "if they won't answer to them?"

"No use to them," said Alice: "but it's useful to the people who name them, I suppose."

— Lewis Carroll, *Through the Looking Glass*

The Clean Water Act passed in 1972 requires that treated wastewater and industrial flows into aquatic habitats must be monitored for impacts on biological communities. These monitoring surveys collect biological data on a regular basis and typically generate large data sets representing hundreds of species or taxa. The basic component of a biological survey is taxonomic data, a listing of names and abundance levels for organisms collected. Taxonomic data are analyzed utilizing various statistical methods. The results are interpreted, reported, and used in regulatory decisionmaking. Taxonomic consistency, subsequent analyses, interpretations, and regulatory decisions cannot be made with confidence. Implementation of taxonomic standardization both within and between regional monitoring programs is a means of achieving and maintaining taxonomic consistency.

In heavily populated and industrialized coastal regions such as southern California, several programs monitoring marine biological communities are conducted by both public and private organizations, and considerable amounts of money and effort are expended. A largely unrecognized obstacle to realizing the full potential of these data is the lack of taxonomic consistency. Taxonomic consistency is difficult to maintain for a variety of reasons: (1) published or unpublished regional taxonomic literature is rarely comprehensive (if it exists at all); (2) undescribed species are common in many habitats; (3) standard taxonomic usage changes over time as new research becomes available and new species are recognized; (4) taxonomic staffs vary in skill, experience, and capabilities; (5) personnel changes within a program lead to change—drift over time; (6) sibling species or sexual dimorphism within a species are common.

Statistical analyses are affected when taxonomic inconsistencies exist in the supporting data. Community indices can be inaccurate due to overestimation or underestimation of species numbers by different taxonomists within a survey. Changes over time in the taxonomy can lead to inaccurate community classification analyses. Tests of statistical significance may be compromised by inconsistency between taxonomists working on the same survey. Comparison of data between programs becomes difficult or impossible when taxonomists either within or among monitoring programs are inconsistent.

Other types of data can be affected by inconsistent taxonomy. Chemical results from bioaccumulation samples can be compromised if the animals used are not consistently identified during collection. Inconsistently or incorrectly identified species used in toxicity testing could lead to misinterpretation of test results. Easily confused sibling species may respond differently to tested contaminants.

There are a number of consequences for regulatory decisionmaking when the analysis of the data is affected. Test sensitivity may be reduced by taxonomic inconsistency. Failure to identify organisms consistently may lead to spurious variability in a database, and decrease the sensitivity of a statistical test by increasing the variance at reference (control) stations. Evidence may be contradictory because of taxonomic confusion. Contradictory evidence can be introduced if the same organism is identified differently in different samples within a single survey, or over time. Thus an "indicator" species could be present (or absent) depending, not on the environmental conditions, but on inconsistent taxonomy. FAlse violations may result from invalid toxicity test results. Toxicity tests based on mixed lots of test animals from similar, but different, species may lead to spurious test results. These caveats might results in the appearance of a discharge permit violation, when none had actually occurred. Approval/denial decisions may rest on questionable evidence. Requests for environmentally safe dischargers could be denied; or worse, requests for environmentally safe dischargers approved, because decisions are based on evidence from flawed taxonomic data. All of the above types of compromised data can lead to inappropriate action at the regulatory level, since such actions must be based on the "best available data and analyses."

SCAMIT was formed in 1982 by a group of marine biologists who recognized the value of regionally standardized data. The nonprofit organization is supported by contributions from regulated agencies, grants from industry, and annual dues. SCAMIT's goals are "to promote the study of marine invertebrate taxonomy and develop a regionally standardized taxonomy." A variety of activities help achieve these goals.

An annual agenda of monthly meetings covering taxonomic problem areas is scheduled (see Table 1). Members regularly exchange specimens that have been inconsistently identified or are new to science. National and regional experts lead taxonomic workshops presenting innovative identification techniques, new keys, and review of voucher collections. Information from meetings and workshops, new literature citations, species voucher sheets, and announcements of interest are distributed to members through a monthly newsletter. Centralized literature and specimen voucher collections are maintained and updated by SCAMIT. Membership in SCAMIT is open to anyone, and currently over 100 individuals and more than 25 organizations participate.

SCAMIT's activities have greatly enhanced taxonomic quality control and regional standardization within and among benthic marine invertebrate data bases in southern California. The organization recommends the following guidelines for implementing taxonomic standardization: (1) recognize the nature and extent of the problem; (2) improve communication between taxonomists; (3) generate new taxonomic information; (4) establish and maintain centralized literature and specimen voucher collections; and (5) support long-term maintenance of ecological survey samples.

Several aspects of biological criteria development can benefit from regional standardization of taxonomy. Biological survey design should include a component for regular calibration of taxonomic

Table 1.—SCAMIT Agenda 1990–91.

June 11	*Nassarius* by Don Cadien at Cabrillo Marine Museum, San Pedro, California
July 9	Hydrozoa by John Ljubenkov at MEC Analytical Systems, Carlsbad, California
August 13	Polychaete Scale Worms by Ross Duggan at Allan Hancock Foundation, University of Southern California
September 10	Latin Grammar for Taxonomy by John Ljubenkov at Cabrillo Marine Museum, San Pedro, California
October 15	Epitoniidae by Helen DuShane at Los Angeles County Museum of Natural History
November 19	Hesionidae by Ron Velarde at Allan Hancock Foundation, University of Southern California
December 10, 11	Barnard Amphipod Workshop at Los Angeles County Museum of Natural History
January 14	Polyclad Flatworms by John Ljubenkov, Carol Paquette, Tony Phillips at Cabrillo Marine Museum, San Pedro, California
February 11	Lovell Taxonomic Consulting, Vista, California
March 11	Nuculanidae by Paul Scott at Santa Barbara Museum of Natural History
April 8	*Tharyx* by Tony Phillips at Allan Hancock Foundation, University of Southern California
May 13, 14	Bryozoan Workshop with Dr. William Banta at Cabrillo Marine Museum, San Pedro, California

data through quality assurance/quality control programs. Selection and assessment of regional or program reference sites should utilize regionally standardized data. In some areas, reference sites might be shared by more than one discharger. Selection of aquatic community components for statistical manipulation and detailed analysis should be discussed with taxonomists to avoid selection of problem species. Taxonomic data should support the selection of endemic species for toxicity testing. Biological indices, commonly used as regulatory tools to manage complex environmental impact issues, are dependent upon the quality and comparability of the underlying taxonomic data. Implementation of taxonomic standardization in other regions should serve to improve national biological criteria for surface waters programs.

> *"If one does not know the names, one's knowledge of things is useless."*
>
> — *Isidorus*

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Integrated Chemical and Biological Monitoring of Sun Creek, McPhearson County, Kansas, U.S.A.

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Water quality in a portion of the Sun Creek catchment, a stream system which receives the effluents from a municipal wastewater treatment facility and an oil refinery, was evaluated based on collection of surface floating Chironomid pupal exuviae and water chemistry. Utilizing cluster analyses as the initial basis for evaluation, both pupal exuviae and chemical data provided supportive information. Cluster analysis of the Chironomid data identified tolerant taxa, facilitative taxa and intolerant taxa. For the chemical parameters the analyses identified a group of parameters associated with organic enrichment and a group of parameters which define ambient water quality. Both chemical data and Chironomid data classified sites similarly, however, Chironomid data provided somewhat better resolution of differences between sites. Correlation analyses between the chemical data and the Chironomid data revealed that the taxa identified as tolerant were positively correlated with the enrichment parameter while those identified as intolerant were negatively correlated with the enrichment parameter. The simultaneous collection of both chemical and biological data provided complementary information which water quality managers could use to make decisions and plan options. Each element provided a slightly different perspective which, when taken together, clearly defines water quality problems and processes.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

The Use of Biocriteria in the Ohio EPA Biological Monitoring and Assessment Program

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Ohio EPA has operated a program of biological surveys since the late 1970s. Their initial purpose was to provide an integrated set of biological and chemical data for use in monitoring and reporting activities and the water quality standards (WQS) program. An outgrowth of this effort has been the development of biological criteria ('biocriteria") as an ambient aquatic life use goal assessment tool. Biocriteria were recently adopted (February 1990) as a part of the Ohio WQS regulations. Concepts important to this approach include a practical definition of biological integrity, the role of ecoregions, the regional reference site approach, and recognizing the characteristics inherent to chemical assessment ("bottom up" approach) and biocriteria ("top down" orientation). These are important concepts in the development and application of biocriteria. Initiating and implementing a biocriteria program requires that several initial decisions be made. These include how to incorporate biocriteria into the existing structure of the WQS regulations, selection of appropriate organism groups, selection of evaluation tools, selection of reference sites, and regionalization considerations. All of these affect how well biocriteria work in a state water quality management program.

Biological field sampling procedures are also summarized with cost and resource requirements. The Index of Biotic Integrity (IBI), modified for application in Ohio, and the Invertebrate Community Index (ICI) are two of the principal evaluation tools used by Ohio EPA. The derivation, calibration, and variability of each is described. Current program uses of biocriteria include water quality standards (use designations, use attainability analysis), NPDES permitting, State Revolving Loan Fund, basic monitoring/reporting (e.g. 305b report), nonpoint source assessment, enforcement/litigation, 404/401 dredge and fill issues, and CSO/stormwater management. An emerging area of use is with Natural Resource Damage Assessments. Examples of biocriteria application are illustrated and include stream specific assessment, trend reporting and assessment, and providing information about rare and endangered species.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Water Quality Indicators for Rivers and Streams: Selection, Stratification and Aggregation for Decisionmaking

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Current state and national assessments of water quality status and trends (the state and national reports pursuant to Section 305(b) of the Clean Water Act are perceived by many to be less than adequate for important management and information purposes. This stems from use of inappropriate measures of status and trends that are inconsistently reported and summarized. Consistent stratification of meaningful indicators of water quality status coupled with standardized methods to aggregate assessment information can greatly enhance the utility of our reporting mechanisms. An aggregation method is developed using four important indicators pertinent to water quality problems in rivers and streams: (1) severity, (2) extent, (3) trend, and (4) recovery potential.

A consistent scheme for reporting problem severity is presented allowing use of ecological (in-stream biological survey) data, chemical water column data, toxicity testing information, or risk estimates for consumption of contaminated fish or water supplies. Problem extent for rivers and streams incorporates both flow and segment length. Each indicator is stratified with values corresponding to each level. The logic for the stratification scheme and values is described. Options for formal development of values are discussed. The aggregation method (with initial straw values developed by the author) is applied to the Deep River system (North Carolina) demonstrating its utility for objective quantification of water quality status, for measuring water quality improvement over time and for illustrating control program effectiveness. Conclusions and potential applications are discussed. These include: (1) coherent summarization of status and trends for a wide range of geographic levels of resolution, (2) better targeting of priority problems, and (3) easier evaluation of effectiveness of control programs.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Aquatic Macroinvertebrates as Biological Indicators of Water Pollution in Arizona

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The Arizona Department of Environmental Quality, in cooperation with the University of Arizona, is reviewing a variety of biological assessment methods to enhance Arizona's surface water quality monitoring program. Very little data regarding water quality and its relationship to aquatic macroinvertebrate communities in arid regions are available. This information will be necessary for future management of Arizona's surface water resources. A semi-quantitative approach of macroinvertebrate sampling (U.S. EPA Rapid Bioassessment Protocols) is currently being tested at 6 sites on the Verde and Santa Cruz Rivers. Physical, chemical and biological data have been collected during spring and summer 1990 to characterize conditions in streams that are either minimally or moderately impacted by anthropogenic activities. Species of macroinvertebrates collected during spring represented 8 classes (Insecta, Crustacea, Gastropoda, Bivalvia, Oligochaeta, Hirudinea, Turbellaria) including 17 orders of aquatic macroinvertebrates. Aquatic insects (immature and mature forms) dominate these communities with representatives of 29 families. Aquatic communities from Verde River collection sites appeared similar whereas those sampled from the Santa Cruz River demonstrated lower diversities (family level) in the effluent dominated sites than the control site. Species typically tolerant of low dissolved oxygen and mediocre habitat (Chironominae) dominated the fauna at effluent dominated sites. The data, while providing valuable aquatic community ecology information, will be useful for the implementation of programs consistent with U.S. EPA guidelines for biological standards and monitoring mandated by the 1987 Clean Water Act.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Development of Diagnostic Procedures to Evaluate Aquatic Resources In Regional Watersheds

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The U.S. EPA laboratory at Duluth, Minn. has been participating in regional water quality studies within the states of Illinois, Minnesota, and Michigan. Locations are in the Upper Illinois, Minnesota and Saginaw river basins. The objectives are to develop diagnostic procedures to identify impacts and assist the sponsoring agency in the integration of this information into a regional goal setting process. Our overall project goal with these regional studies is to determine biological and hydrological linkages that transcend geographical boundaries and serve as guidelines for defining watershed health. The example presented is the Minnesota River study being coordinated by the Minnesota Pollution Control Agency and conducted by a multiagency task force composed of state, university, and federal participants. The general approach is to evaluate a variety of procedures to define watershed health. The physical procedures measure habitat quality, and the chemical and laboratory toxicity tests measure the quality of the ambient surface and sediments. The biosurveys (with fish and macroinvertebrates) define the present status of instream biota and reveal the severity of degradation. All procedures assist in defining stressors and aid in the categorization of priority reaches needed for further control measures by regulatory authorities.

If you would like further details on this subject matter, please feel free to contact the participant; addresses can be found in the Attendees List starting on page 163 of this document.

Biological Criteria: Research and Regulation

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