



**Region IV Workshop on
Biomonitoring and Biocriteria**

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This report reflects the advice and opinion of the participants, not necessarily EPA or state policies on any subject. It is hoped that the results of this workshop will provide a helpful perspective as we begin the process of biocriteria policy and program development. Please refer any questions, thoughts, or comments on this report to

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EXECUTIVE SUMMARY

This report summarizes the proceedings of the Region IV Biocriteria Workshop held in Athens, GA, March 7-9, 1989. The biocriteria workshop was sponsored by the U.S. Environmental Protection Agency Region IV Water Quality Management Branch and Ecological Support Branch in response to the 1987 Office of Water report, *Surface Water Monitoring: A Framework for Change*. The report called for increased ambient biomonitoring and an improved framework for monitoring, assessment, and reporting. The primary purpose of the workshop was to provide support and assistance to the region's eight states in developing specific narrative and numerical biocriteria that are consistent with Section 304(a)(8) of the Clean Water Act. Workshop participants included state water quality biologists who represent each of the region's eight state environmental regulatory agencies, a Tennessee Valley Authority biologist, academics and consultants specializing in ecology and aquatic resources, and EPA scientists and administrators from Region IV. The workshop emphasized free-flowing, wadeable streams and rivers. The primary objectives of the workshop were

- to survey the biomonitoring programs of Region IV states
- to briefly compare approaches and methods of specific components of biomonitoring methods and biocriteria programs
- to recommend consensus approaches and methods to improve comparability between programs in the Region
- to identify the primary resource and research needs for the states in Region IV

Aquatic ecosystems are exceedingly complex and dynamic. To assess them rationally, an array of biological metrics that conveys information about their structural and functional attributes must be developed. The past 10 years of aquatic ecosystems research and assessments indicate a need to change from the chemical-specific toxicological approach alone to an integrated approach that fully incorporates biological surveys, biocriteria, and physical habitat. Assessing ambient biological conditions has become possible because sampling tools and protocols have been markedly improved, tested, and refined. Thus, many issues that limited the use of biocriteria and biomonitoring are no longer relevant. Also more tractable definitions of biological integrity and improved analytical tools to assess this integrity have standardized sampling and analyses.

Conclusions

- Biocriteria, combined with determinations of physical habitat, chemical concentration, and toxicity testing, provide an integrated, conceptually valid approach for assessing aquatic resources. Biocriteria are especially useful for determining whether other established criteria are protecting aquatic life.
- Biocriteria can quantitatively measure the interim goals of restoring and maintaining biological integrity as mandated in Section 101(a)(2) of the Water Quality Act.
- Biocriteria should be established on an ecoregion basis because each ecoregion has a unique biotic potential. However, the goal of biocriteria, that is maintaining biological integrity in a water resource system, should not be altered within ecoregions unless a site's natural habitat differs substantially from the reference.
- A sound biomonitoring program should include benthos, fish, and aquatic plant life monitoring capabilities.
- Despite limited resources, several Region IV states have successfully implemented initial phases of biocriteria.
- Progress in developing biocriteria has been hampered by a lack of resources and will continue to be severely restricted without additional resources.

Recommendations

- Ideally, states should have numeric biocriteria in place within 5 years. Narrative criteria with an implementation plan are appropriate interim steps. Region IV assistance is essential to achieving biocriteria development goals.
- States should establish several relatively unimpacted reference sites per ecoregion. These sites should be tested for seasonality, size, and intra-regional variability. Reference sites should be incorporated into monitoring programs.
- A series of field validations, sponsored by Region IV, should be held to provide assistance for training and methods comparisons.
- A menu of sampling methods should be developed for the Region IV states. The menu will allow each state to choose appropriate methods based on its data needs and resources. The sampling methods must be rigorously defined, standardized, and documented in a methods manual.
- Habitat evaluations should be standardized using numerical descriptors. Habitat analysis should include watershed factors.
- Region IV states should define a set of metrics that allows a rigorous, statistically defensible sampling and data analysis program.
- A quality assurance/quality control (QA/QC) program must be developed and implemented. This program should include standardized biologist training and evaluation of sampling and analysis methods.

- Database management requirements are large and are not yet being met. Significant resources are required for computer systems, software, data entry, and data analysis.
- EPA Region IV should develop a regional policy supporting biomonitoring and biocriteria development. The policy should incorporate individual state resources and needs.

1. INTRODUCTION

This report summarizes the proceedings of the U.S. Environmental Protection Agency (EPA) Biocriteria Workshop held in Athens, GA, March 7-9, 1989. The EPA Region IV Biocriteria Workshop was sponsored by the Water Quality Management and the Ecological Support Branches at EPA Region IV in response to the 1987 Office of Water report, *Surface Water Monitoring: A Framework for Change*. This report called for increased ambient biomonitoring and an improved framework for monitoring, assessment, and reporting. The primary purpose of the workshop was to support and assist the region's eight states in developing specific narrative and numerical biocriteria that are consistent with the mandate stated in Section 304(a)(8) of the Clean Water Act (CWA). The workshop also supported the Water Quality Act (WQA) of 1987, Section 303(c)(2)(B), requirements for EPA to develop criteria based on biological assessment methods when numerical criteria have not been established for the priority pollutants listed in Section 307(a) of the CWA. Biocriteria regulations can provide a quantitative measure of the interim goals (i.e., restoring and maintaining biological integrity) of Section 101 of the WQA.

Biocriteria and biomonitoring data will also improve the implementation of other sections of the CWA:

- methods of measuring the effects of pollutants on biological integrity, *Section 105*
- guidelines for evaluating Nonpoint Sources (NPS), *Section 304(f)*
- lists of waters unable to support balanced biological communities, *Section 304(l)*
- biennial reports of the extent to which waters support balanced aquatic communities, *Section 305(b)*
- assessments of lake trophic status and trends, *Section 314*
- lists of waters that cannot attain designated uses without additional NPS controls, *Section 319*
- prohibitions against dredge and fill disposal adversely affecting balanced wetland communities, *Section 404*
- reclassification of water resources (e.g., outstanding resource water)

All Region IV states conduct biomonitoring to provide information to water quality regulatory programs, and many state programs have accumulated enough practical experience and technical

expertise to take the lead in developing biocriteria. With this in mind, the Region IV Biocriteria Workshop was designed with four primary objectives:

- to survey the biomonitoring programs of the Region IV states
- to briefly compare methods and approaches used by Region IV states in their biomonitoring and biocriteria programs
- to recommend consensus methods and approaches to improve the comparability between programs in the region
- to identify the primary resource and research needs for the states in Region IV

This information will enable EPA Region IV to assume the lead in providing the states with technical and programmatic guidelines for developing biocriteria.

Workshop participants included state water quality biologists who represented each of the region's eight state environmental regulatory agencies, a Tennessee Valley Authority biologist, academics and consultants specializing in ecology and aquatic resources, and EPA scientists and administrators from Region IV. A list of workshop participants is included as Appendix A.

The workshop consisted of two parts: presentations by individual speakers (see Table 1.1) and group discussion. The second component of the workshop involved small discussion groups where participants addressed issues relative to reference sites, sampling methods, habitat evaluation, data analysis and database management, and quality assurance/quality control (QA/QC) programs. When appropriate, consensus positions were defined and/or recommendations were made. The groups also discussed key steps in developing biocriteria. The workshop emphasized free-flowing, wadeable streams and rivers.

Table 1.1 Workshop Presentations

Name	Topic
Mr. Mike McGhee	Welcome
Mr. Fritz Wagener	Water quality standard perspective
Dr. James Karr	Need for biological monitoring; advantages and disadvantages of various approaches for assessing biological integrity
Dr. Robert Hughes	Use of the ecoregion approach and the emerging strategic triad concept in water quality
Dr. Richard Wiegart	Ecological research initiatives
Region IV states	Status of their biomonitoring and biocriteria programs

2. OVERVIEW¹

Biomonitoring data and biocriteria should be used in conjunction with chemical water quality assessments and chemical-specific criteria. This concept is the strategic triad for protection of water quality. Biomonitoring data and biocriteria directly define and measure instream biological integrity and must be fully integrated into surface water programs. Additionally, habit assessment should be incorporated to allow detection of significant non-toxic stressors. Biomonitoring data are generally used in a reactive manner to detect inadequacies in current controls. In contrast, chemical-specific criteria are a proactive tool used, for example, to set limits for discharge permits. These proactive and reactive functions form an important balance in the regulatory process.

Biomonitoring provides a valuable set of tools to directly determine whether the objective of restoring and maintaining the biological integrity of the nation's waters (Section 101 of the 1987 WQA) has been met. The past 10 years of aquatic ecosystems research and assessments indicate a need to change from the chemical-specific/toxicological approach alone to an integrated approach that fully incorporates the strategic triad. Assessing ambient biological conditions has become more reliable because sampling tools and protocols have been markedly improved, tested, and refined. Thus, many of the issues that limited the use of biocriteria and biomonitoring are no longer relevant. Also, more tractable definitions of biological integrity and improved analytical tools to assess this integrity have standardized sampling and analyses.

Historically, the EPA and the states have developed a variety of standards to achieve the goals of the 1987 WQA. These standards specify beneficial uses for waterbodies and criteria to evaluate whether the uses are being attained (and thus, whether the standards are being met). Most of these criteria have followed the traditional approach of developing chemical-specific limits deemed to be protective for the designated uses. These limits were usually established by performing single chemical toxicity tests on test organisms. More recently, whole-effluent testing has been added to the array of assessment tools.

This approach to water quality has produced significant improvement in the nation's waters. Laboratory-derived criteria are particularly appropriate for designated uses related to human activities such as drinking water or human contact (swimming, water skiing). Chemical-specific

¹ From U.S. Environmental Protection Agency, 1987, Pages 2-3 Reprinted with changes

criteria and subsequent water quality sampling are essential to establish effluent limitations in the National Pollutant Discharge Elimination System (NPDES) permits program. The chemical-specific criteria approach also has been applied to protecting designated aquatic life uses. However, an increasing body of evidence indicates that regulatory agencies cannot solely rely on these criteria without also sampling the ambient biological communities to verify the results of pollution control measures and to detect non-toxic stressors.

The existing criteria are insufficient to protect aquatic life uses for two main reasons. First, they are derived from laboratory-based simulations, which cannot address all factors affecting resident aquatic communities (e.g., habitat limitations, additive impacts of multiple dischargers). Thus, they are only surrogates for achieving desired results. Secondly, chemical-specific criteria cannot always address potential impacts from habitat modifications and many nontoxic pollutants, particularly those from nonpoint sources such as sediments and nutrients, which affect a vast majority of river miles. These issues are discussed in more detail in the 1987 Office of Water report, *Surface Water Monitoring: A Framework for Change*.

EPA and the states have previously been discouraged by actual and perceived problems in sampling and evaluating the health of the ambient aquatic life, which made them hesitant to use this information in regulation. Natural communities were thought to be too variable and too complex to be precisely and consistently measured. As long as these beliefs prevailed, regulators could not justify using biological criteria in the same way as chemical criteria. Indices (such as species diversity) that were intended to reduce this complexity proved unreliable; there was considerable debate about sampling methods (what kinds, how extensive or intensive, and where to sample) and which organisms to sample. Finally, the costs of sampling and evaluating ambient biological conditions were considered too high for the level of information obtained.

One result of recent improvements is that expensive, intensive biological surveys of all organisms over long periods are not necessary for many regulatory purposes. Research in field methods has shown that reliable data, useful for monitoring, can be collected without excessive expense. This makes biomonitoring data especially valuable as a screening tool, while reserving more expensive investigatory techniques for more complex situations, such as where a high probability of litigation exists.

3. CURRENT STATUS OF STATE PROGRAMS IN REGION IV

Each state presented a synopsis of its biomonitoring and biocriteria activities. The areas of interest included a brief review of biocriteria regulations and use of biomonitoring information, reference streams, ecoregions, sampling methods, habitat evaluation, data interpretation (metrics), database management, and QA/QC programs. The objective of the state presentations on the status of their biomonitoring/biocriteria programs was to gain insight into specific activities that are critical to implementation of biocriteria into water quality standards. These presentations varied considerably in format and content, reflecting the substantial differences between states in both approach and status of their programs.

3.1 Biocriteria Regulations and Use of Biomonitoring Information

Each state in Region IV is conducting some form of biomonitoring; however, the extent, nature, and the manner in which the information is used varies considerably between states. Four states (Kentucky, Alabama, South Carolina, and Georgia) have a general narrative statement in their water quality regulations requiring the protection of aquatic life. Two other states (Florida and North Carolina) use biological criteria to protect biological integrity of aquatic resources.

Florida is the only Region IV state with a numerical biocriteria regulation. Florida's biocriteria regulation, which has been in place since 1975, defines biological integrity as the maintenance of $\geq 75\%$ of the established background diversity (as measured by the Shannon-Weaver Diversity Index). The Florida rule mandates the type of sampling, the number of samples, and the specific metric to be applied. There are several concerns with this approach (e.g., the reliability of the Shannon-Weaver Index as an effective indicator of biological integrity and the inability to adopt improved sampling and data analysis methods). Florida makes extensive use of its biocriteria information in all facets of its water quality program.

Kentucky, Tennessee, and Alabama biomonitoring teams file reports to their water quality divisions. In Alabama, biological information is used in conjunction with water quality demonstration studies, standards review, ambient monitoring and special problems monitoring. In Kentucky, South Carolina, and Tennessee biological data is used for trend monitoring, reference data, the biennial 305(b) report to Congress on Water Quality, and Health Advisories for fish consumption. Kentucky, South Carolina, and Tennessee also have an intensive survey program that collects biological, physical, chemical, and sediment data for stream use designations and investigation of the effects of

point source discharges and nonpoint source run-off. North Carolina appears to make the most extensive use of narrative biomonitoring information in Region IV. North Carolina has had defacto biological criteria in place for three ecoregions since 1983. This state's approach to biocriteria is flexible, can be altered as natural changes occur or improved methods are developed, and relies on the expert judgment of experienced personnel. North Carolina's use of narrative biocriteria information includes

- Investigations of Point Source Dischargers
 - Special Studies, Complaints
 - Toxicity Reduction Surveys
 - "Before and After" surveys
- Investigations of Nonpoint Source Runoff
 - Test of Management Strategies
- Use of Attainability Designations
 - Water Use Reclassifications
 - Outstanding Resource Water
 - High Quality Water (Proposed)
- Basin Assessment
- Trend Analysis (BMAN)

3.2 Reference Sites

North Carolina has selected at least one reference site for each of its three defined ecoregions, based primarily on landuse. Mississippi is establishing a reference site for each of its five defined ecoregions and has historical monitoring on 22 stream sites. However, many of these streams are impacted, particularly within the Delta. Kentucky has proposed a reference reach program; specific sites are under consideration. Alabama, Georgia, and Florida have not yet established independent reference sites. In 1983, Tennessee sampled 20 unimpacted reference sites seasonally (twice) for benthos, fish, and algae. Alabama has several candidate sites where the Alabama Geological Survey has conducted intensive biomonitoring. Georgia has 10-15 candidate reference sites; the state anticipates selecting a site for each ecoregion by 1994. The states in Region IV have not clearly defined their criteria for reference sites and have not incorporated the biomonitoring results into their biocriteria programs.

3.3 Ecoregions

The ecoregion concept received enthusiastic support from the state biologists in Region IV. Three states (North Carolina, Kentucky, and Georgia) have adopted some form of ecoregions and assigned drainage basins to the appropriate ecoregion. Because the states do not share the same criteria or system of defining ecoregions, comparisons or cooperation across state lines is difficult. The states that have assigned ecoregion classifications have not defined the biotic potential for biocriteria

standards. The remaining states have not yet incorporated this approach into their biomonitoring programs.

3.4 Sampling Methods.

Most biological data bases represent a compromise between resources expended and information gained. This compromise is one factor that has led to a fairly wide variety of state biomonitoring programs. Another factor is the ultimate use of the data. Regardless of the use of the data, all states have developed standard field methods and quality assurance to ensure consistent and defensible results. It is essential that biomonitoring is performed with the same rigor as procedures used to monitor chemicals and test for toxicity.

All Region IV states sample macroinvertebrates. Artificial substrate samplers are used by all the states except North Carolina and Mississippi, which is currently replacing them with natural substrate samplers (Table 3.1). Kick sampling is conducted by Alabama, Kentucky, North Carolina, South Carolina, and Tennessee, and selected picks are conducted by Alabama, Kentucky, Mississippi, North Carolina, South Carolina, and Tennessee. Sweep nets are used to collect samples in Mississippi, North Carolina and Tennessee, and leaf packs are used by Mississippi, North Carolina, South Carolina, and Tennessee. The other methods employed (epifauna samplers, sand samplers, surber samplers, timed streamside sampling, and limestone baskets) are each used by only one or two states.

Identifying specimens from any of the collection methods is time consuming, and states use different metrics to evaluate data according to their needs and resources (Table 3.2). Some of the more common metrics were EPT richness, total taxa richness, the Biotic Index (used to assess organic pollution), abundance, indicator groups, equitability, and the Shannon-Weaver index. The other metrics listed in Table 3.2 are used by only one or two states.

Fish are collected by North Carolina and Kentucky water quality agencies as part of their biomonitoring program. In Florida and Georgia a different state agency collects fish. Most states focus on game fish. Although fish require a greater field effort to collect than macroinvertebrates, they can be identified to a relatively low taxonomic level in the field, and data are quickly available for analyses. Electrofishing, seining, and trammel/gill nets are used to collect fish. Kentucky and North Carolina calculate the Index of Biotic Integrity (IBI) for fish. North Carolina also determines fish density, reproductive success, and taxa richness. Most of the states monitor toxics in fish tissue in conjunction with biomonitoring.

Georgia, Kentucky, Mississippi, North Carolina, South Carolina, and Tennessee collect algae data. Georgia, Kentucky, Mississippi, South Carolina, and Tennessee use periphytometers to collect

Table 3.1 Methods for Collecting Biomonitoring Samples

Organism Group	Method	States							
		AL	FL ^a	GA ^a	KY	MS	NC	SC	TN
Macroinvertebrates									
Fine Mesh Samplers									
	Epifauna samplers						x		
	Sand samplers						x		
Artificial Substrate Samplers									
	Limestone baskets			x					
	Multiple plate samplers	x	x	x	x	x		x	x
Disturbance Samplers									
	Sweep nets					x	x		x
	Kick samples	x			x		x	x	x
	Surber samplers								x

Table 3.1 Continued.

Organism Group	Method	States							
		AL	FL ^a	GA ^a	KY	MS	NC	SC	TN
Other	Leaf packs					x	x	x	x
	Selected pick	x			x	x	x	x	x
	Timed, streamside sampling			x				x	
Algae	Periphytometers			x	x	x		x	x
	Substrate samplers				x			x	
	Water sample-plankton			x	x		x	x	
	Natural substrate				x				
Fish	Electrofishing	x			x		x	x	x
	Seine				x		x	x	
	Trammel/gill nets				x		x	x	x

- ^a A different state agency has responsibility for fish collections in Florida and Georgia

Table 3.2 Metrics Used by States in Region IV for Analysis of Biological Data and Data Management

Organism Group	Method	States							
		AL	FL ^a	GA ^a	KY	MS	NC	SC	TN
Macroinvertebrates									
Taxa Richness									
	Chironomid richness					x			
	EPT/Chironomid ratio					x			
	EPT richness	x			x	x	x	x	
	% change in total taxa richness						x	x	
	Total taxa richness	x		x	x	x	x	x	x
Biotic Index									
	Biotic index		x			x	x		
	Indicator Assemblage Index (IAI)	x							
	Indicator groups	x					x	x	x
Diversity Related									
	Abundance	x			x		x	x	x
	Equitability	x		x				x	x
	Shannon-Weaver diversity	x	x	x		x		x	x
	% change in abundance						x	x	
	% dominants	x							
	Dominants in common						x		
	Index of Community Integrity (ICI)					x			

Table 3.2 Continued.

Organism Group	Method	States							
		AL	FL ^a	GA ^a	KY	MS	NC	SC	TN
Other	Morphology						x		
	% similarity				x			x	
	Functional feeding groups				x	x			
Algae	Biovolume						x		
	Chlorophyll <u>a</u>			x	x	x	x	x	x
	% community similarity				x				
	Species present						x	x	
	Taxa richness				x				
	Density						x	x	
	Biomass								x
Fish	Density						x		
	Index of Biotic Integrity (IBI)				x		x		
	Reproductive success						x		
	Taxa richness				x		x		
	Toxics burden				x	x	x	x	x

Table 3.2 Continued.

Organism Group	Method	States							
		AL	FL*	GA*	KY	MS	NC	SC	TN
Data Management	Paper files	x	x	x	x	x	x	x	x
	Personal computers	x	x		x	x	x	x	
	Main frame				x		x	x	

- * A different state agency has responsibility for fish collections in Florida and Georgia.

attached algae, and Georgia, Kentucky, North Carolina, and South Carolina collect water samples (lakes only) for determinations of planktonic algae. Georgia, Kentucky, Mississippi, North Carolina, South Carolina, and Tennessee determine chlorophyll *a* as an estimate of biomass. Kentucky also determines community similarity and taxa richness of the algae, and North Carolina and South Carolina determine density and species present. North Carolina also determines biovolume.

3.5 Habitat Evaluation

All states in Region IV collect physical data and evaluate habitats as part of routine surveys. The format and intensity of these surveys vary considerably between the states. Several states take only rudimentary notes on the sample site. No state has developed a rigorous numerical format to allow precise comparisons between sample sites or to eliminate confounding factors for data interpretation. Understanding the inseparable link between the watershed and stream assists in addressing nonpoint and nonchemical degradation of the biological community. To accomplish this, habitat evaluations must look beyond instream and bankside parameters. The southeastern states attempt to characterize the landuse or other factors in watersheds surrounding sample sites when appropriate.

3.6 Data Management and Application

Data management and analysis capabilities are highly varied among the states. All have data in paper files (Table 3.2), and all would like to have these data computerized in workable software packages. In Region IV states, biomonitoring data are used to support use attainability designations, or for permitting compliance. Florida and North Carolina have computerized (PC and mainframe) data bases and also have the most developed biocriteria. These states also use the data to detect trends; to determine nonpoint source effects; to make basin assessments; to identify outstanding natural resource waters, nutrient sensitive waters, or eutrophication; or to test management strategies.

3.7 Quality Assurance/Quality Control

The states all use some EPA sampling methods and procedures as defined by the *Biological Field and Laboratory Methods*. All states agree that the manual needs to be updated by EPA Region IV to account for improvements in sampling methodology and data analysis techniques. No state has a formal training and certification program for field biologists.

3.8 EPA Technical Support

A consistent theme in each presentation was that these states need guidance and support from EPA to implement biocriteria. EPA should prepare a technical support document for developing

biocriteria and using ambient biological sampling in surface water programs. The document should address a variety of topics including greater resolution of ecoregions, managing data, developing software, and establishing a menu from which the states can pick methods for stream and habitat evaluations. States need flexibility in choosing methods and approaches suitable to their needs; however, those methods need to be as sophisticated and reliable as physico/chemical monitoring. EPA should support the development, evaluation, implementation of refined-use designations and numerical biocriteria by the states. These numerical criteria should be used to translate narrative criteria for protecting aquatic life uses into more quantifiable measures of attainment.

4. PREFERRED OPTIONS AND PRACTICAL RECOMMENDATIONS

Each of the two workshop discussion groups was charged with the task of defining and assessing key concepts and issues in biocriteria. Each group attempted to define key concepts relative to reference sites, sampling methods and timing, habitat evaluation, and data analysis and management. Each issue was examined in detail and, where appropriate, a consensus was reached.

4.1 Biocriteria

The goal of biocriteria is to protect water resources through evaluation of biological integrity. Combined with determinations of chemical concentration/toxicity testing, physical habitat assessments, and biocriteria form the strategic triad for protecting aquatic resources. The strategic triad provides an integrated, conceptually valid approach to assessing aquatic resources. The criteria can be either narrative or numerical.

Biocriteria narrative standards are in place in Arkansas (Shakelford 1988), Maine, and Ohio (Ohio EPA 1987) and have been proposed in North Carolina. The state of Maine's approach has been to set narrative standards in the law that establish general characteristics necessary for a biological community to attain a given classification and a given level of integrity (Courtemanch et al. 1989). The narrative criteria are basically a refined use classification. Although narrative criteria provide a basis of protection for aquatic resources, the workshop consensus was that the ideal situation would be an ecoregion-specific numeric standard. The numeric biocriteria need to allow for flexibility and account for variability. Ohio has proposed numerical biocriteria, and several other states (Arizona, Idaho, Nebraska, and Wisconsin) are developing them.

Even though the idealistic goal of biocriteria is to establish numeric criteria, initial biocriteria will rely on narrative. The narrative criteria should have support from documents, have a methodology in place, and be open for review. Regardless of whether a biocriterion is numeric or narrative criteria, it must be defensible in a court of law.

4.2 Ecoregions

The ecoregion concept, which groups naturally similar ecosystems into regions that are substantially less diverse than the entire nation or a state, is critical to the successful development and application of biocriteria. Sites stratified by ecoregion have similar ecological potentials that can be quantified with known levels of precision (Hughes and Larsen 1988). Omernik (1987) developed an ecoregion

map of the southeastern United States. These ecoregions incorporate less ecosystem variation than an entire state or major river basin, and they are more widely applicable than ecological regions defined by a single characteristic or purpose.

Workshop participants agreed that biocriteria should be established on an ecoregion basis. They also agreed that Omernik's defined ecoregions were a starting point, but that in some cases the ecoregions would need to be refined to minimize natural variability for between-system comparisons. The participants also noted the potential problem of ecoregions overlapping political boundaries; therefore, a procedure must be established to facilitate interstate cooperation. Interstate cooperation will be required to define ecoregions and should be encouraged in biomonitoring and biocriteria development.

4.3 Reference Sites

There are two distinct types of reference sites: site-specific and regional. The site-specific site is the traditional upstream sample point that is used as a reference for suspected point discharges. State standards may require that a demonstrable difference not exist between upstream (reference) and downstream areas. A regional reference site serves as a relatively unimpacted benchmark for disturbed streams in a given ecoregion (Hughes et al. 1986). Regional reference sites are required as a reference for nonpoint source discharges.

Reference sites should mimic, as closely as possible, the natural characteristics of the sites of concern. The reference sites should be in the same ecoregion and have similar types of biological and chemical data collected. They should have similar stream morphology, stream order, and watershed size/discharge, and should be minimally impacted. This will be difficult where nonpoint pollution, including atmospheric deposition, is a problem. Under ideal circumstances, there should be a minimum of three size classifications of reference sites within an ecoregion and at least six sites per size class to capture the degree of natural variability in the ecoregion.

When defining a reference site, the stream should be sampled at least two times per year, preferably four, for macroinvertebrates and one to two times per year (in late summer or autumn) for fish. Greater sampling frequency may be needed to detect the effects of seasons on transient, elusive, ephemeral, and rare species. Historical data should be used when available. However, resampling reference sites validates the program and assesses long-term climatic and landscape changes. Therefore, reference streams should be incorporated into a trend monitoring program.

If an ecoregion lacks suitable reference sites, the least impacted sites in the ecoregion may suffice. Historical data for a stream that was once suitable as a reference stream can also be used provided that methodologies are comparable.

Suitable reference sites may not be in the same state as the site of interest. Two options exist: (1) use an existing instate reference site or (2) interact with colleagues in the adjacent state. Although excellent communication and interactions now exist among Region IV biologists, communication needs to be improved to facilitate the exchange of information among states. This exchange could be achieved through memoranda of agreement, including delineation of higher resolution ecoregions and maintenance of a reference site data base by the states in EPA Region IV.

4.4 Sampling Issues

Fish, macroinvertebrates, and algae are all suitable taxa to monitor for establishing criteria.

Each state should use similar methods for biomonitoring and levels of species identification to maximize data comparability. Different methodologies are adapted to different water resource types. Rigorous identification and use of multiple metrics enhances our ability to make accurate decisions and, thus, provide greater protection for water resources shared by neighboring states. Comparable sampling methods also allow interstate comparisons of the limited number of reference sites, thereby maximizing the use of such data. Different sampling approaches (see Table 3.1) now used by the states should be standardized to increase data comparability on a per-unit basis. A suite of methods should be chosen based on sound scientific criteria; once established, sampling methodologies should remain in place. When a change in sampling strategies is considered, the alternate methods should be compared to allow conversions between older and newer methods.

A menu of methods needs to be developed for the Region. Workshop participants recommended that the EPA sponsor field studies to examine method comparability and efficiency of each method for use in Region IV states. When the studies are complete, a methods workshop for Region IV states should be conducted. Methods currently used by the states in Region IV are summarized in Table 3.1.

The type of sampling conducted depends on the metrics examined and the degree of resolution required for making decisions. Each ecoregion may have a different set of metrics with different expectations. In Ohio, for example, there are different expectations for different sized streams and different ecoregions. Metrics differ by stream size, but are the same statewide. In Region IV, metrics and expectations could be developed as they were in Ohio, or states could lump all metrics together and then determine different metrics for different ecoregions.

Quantitative sampling is usually employed if an estimate of organisms per unit effort or space is required. Qualitative sampling is conducted to provide an inventory (e.g., presence/absence, abundant/common/rare) of organisms. Both yield usable data; which data are used -- hence the method employed -- depends on the data objectives. The majority of Region IV states use qualitative, multi-habitat collection techniques for macroinvertebrates. Appropriate statistics should be applied to the data derived from these methods to ensure that comparisons can confidently be made. However, quantitative data tractable to statistics will be required for long-term trend monitoring and regulatory purposes.

Workshop participants agreed that the practice of using rapid bioassessment protocols and diversity indices needs to be re-examined. Data analysis methods are sensitive to sample size, thus limiting the usefulness of some rapid bioassessment protocols in a biocriteria program. Some of the rapid bioassessment protocols are appropriate for biocriteria and should be included in the Region IV method comparison study. However, *rapid* should be omitted from the method name. Additionally, the methods need to be redefined.

Diversity indices, once widely used, have largely been supplanted. Although they work under select conditions, they are not a valid parameter for biotic criteria.

A key component of sampling is defining and stratifying inherent natural variability of stream ecosystems. Natural variability and ways to stratify it must be taken into consideration when establishing biocriteria that are defensible in a court of law. Natural variation in the biota of streams is both spatial and temporal. Spatial variability can be reduced by factoring stream morphology, stream size, habitat, and ecoregion into study designs. Ohio's biocriteria program attempts to reduce spatial variation by incorporating watershed area and ecoregion in its experimental design. Temporal variability can be reduced by incorporating stream physical variables (e.g., stable flow, seasonal temperature) and biological attributes of organisms (e.g., longer lived species, emphasis on structural and functional properties of assemblages) in the study design.

The use of multiple metrics, reference sites, and ecoregions can reduce the effect of natural variation. Expectations for the biota in a stream are determined from reference sites, previous experience, or the literature. Sampling, at comparable stream conditions and season, is conducted to determine whether expectations are met. IBI and Invertebrate Community Index (ICI) are examples of measures that reduce variability.

Biological trend monitoring can provide a baseline for establishing biological criteria. Historical insights into the biota of a stream, or in a region, are invaluable.

4.5 Habitat Evaluation

Habitat evaluations should be an integral part of biomonitoring. Evaluation of habitat quality is important because differences in habitats between sites may confound interpretation of sampling data. In addition, changes in habitat at a particular site may be the major stress on the biological community.

Each state should develop a standard habitat evaluation procedure (possibly with ecoregion specific metrics and scoring criteria) using numerical descriptors for various parameters. Interstate and Region IV cooperation will assist development of the evaluation procedures. Developing a methods manual to provide guidance on procedures would be beneficial. Existing habitat evaluation procedures, developed by the U S. Fish and Wildlife Service, U S. Forest Service, EPA and several state agencies, may serve as models. A field workshop for methods comparison and development is critical to ensure comparable results ecoregion wide.

4.6 Data Analysis and Data Base Management

Metrics need to be tailored to the state using them and to the state's objectives. In Ohio, for example, IBI metric expectations were developed on a statewide basis. This provides the state with data that meets biomonitoring needs and allows for comparisons across the state. The data are used to make decisions for permitting and use/attainability determinations. Region IV states need to define a set of metrics, as Ohio has done, that allow a rigorous, statistically defensible sampling and data analysis program. The metrics should measure shifts in the structural and functional composition of aquatic communities from conditions expected in unimpacted or minimally impacted regional reference sites.

To enhance our ability to make water resource decisions and to report results, biomonitoring data must be organized in computerized data bases. Data in paper files are much less likely to be used than computerized data. Technology exists that will facilitate data transfers from paper to computer; however, resources to complete the data transfers do not yet exist. States in Region IV have widely varying capabilities in computer and data management systems. Those states that are more advanced in computerization could share techniques and knowledge with the other Region IV states. A regionwide common data management system would facilitate sharing data from ecoregions that cross state boundaries. Meetings of the Southeastern Water Pollution Biologists Association (SEWPBA) are one potential regionwide forum for discussion and technology transfer in this area.

4.7 Quality Assurance/Quality Control

Experienced and trained biologists as well as quality assurance and quality control procedures, insure the success of a biomonitoring program. An effective biomonitoring program will require eventual

certification of state biologists and consultants. Collection, analysis, and interpretation must be done by biologists experienced in techniques and ecology. Inexperienced personnel can be trained on reference streams where the biota and stream physical conditions are known. Personnel standards can be achieved through scientific society certification (e.g., the Ecological Society of America) or by establishing education, training, and experience requirements.

Quality Assurance/Quality Control is an important part of sampling. Scientifically sound sampling protocols are paramount to accurate, reliable data. States have standard operating procedures in place through EPA Region IV programs. Although these procedures are established, it is important to consider flow conditions, time of year, and time of day when designing sampling programs. Consistency in sampling is important; it can reduce sampling error and increase data comparability.

5. PROGRAM NEEDS

During the workshop, the states identified areas where they need assistance in program development. Biomonitoring is an active, ongoing program in the states; although the states do not require assistance for any new programs, they did name specific areas that they would like expanded in their existing programs. These areas are staff, data management, metrics determinations, sampling methods, ecoregion definition, communications and cooperation, and refinement of data interpretation techniques. At the conclusion of the workshop, there was a consensus among participants that a week-long SEWPBA field/laboratory workshop addressing these topics is necessary.

All states share the greatest need: to increase staff. Most states do not have the personnel necessary to quickly develop and implement biocriteria (Table 5.1). Another need common among the states is for increased data management. Computers and software are required to get data from the field and file cabinets into a computerized data management system. Manually compiled data are relatively

Table 5.1. Rating of Resources Necessary for Each State to Conduct Assessments to Establish Biocriteria^a

State	Metric			Data Management	Assistance from Other State Agencies
	Fish	Benthos	Plant		
Alabama	1	2	0	.5	1
Florida	0	4	4	4	4 (fish)
Georgia	1	2.5	.5	1	4 (fish)
Kentucky	3.5	3.5	2.5	1	3
Mississippi	.5	1.5	2.5	1.5	2
North Carolina	3	4	3	2.5	3
South Carolina	2	3	1.5	1	2
Tennessee	2	3	1	0	3

Scale of 0-5 was used: 0 = no resources available and 5 = all resources available

^a Two workgroups were surveyed; each number rating represents the average of both workgroups

inaccessible and are less likely to be used than computerized data. Additionally, historical data that yield insights into past water quality and biota may be effectively lost without computerization. Computerization will also improve the efficiency of data analysis and will provide the means to statistically analyze large data bases. Computerization is critical to the full implementation of biocriteria into the standards program.

Region IV states need to address how they intend to quantify features of the biological community they intend to examine. For example, are the states going to use multiple metrics of community structure separately (e.g., diversity, richness, and abundance) or combine individual metrics into a few index values (e.g., IBI; ICI; Fausch et al. 1989)? The IBI and ICI are gaining wider acceptance. A regionwide overview outlining the Region's approach must be developed. Once the general framework is established, several choices of metrics should be made available to each state; this concept was termed a metrics menu at the workshop. The metrics menu approach provides the necessary comparability at the EPA Region level, but allows flexibility for each state.

Sampling methods should be standardized across Region IV states. To allow for flexibility, however, there should be several standardized methods available for sampling the organisms chosen for monitoring. Each method should yield results that can be used for assessing biotic integrity. Several metrics, such as those listed in Table 3.2, are available for fish, macroinvertebrates, and algae. Ideally, a multiple metric assessment, such as IBI, will be used. This type of assessment should incorporate the following criteria:

- The measure must be biological.
- The measure must be interpretable at several trophic levels or provide a connection to other organisms not directly involved in monitoring.
- The measure must be sensitive to the environmental conditions being monitored.
- The response range of the measurement ought to be suitable for the intended application.
- The measure must be reproducible and precise within defined and acceptable limits for data collected over space and time.
- The variability of the measure must be low (Karr et al. 1986).

Differences among the state's resources make it appropriate for the states to exercise their own options in selecting methods. Flexibility will be a key factor in the success of Region IV biocriteria. Data comparability within the ecoregion (i.e., reference sites versus sample sites) and data defensibility are critical for implementation of biocriteria. Participants suggested that EPA update

the 1973 biological methods manual (EPA/670-4-⁷³31-001) and that the revised manual become the Region IV methods manual.

Ecoregions provide the geographical framework for biocriteria; therefore, determining ecoregion boundaries is critical to the program's success. Despite the publication of *Ecoregions of the Southeastern United States* (Omernik 1987), states and Region IV may need to refine such ecoregion delineations to provide a consistent, regionwide approach.

Because ecoregions cross several state boundaries, communication among the states in Region IV is critical to the success of biocriteria. Currently, communication at the biologist level is quite good (evidence SEWPBA); however, communication needs to be improved at the administrative level. Memoranda of agreement should be established among the states to facilitate interstate cooperation.

GLOSSARY¹

This section defines some of the terms used at the workshop. These are only working definitions, and are somewhat general; they are presented for purposes of clarification and continuity in this report. It is apparent from discussions at the workshop that many of these terms are used inconsistently or are misunderstood. The EPA should provide clear definitions for these terms and foster consistent usage.

AMBIENT (INSTREAM) BIOLOGICAL SAMPLING -- the process of collecting a representative portion of the organisms living in the waterbody of interest, to determine the characteristics of the lotic or lentic aquatic community. Fish, algae, and benthic macroinvertebrates are usually sampled. This term includes short- or long-term surveys and monitoring.

BIOASSAY -- the procedure of exposing test organisms, in a laboratory setting, to various concentrations of suspected toxicants or dilutions of whole effluent.

BIOASSESSMENT -- assessment of the condition of a waterbody using any available biological methods. Biomonitoring and bioassay are common bioassessment methods.

BIOCRITERIA -- the numerical or narrative expressions of the biological characteristics of ambient aquatic communities. (Often structural measures, e.g., species composition, organism abundance, and diversity). Biocriteria, as generally applied in state programs, are designed to reflect attainable characteristics under minimally impacted conditions. As such, biocriteria describe the ecological potential for aquatic community health in a given watershed, drainage basin, or ecological region.

BIOLOGICAL INTEGRITY -- a measure of the state of health in aquatic communities. A healthy aquatic community is a balanced community of organisms having a species composition, diversity and functional organization comparable to that found in natural (unimpaired) habitats in the region (Karr et al. 1986). Also called "Biotic Integrity."

BIOMONITORING -- monitoring conducted to ensure standards or effluent limitations are being met using either the ambient community or toxicity tests.

BIOSURVEILLANCE -- used synonymously with biomonitoring in this report. Also can be used to describe a series of systematic biomonitorings.

BIOSURVEY -- used synonymously with ambient biological sampling in this report.

CHEMICAL-SPECIFIC CRITERIA -- criteria that set specific allowable concentrations of individual chemicals in the water. These criteria are presumed to be protective of the designated aquatic life uses, as well as other uses, e.g., drinking water or human health ("swimmable" conditions).

CRITERIA -- the conditions presumed to support or protect the designated use(s), e.g., dissolved oxygen not less than 5 mg/L to protect a coldwater fishery use designation.

DESIGNATED USES -- the purposes or benefits to be derived from a waterbody, e.g., drinking water, aquatic life.

ECOREGIONS -- broad scale areas with a common ecological characteristic, e.g., Central Corn Belt Plains, Western Allegheny Plateau. Also called Ecological Regions.

IN SITU BIOASSAY -- assay conducted on test organisms, in the ambient water or discharge mixing zones, for known exposure periods, e.g., with caged fish or clams.

STANDARDS -- the legally established state rules consisting of two parts, designated uses and criteria.

TOXICITY TEST -- used synonymously with bioassay in this report.

WHOLE EFFLUENT TESTING -- a bioassay using the complete discharge "as it comes from the pipe," as opposed to separate bioassays on the individual component chemicals.

¹ From U.S. Environmental Protection Agency, 1987, pages iii-iv

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APPENDIX A

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