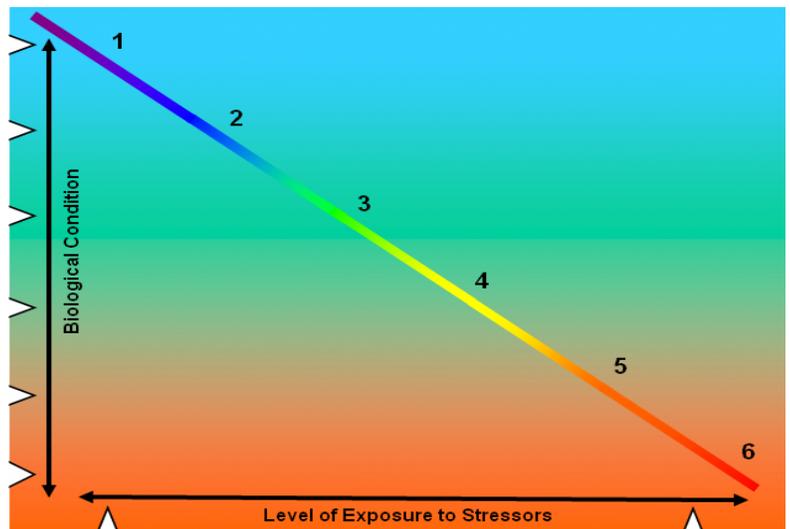


Calibration of the Biological Condition Gradient for High Gradient Streams of Connecticut



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CALIBRATION OF THE BIOLOGICAL CONDITION GRADIENT FOR HIGH GRADIENT STREAMS OF CONNECTICUT

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EXECUTIVE SUMMARY

In recent years, several states and the US EPA have developed a framework to support improved biological assessment. The framework, called Tiered Aquatic Life Use (TALU) supports development of tiered biological criteria in a state's water quality standards that can protect the best quality waters, that can be used as a tool to prevent or remediate cumulative, incremental degradation, and that can help to establish realistic management goals for impaired waters.

In recent years, several states and the US EPA have developed a framework to support improved biological assessment. The framework, called Tiered Aquatic Life Use (TALU), supports development of tiered biological criteria in a state's water quality standards that can protect the best quality waters, that can be used as a tool to prevent or remediate cumulative, incremental degradation, and that can help to establish realistic management goals for impaired waters. The basis of the TALU framework is recognition that biological condition of water bodies responds to aggregate human-caused disturbance and stress, and that the biological condition can be measured reliably. For TALU implementation, biological condition is measured on the Biological Condition Gradient (BCG), a universal measurement system or yardstick that is calibrated on a common scale for all states and regions.

This document describes the calibration of the BCG to high-gradient streams of Connecticut, which are routinely sampled in Connecticut DEP's monitoring program. The BCG includes decision criteria to assign streams to levels of the BCG, and thus it can be directly applied to tiered aquatic life uses in Connecticut's Criteria and Standards. The BCG is a more accurate and representative way to classify the condition of water bodies than previous methods, because its measurement standard is based on natural, undisturbed condition rather than a sliding scale of local conditions. Although it is intended to be a universal scale, it is not "one-size-fits-all" and takes into account natural classes and variability.

Description of the BCG

The Biological Condition Gradient is a conceptual model that describes changes in aquatic communities. It is consistent with ecological theory and has been verified by aquatic biologists throughout the US.

Specifically, the BCG describes how ten biological attributes of natural aquatic systems change in response to increasing pollution and disturbance. The ten attributes are in principle measurable, although several are not commonly measured in monitoring programs. The attributes are:

1. Historically documented, sensitive, long-lived or regionally endemic taxa
2. Sensitive and rare taxa
3. Sensitive but ubiquitous taxa
4. Taxa of intermediate tolerance
5. Tolerant taxa
6. Non-native taxa
7. Organism condition
8. Ecosystem functions

9. Spatial and temporal extent of detrimental effects
10. Ecosystem connectance

The gradient represented by the BCG has been divided into 6 BCG Levels of condition that biologists thought could be readily discerned in most areas of North America:

1. Natural or native condition
2. Minimal changes in structure of the biotic community and minimal changes in ecosystem function
3. Evident changes in structure of the biotic community and minimal changes in ecosystem function
4. Moderate changes in structure of the biotic community with minimal changes in ecosystem function
5. Major changes in structure of the biotic community and moderate changes in ecosystem function
6. Severe changes in structure of the biotic community and major loss of ecosystem function

The BCG and a multimetric index calibrated for Connecticut streams

This report summarizes the findings of a panel of aquatic biologists in Connecticut who applied and calibrated the general BCG model to benthic macroinvertebrate data from Connecticut streams. Data from Connecticut's monitoring program were examined to determine if the data were adequate to apply to the BCG. The panel was able to assign species in the database to the first five attributes listed above, and the panel assigned a set of test sites to BCG levels 2 to 6 based on the sample data.

No Level 1 sites (pristine, natural condition) were identified in Connecticut's database. The panel assigned 48 samples to levels of the BCG. For some samples, the panel's evaluation reflected some ambiguity between adjacent levels, such that a sample may have had characteristics intermediate between two levels. From the general descriptions of each of the levels, the panel developed a set of operational rules for assigning sites to levels. These rules ensure consistent decision-making and captured the consensus professional judgment of the panel. Finally, we developed a computerized decision analysis model based on mathematical set theory to replicate the expert panel decisions. This model explicitly uses linguistic rules or logic statements, e.g., "If taxon richness is high, then condition is good" for quantitative, computerized decisions. The decision model can also produce ambiguous decisions among levels, and the model's ambiguity often matched the panel's ambiguity. The model exactly matched the panel decision in 45 of 48 cases (94% concordance). For the remaining 3 cases, the model selected the panel's minority decision as its level of greatest membership.

A multimetric index was also developed for the macroinvertebrate data. Several alternative indexes were evaluated based on the degree of separation of reference site and stressed site index scores, the reliability that the index could separate the stressed sites from the reference sites, variability of index scores among reference sites, and verification results.

The multimetric index included the following metrics:

- Ephemeroptera taxa (scoring adjusted for watershed area)
- Plecoptera taxa
- Trichoptera taxa
- Percent sensitive EPT (scoring adjusted for watershed area)
- Scraper taxa
- BCG Taxa Biotic Index
- Percent dominant genus

The BCG decision model and the multimetric index were overall in concordance on the assessments from the 2 methods. The scoring range of the multimetric index was broken into categories corresponding to BCG levels. This resulted in disagreement of 32% of multimetric scores compared to the BCG decision model, always by a single level. Where the two models did not agree, the expert panel felt that the BCG decision model reflected the true BCG level for the site, but that the anomalous index score showed a potential unusual situation for the site: a particularly good or poor condition within the given BCG level (e.g., a very high Level 4 site), but not enough to rate the site in the next Level.

Data from a set of 20 sites that had been sampled in multiple years were analyzed for variability. The data collected by Connecticut, and the indexes derived from them, show remarkable stability when samples from the same sites are compared among years. The maximum difference within sites was 21 points of the MMI (of 100 points), and 1 level of the BCG.

Implementation of Tiered Aquatic Life Use

Connecticut has 3 Designated Use classes for streams that meet water quality criteria (Connecticut DEP, 2002):

- Class AA: all waters that are designated for an existing or proposed drinking water supply, e.g., all waters upstream of existing drinking water intakes are Class AA;
- Class A: all waters with no permitted discharges that may be potential drinking water supplies;
- Class B: All other waters (mostly with permitted discharges).

In Connecticut's current water quality standards for aquatic life use, there is no reliable mechanism to recognize and protect high quality aquatic communities. Adoption of Tiered Aquatic Life Uses, even within the context of the current AA, A, B classification, would allow the State to protect its best waters, and at the same time to establish tangible and attainable restoration goals for biologically impaired waters, including waters subject to UAA and site-specific criteria.

An approach proposed in this document would be to establish Aquatic Life Use Tiers I, II, and III, corresponding to BCG levels 2, 3, and 4. Waters with a biological community in BCG Level 2 would receive the highest aquatic life classification (Tier I), and would possibly qualify for outstanding natural resource waters.

Consensus of the Connecticut biologists was that BCG Level 4 is minimally acceptable: streams that were rated at low BCG Level 4 or at Level 5 were deemed to fail biocriteria as currently applied in Connecticut. Under the proposed implementation of Tiered Aquatic Life Uses, this current minimum would be retained, and all waters would have a default (unassessed) assignment to Tier III (=BCG level 4). Streams would be reassigned to Tiers I and II upon biological assessment, and finding that they meet the biological conditions for those Tiers.

Issues to Resolve for Implementation

The above system proposes 3 aquatic life use tiers, such that unassessed waters are assumed to be the lowest acceptable Tier (III), which is equivalent to the current minimum acceptable biological condition. Finding that a water body attains a higher Tier is a permanent upward ratchet: waterbodies are protected from degrading from Tier I to II, II to III, etc.

There are several issues to resolve in order to implement Tiered Aquatic Life Uses:

- How can the State protect sites before they become degraded? Waterbodies that meet Tier I could be reclassified (upgraded) as an Outstanding Natural Resource Water. How can the state protect Tier II waterbodies?
- How does the State enforce the biocriteria after degradation is found? For example, suppose a Class A stream was sampled, and found to meet ALU Tier II. Five years later, it is found to have degraded (permanently) to Tier III because several developments have been built in the watershed. Does that stream now go on the TMDL list? Or is there another mechanism under Antidegradation to restore/rehabilitate streams?
- Assignment to tiers should ultimately be based on existing, long-standing unalterable human land use and infrastructure. For example, rural streams with mixed land use in their watersheds may be expected to attain Tier II. The “Expected attainment” is also the restoration goal for water bodies that do not attain the expected Tier.

ACKNOWLEDGMENTS

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DISCLAIMER

This report utilizes data collected by Connecticut Department of Environmental Protection (CT DEP). All data passed CT DEP's QA/QC procedures. No further data quality requirements were defined by EPA; hence, no further data QA was necessary for this project. Quality of the data used for this report has not been evaluated by EPA.

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1.0 INTRODUCTION

The U.S. Environmental Protection Agency (EPA) has supported efforts to develop uniform assessments of aquatic resource condition and to set more uniform aquatic life protection and restoration goals (Davies and Jackson, 2006). These efforts have led to a conceptual model that describes ecological changes—from pristine to completely degraded—that take place in flowing waters with increased anthropogenic degradation (Davies and Jackson, 2006). This model, called the Biological Condition Gradient (BCG), promotes a more consistent application of the Clean Water Act by identifying levels or condition classes that can be operationally defined in a consistent manner (Figure 1-1) across regions and stream types.

Tiered aquatic life uses (TALU) and the BCG require assessors to consider ecological information in making assessments. Biological condition levels are narrative statements on presence, absence, abundance, and relative abundance of several groups of taxa, as well as statements on system connectivity and ecosystem attributes (production, material cycling). The statements are consensus best professional judgments based on years of experience of many biologists in a region and reflect accumulated biological knowledge. It should be noted that empirical developments of the BCG such as this one have only made use of structural attributes of the system.

Levels of Biological Condition

Natural structural, functional, and taxonomic integrity is preserved.

Structure & function similar to natural community with some additional taxa & biomass; ecosystem level functions are fully maintained.

Evident changes in structure due to loss of some rare native taxa; shifts in relative abundance; ecosystem level functions fully maintained.

Moderate changes in structure due to replacement of sensitive ubiquitous taxa by more tolerant taxa; ecosystem functions largely maintained.

Sensitive taxa markedly diminished; conspicuously unbalanced distribution of major taxonomic groups; ecosystem function shows reduced complexity & redundancy.

Extreme changes in structure and ecosystem function; wholesale changes in taxonomic composition; extreme alterations from normal densities.

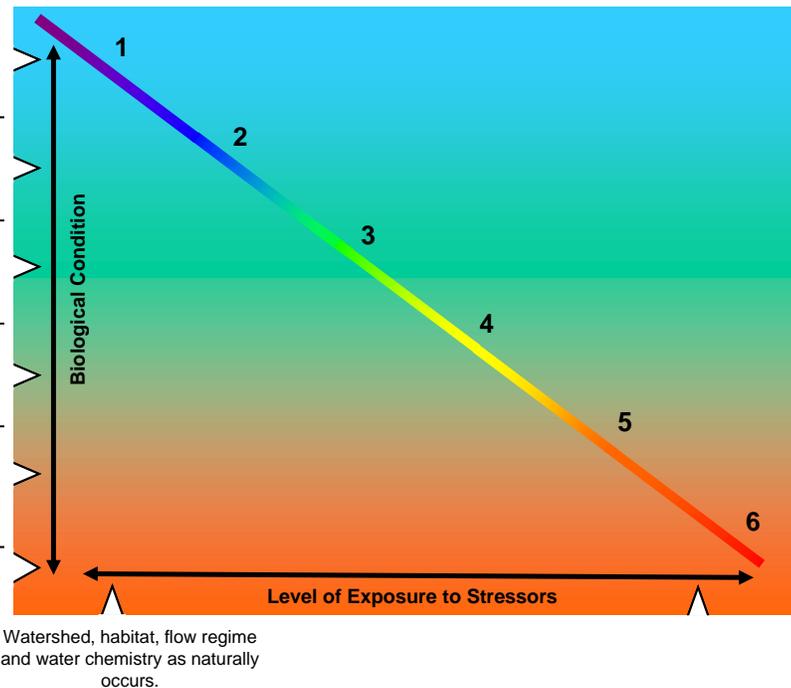


Figure 1-1. Schematic of biological condition gradient, showing six levels of condition.

A central aspect of developing tiered aquatic life uses is to describe the Biological Condition Gradient from unimpaired, relatively pristine waterbodies to severely impaired. The BCG has been described in a general sense for North America and for several regions within North America. The end assessments, the numbered levels shown in Figure 1-1, are on a single scale

that can be applied nationwide. As a universal scale, the BCG can be calibrated to local conditions using specific, local expertise to apply it to conditions within a state.

This report describes the application of the BCG to streams of Connecticut and the development of a more traditional multimetric index; either of which can be used for defining levels for restoration goals and aquatic life protection criteria. For clarity, we reiterate three important definitions: aquatic life use tiers, BCG levels, and BCG attributes. The tiers of the aquatic life use framework refer to programmatic categories of expected use attainment for waterbodies within a state. These should not be confused with the BCG levels, which are narrative descriptions of the biological condition with respect to a gradient from completely natural to severely disturbed. Unless specifically stated, we refer to levels of the BCG from this point forward. The BCG attributes are characteristics of the biological community, individual organisms within the community, and the physical environment. BCG attributes are used to help recognize BCG levels. The predominant BCG attributes used in this analysis are coded as numerals I through VI, which is the same range as the scale of the BCG levels, but level and attribute numbers are not identical or interchangeable.

1.1 The Biological Condition Gradient

Stream communities change in response to pollution, and aquatic biologists have developed indexes to reflect and standardize these changes. Communities are altered on a relatively predictable gradient from pristine to slightly impaired to severely impaired. Indexes that reflect the gradient have included the Saprobien index (Cairns and Pratt, 1993), the index of biotic integrity (IBI; Karr et al., 1986), similarly constructed indexes for macroinvertebrates (Barbour et al., 1999), simple diversity and richness indexes that follow the general loss of native taxa with impairment (e.g., Cairns et al., 1993), and more complex indexes that compare observed taxa to model-predicted expected taxa, such as the River Invertebrate Prediction and Classification System (RIVPACS; Clarke et al., 1996).

1.1.1 Biological Attributes

The BCG systematizes the cumulative knowledge of how aquatic communities change with disturbance by first identifying critical attributes of the community, and then by describing how each attribute changes in response to human disturbance. Through a series of national, EPA-sponsored workshops, a technical workgroup of State, Tribal, academic, and federal biologists described the BCG using the following 10 attributes (EPA, 2005; Davies and Jackson, 2006):

- I. Historically documented, sensitive, long-lived or regionally endemic taxa: refers to taxa known to have been supported in a waterbody or region prior to enactment of the Clean Water Act, according to historical records compiled by state or federal agencies or published scientific literature. Sensitive or regionally endemic taxa have restricted, geographically isolated distribution patterns (occurring only in a locale as opposed to a region), often due to unique life history requirements. They may be long-lived, late maturing, low fecundity, limited mobility, or require a mutualist relation with other species. May be among listed endangered/threatened or special concern species. Predictability of occurrence is often low, therefore, requiring documented observation. Recorded occurrence may be highly dependent on sample methods, site selection and level of effort.
- II. Highly Sensitive Taxa: taxa that naturally occur in low numbers relative to total population density but may make up large relative proportion of richness. They may be ubiquitous in occurrence or

may be restricted to certain micro-habitats, but because of low density, recorded occurrence is dependent on sample effort. Often stenothermic (having a narrow range of thermal tolerance) or cold-water obligates; they are commonly k-strategists (populations maintained at a fairly constant level; slower development; longer life-span). They may have specialized food resource needs or feeding strategies and are generally intolerant to significant alteration of the physical or chemical environment; is often the first taxa observed to be lost from a community.

- III. Intermediate Sensitive Taxa, (or Sensitive and Common Taxa): taxa that are ordinarily common and abundant in natural communities when conventional sample methods are used. They often have a broader range of thermal tolerance than Sensitive- Rare taxa. These are taxa that comprise a substantial portion of natural communities, and that often exhibit negative response (loss of population, richness) at mild pollution loads or habitat alteration.
- IV. Taxa of Intermediate Tolerance: taxa that make up a substantial portion of natural communities; may be r-strategists (early colonizers with rapid turn-over times; “boom/bust” population characteristics). They may be eurythermal (having a broad thermal tolerance range). May have generalist or facultative feeding strategies enabling utilization of relatively more diversified food types. Readily collected with conventional sample methods. May increase in number in waters with moderately increased organic resources and reduced competition but are intolerant of excessive pollution loads or habitat alteration.
- V. Tolerant Taxa: Taxa that make up a low proportion of natural communities. These taxa often are tolerant of a broader range of environmental conditions and are thus resistant to a variety of pollution or habitat induced stress. They may increase in number (sometimes greatly) in the absence of competition. Commonly r-strategists (early colonizers with rapid turn-over times; “boom/bust” population characteristics), able to capitalize when stress conditions occur. These are the last survivors in severely disturbed systems.
- VI. Non-native or Intentionally Introduced Species: with respect to a particular ecosystem, any species that is not found in that ecosystem. Species introduced or spread from one region of the U.S. to another outside their normal range are non-native or non-indigenous, as are species introduced from other continents.
- VII. Organism Condition (especially of long-lived organisms): general indicators of organism health, such as deformities, anomalies, lesions, tumors, or excess parasitism are all external indicators of condition.
- VIII. Ecosystem Function: function includes trophic levels, production, respiration, total biomass and biomass in functional levels, P/R ratios, etc.
- IX. Spatial and Temporal Extent of Detrimental Effects: the spatial extent of damage or degradation from a particular source.
- X. Ecosystem Connectance: natural connections and relation among ecosystem units, such as extent fragmentation, connections of riparian areas with the stream and floodplain, etc.

The last three attributes, Ecosystem Function, Spatial and Temporal Extent, and Ecosystem Connectance, were all deemed ecologically important by the workgroups that developed the BCG (Davies and Jackson, 2006), but none have been applied or tested in either regional or state contexts. There is disagreement among ecologists whether measures of ecosystem function provide unique information on condition not already provided by the more common structural measures. Attributes IX and X, both spatial attributes, were considered by some ecologists to be measures of stress, and not biological response to stress. Routine monitoring programs (including Connecticut’s) do not normally collect information on these attributes (VIII to X). In

this development for Connecticut, we did not use attributes VII to X. Attribute VII, Organism Condition, is commonly measured by agencies that monitor fish and edible shellfish.

1.1.2 Levels of the Condition Gradient

At the national workshops, biologists agreed that in most stream ecosystems it was possible to discriminate six levels in the condition gradient, ranging from undisturbed natural condition to severely degraded and almost devoid of natural life. The levels are described in terms of changes in the structure and function of native aquatic communities. Although the condition levels are described in terms of both structure and function, empirical application of the BCG have so far not incorporated the functional or spatial attributes.

1. Natural or native condition: Native structural, functional and taxonomic integrity is preserved; ecosystem function is preserved within the range of natural variability.
2. Minimal changes in structure of the biotic community and minimal changes in ecosystem function: Virtually all native taxa are maintained with some changes in biomass and/or abundance; ecosystem functions are fully maintained within the range of natural variability.
3. Evident changes in structure of the biotic community and minimal changes in ecosystem function: Evident changes in structure due to loss of some rare native taxa; shifts in relative abundance of taxa but sensitive-ubiquitous taxa are common and abundant; ecosystem functions are fully maintained through redundant attributes of the system.
4. Moderate changes in structure of the biotic community with minimal changes in ecosystem function: Moderate changes in structure due to replacement of some sensitive-ubiquitous taxa by more tolerant taxa, but reproducing populations of some sensitive taxa are maintained; overall balanced distribution of all expected major groups; ecosystem functions largely maintained through redundant attributes.
5. Major changes in structure of the biotic community and moderate changes in ecosystem function: Sensitive taxa are markedly diminished; conspicuously unbalanced distribution of major groups from that expected; organism condition shows signs of physiological stress; ecosystem function shows reduced complexity and redundancy; increased build-up or export of unused materials.
6. Severe changes in structure of the biotic community and major loss of ecosystem function: Extreme changes in structure; wholesale changes in taxonomic composition; extreme alterations from normal densities and distributions; organism condition is often poor; ecosystem functions are severely altered.

1.2 Development of Attributes and Gradient for Connecticut

Aquatic biologists familiar with Connecticut streams convened in a workshop to develop both the ecological attributes and rules for assigning sites to levels in the gradient. Their expertise included aquatic ecology, benthic macroinvertebrate sampling and monitoring, water quality, and

fisheries biology. Although the BCG is intended to be developed and applied for as many taxonomic groups as possible (e.g., benthic macroinvertebrates, periphyton, fish, herpetofauna, vascular plants, etc.), this development of the gradient included systematic application to benthic macroinvertebrates only, collected by the methods used in Connecticut's monitoring program. Integration of fish and other taxonomic groups into the descriptions of the BCG must await future iterations of the process. As in other applications, we developed the BCG using only Attributes I–VI, because the monitoring program does not collect information on the other attributes.

After reviewing EPA's conceptual model of the biological condition gradient, the group reviewed the list of taxa identified in the Connecticut ambient monitoring program to assign taxa to attribute groups I–VI. Appendix A includes the taxa list and assigned attribute groups. The group then considered data from selected monitoring sites and assigned the sites to levels in the BCG based on the taxa present in the sample. Details of these processes are presented in the Methods section.

1.3 Aquatic Life Uses

A biological condition gradient requires strong scientific knowledge on the response of aquatic biological assemblages to stressors, as well as the biota inhabiting a region. Using the scientific information to better assess and manage living aquatic resources also requires a legal foundation that permits the determination of scientifically defensible management goals (policies, designated uses, standards, criteria) in keeping with the goals of the Clean Water Act. Finally, developing a quantitative methodology for assessing waterbodies in relation to the BCG requires a scientifically sound biological monitoring program.

Under the Clean Water Act a state can identify use classes, called Designated Uses, for its waterbodies. As biological condition can be divided into levels, so can designated aquatic life uses of waterbodies be divided into tiers corresponding to the biological expectation for the different uses. The relationship between aquatic life use (ALU) tiers and BCG levels must be addressed in the context of State programs and policies. BCG development may be required for each tier of ALU (where the ALU tier is defined by environmental classification), or BCG levels may coincide with aquatic life use tiers (where the expected biological condition is the basis for the ALU tier). In this report, we focus on the BCG level development.

2.0 METHODS

The calibration process includes

- assessment of the state's biological monitoring program to support quantitative calibration of a regional BCG;
- identification of attributes of condition that will be used to build the BCG; assigning taxa to the attributes;
- development of the regional model of the BCG, and its calibration for operational assessment; and
- analysis of biological condition using additional tools to confirm BCG model development and to aid in its application.

The development process is iterative and may require several passes through the process to converge on a coherent, locally calibrated BCG that is scientifically defensible.

2.1 Connecticut Ambient Monitoring Program

Consistent, high quality biological monitoring information is key to developing a quantitative assessment system within a BCG framework. Connecticut DEP operates a sizable ambient monitoring program throughout the state (CT DEP, 2005). The following description is excerpted from DEP's draft report (CT DEP, 2004) "Ambient Water Quality Monitoring: Rotating Basin Approach Data Summary (1996–2001)":

Connecticut contains a total of approximately 5,830 miles of rivers and streams (EPA, 1993). The Connecticut DEP has organized the hydrography of the State into a hierarchical system of natural drainage basins comprised of four basic levels of magnitude (CT DEP, 1981). Major basins represent the greatest level of magnitude and are roughly equivalent, but not identical to, USGS eight digit cataloging units. Major basins are comprised of three categories of sub basins; in order of decreasing magnitude, these are regional, sub-regional, and local basins. The distribution of drainage basin units at each level of magnitude is listed below.

Beginning in 1996, the Bureau of Water Management (BWM) initiated a rotating basin approach to monitoring and assessment. This approach is consistent with the current 305(b) guidelines and the overall goal of a more comprehensive statewide assessment by ultimately increasing the number of river miles monitored.

To accomplish this plan the State was divided into five hydrologic assessment units comprised of one or two CT DEP major basins, or USGS cataloging units. The assessment units are... shown in Figure 2-1. Monitoring and assessment efforts will be concentrated on one unit each year for a five-year period. Implementation began during the fall of 1996.

Sample Collection:

The primary collection method follows EPA Rapid Bioassessment Protocol III (RBP III) for Streams and Rivers (Plafkin, 1989). RBP III involves collecting 12 kick samples (stops) throughout riffles at sampling sites using a rectangular net (18"x18"x10") with 800 × 900 μm mesh. The stops are spread out as best as possible both up, down, and across the riffle. The resulting sample is meant to represent the community as a whole within the riffle. The contents from all 12 stops are composited into sample container(s) and preserved in the field with 70% ethyl alcohol. An alternate method when

habitat may be limited to the benthic community is RBP I (Plafkin et al., 1989). The primary difference with RBP III is that the organisms are removed directly from the debris in the net and are not sub-sampled in the laboratory under controlled conditions. Samples collected using the RBP I method are termed "NQ-Pick", for Non-quantitative Pick.

Benthic community sites for each basin are sampled during the fall benthic community index period (October 1–November 30). Benthic community sampling can also occur during a spring index period (April 1–May 31), but based on experience, DEP considers the fall index period to better represent the worst-case condition. Since differences in habitat conditions are minimized with this approach, differences in the benthic communities of two sites should be primarily due to water quality differences. A complete description of sampling protocol is available in the Ambient Biological Monitoring-Benthic Macroinvertebrates Quality Assurance Project Plan (CT DEP, 2003b).

Laboratory Analysis:

Identification is to lowest practical taxa based on a 200-organism minimum sub-sample. Based on the organisms present in the sub-sample, a series of community structure metrics are calculated and compared to metrics a reference site. A reference site is a specific locality on a waterbody that is minimally impaired and is representative of the expected ecological integrity of other localities on the same waterbody or nearby waterbodies. The final result of RBP III is an assessment of the impairment level of the benthic community.

Rotating Secondary Physical/Chemical Monitoring Network

This network is intended to supplement the primary network sites by providing physical/chemical data on selected rivers. Sampling frequency is quarterly for one year, which is consistent with the rotating basin schedule. Third quarter sampling events are coincident with critical stress periods characterized by low stream flow and elevated water temperature. Sampling site selection is based on a targeted approach considering sub basin size, location of wastewater discharges, land use, and resource value. Conventional water quality parameters, toxic metals, and indicator bacteria are measured by means of grab samples. Personnel from the DEP, Bureau of Water Management, perform sample collection and field measurements. The Connecticut Department of Public Health (CT DPH), Laboratory Division, conducted laboratory analyses (CT DEP, 2004).

Land Use/ Land Cover data

Land cover data were obtained from the University of Connecticut's Center for Land use Education and Research (CLEAR); (<http://clear.uconn.edu/projects/landscape/index.htm>). We used land cover estimated for 2002, in the following categories:

- Impervious surface
- Developed land (built-up, roads)
- Turf and grass (e.g., lawns and parks)
- Agriculture and grass (e.g., pasture)
- Deciduous forest
- Coniferous forest
- Water
- Forested wetland
- Non-forested freshwater wetland
- Tidal wetland
- Utility right-of-way
- Barren land

Connecticut DEP had delineated catchments for each sampling site in the data base and calculated the area of each land cover category within the catchment. Because the CLEAR database did not extend beyond Connecticut's borders, some sites with catchments partially in Massachusetts and New York had incomplete land cover data. For data analysis, the two forest, water, and three wetland categories were added to define a "natural land cover" category.

Data Management

Currently, all benthic data and associated metadata are entered into a Microsoft Access database, where metrics and summary information are generated through queries. Some of the benthic data is still brought into Access through Excel sheets, but CT DEP is working towards entering all of the benthic data from the taxonomist's logbook directly into Access via existing entry forms (M. Beauchene, pers. comm. to J. Gerritsen).

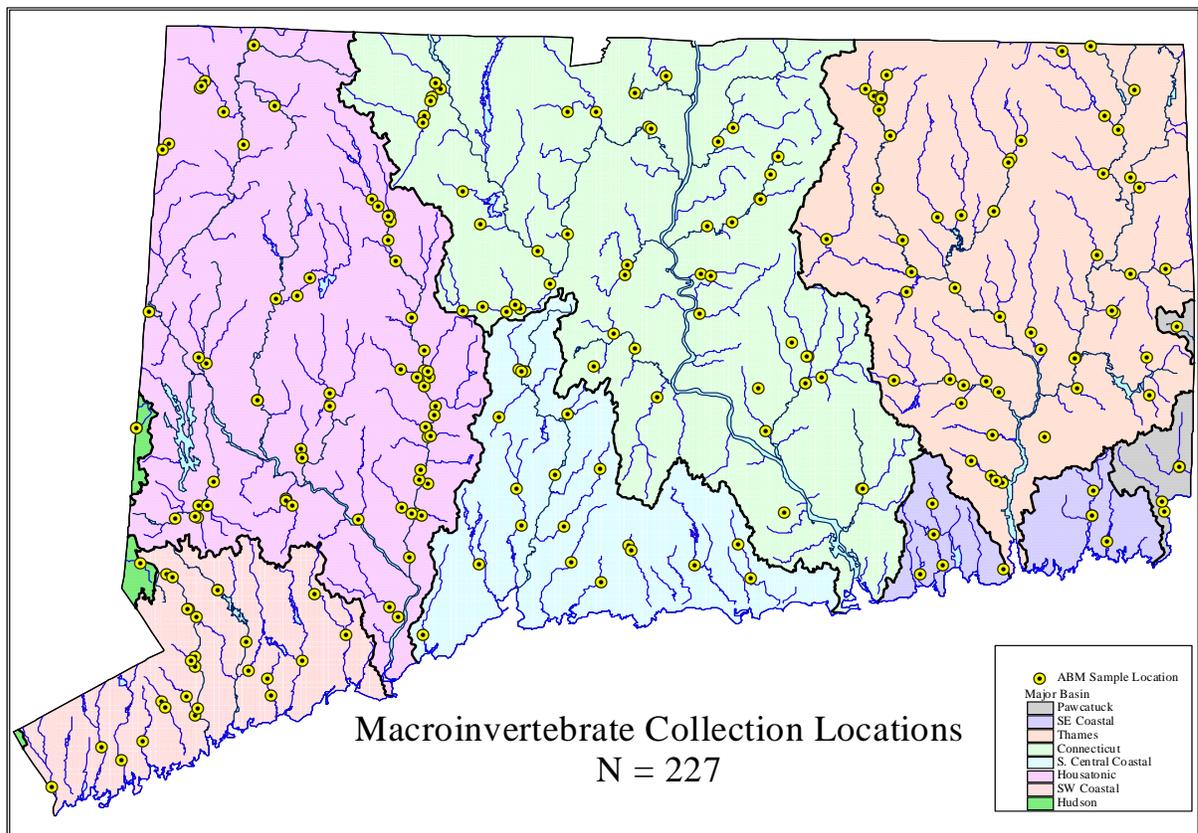


Figure 2-1. Distribution of sampling sites across Connecticut as of 2001, showing major basins (from CT DEP, 2004).

2.2 Identifying Attributes

2.2.1 Preliminary Disturbance Gradient

We identified several stress categories for Connecticut monitoring sites, based on land use and chemistry of samples in the database. Connecticut DEP was a participant in the New England Wadeable Streams survey (Snook et al., 2007), and analyses from that survey are applicable to Connecticut. The NEWS data had shown that urban land use, natural land cover, population density, and chloride concentration were excellent predictors of biological condition. For the Connecticut data we had no population densities, but we did have a “developed” (built-up) category, as well as estimates of impervious surface. Candidate BCG level 1 sites in the NEWS data had < 5 persons per square mile, < 0.5% urban, > 90% natural land cover, and < 5 mg/L chloride. None of the sampled sites in Connecticut met all the NEWS BCG level 1 criteria, so we developed criteria for “Least stressed” in Connecticut, and 6 other categories:

1. Least Stress: Meets all 4 least stressed criteria (Table 2-1), and all 4 metals below metal thresholds (Table 2-2) (n=24)
2. Candidate Least: Meets land cover criteria for least stressed, but chemical (chloride and metals) data were missing (n=1)
3. Slight stress: Meets slight stress criteria and all 4 metals below thresholds (natural land cover or chloride fail least-stressed) (n=37)
4. Moderate stress: Fails one or more slight stress criteria, or single parameter above “high stress” criteria, and no metals above threshold (n=87)
5. Metal contamination: Any one metal greater than threshold (Table 2-2) but otherwise Moderate Stress or better (n=22)
6. Heavy stress: Any 2 or 3 high stress criteria met (Table 2-1) (n=47)
7. Severe stress: All 4 high stress criteria met (Table 2-1) (n=17)

Table 2-1. Stress criteria

Parameter	Least stress	Slight stress	Moderate Stress	High stress
Natural land cover*	> 80%	70%–80%	60%–70%	< 60%
Developed land	< 10%	< 10%	10%–25%	> 25%
Impervious surface	< 4%	< 4%	4%–10%	> 10%
Chloride	<15 mg/L	15–20 mg/L	20–30 mg/L	> 30 mg/L
Decision Criteria	meets all, no metals	1 or 2 in “Slight” category, others “Least”, no metals	Any in “Moderate” or single parameter in “High” category, no metals	Heavy stress: 2 or 3 parameters in “High”; any metals Severe stress: All 4 in “High” category, any metals

*defined as the sum of deciduous, conifer, open water, and all wetland categories

Table 2-2. Metals Criteria (all dissolved)

Metal	Threshold (mg/L)	notes
Copper	0.008	Highest concentrations all impaired
Iron	0.4	Effect marginal
Nickel	0.01	Highest concentrations showed effects, but confounded by detection limit
Zinc	0.02	Strong effect

Screening thresholds for metals (Table 2-2) were determined from scatterplots of number of mayfly or stonefly taxa in the samples vs. metal concentrations (Figure 2-2). These two orders are generally considered highly sensitive to metal contamination, in part due to the large number of chloride cells on their surfaces (e.g., Buchwalter and Luoma, 2005). Metals not included in Table 2-2 (Al, Cd, Hg, Pb, Se) were not associated with biological responses (Al, Hg, Pb), or had too few detectable observations in the data (Cd, Se). Data were not stratified for several other potential stressors (nutrients, BOD, total solids, coliform) because they were redundant with the stressor gradient classes described above. Using the criteria of Tables 2-1 and 2-2, we identified 24 Least Stressed sites as potential reference sites, and 17 Severely Stressed sites to help calibrate the BCG and the MMI (Table 2-3).

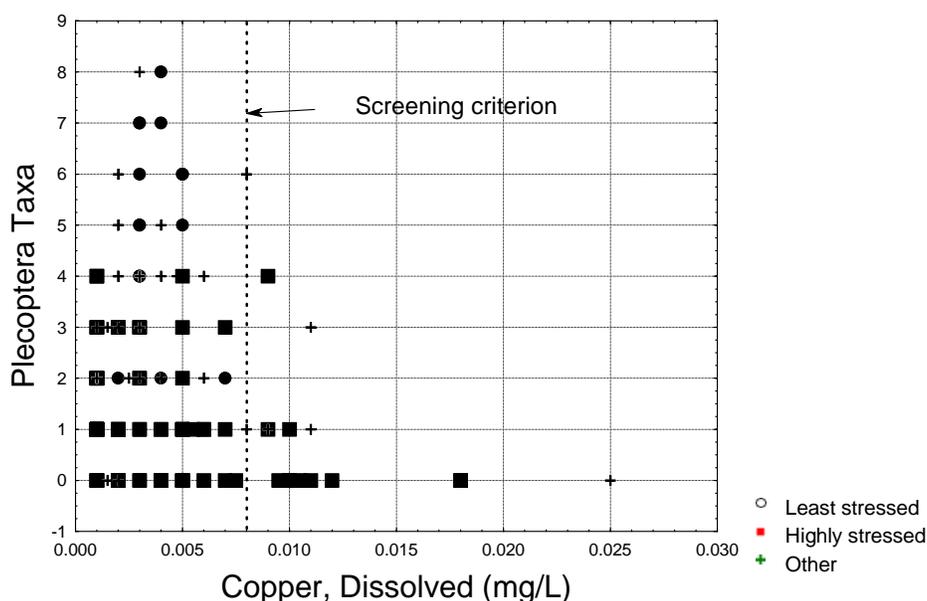


Figure 2-2. Number of Plecoptera (stonefly) taxa and dissolved copper concentration. The screening criterion was determined from sharp decline of stonefly taxa above 0.008 mg/L copper.

Table 2-3. Least stressed and severely stressed sites used in BCG and index development.

Site Code	Waterbody Name	Basin	Town	BCG, MMI	Latitude	Longitude	Ecoregion	Area Sq. Mi.
				Calibration Verification ¹				
Least Stressed Sites								
911	Beach Brook	4319	Granby	MC, B	41.94597	-72.8575	NEHighlands	2.09
468	Bigelow Brook	3203	Eastford	MC	41.86705	-72.0921	NECoastalZone	29.279
924	Clark Creek	4000	Haddam	MC, B	41.44255	-72.4735	NECoastalZone	2.41
907	East Branch Salmon Brook	4320	Granby	MV, B	42.01354	-72.8435	NEHighlands	4.57
62	Eightmile River	4800	Lyme	MC	41.43121	-72.3376	NECoastalZone	20.65
930	Eightmile River	4800	Lyme	MV	41.43003	-72.3392	NECoastalZone	43.19
72	Farmington River	4300	Farmington	MV	41.75077	-72.8717	NECoastalZone	386.79
96	Hammonasset River	5106	Madison	MV	41.32782	-72.6116	NECoastalZone	22.43
122	Hollenbeck River	6200	Canaan	MC	41.94308	-73.3058	NEHighlands	17.56
469	Mount Hope River	3206	Mansfield	MC, B	41.79706	-72.1716	NECoastalZone	28.07
740	Mountain Brook	4320	Granby	MC, B	41.97407	-72.8375	NEHighlands	2.09
189	Natchaug River	3200	Chaplin	MC	41.80083	-72.1183	NECoastalZone	73.17
627	Quaker Brook	8101	New Fairfield	B	41.51020	-73.5289		5.09
462	Roaring Brook	3104	Willington	MC	41.90402	-72.2891	NECoastalZone	22.01
780	Sages Ravine Brook	6001	Salisbury	MC, B	42.04953	-73.4301	NEHighlands	3.54
317	Sandy Brook	4304	Colebrook	MC, B	41.97403	-73.0406	NEHighlands	36.93
743	Sandy Brook	4304	Colebrook		41.99041	-73.058	NEHighlands	34.77
746	Sawmill Brook	6401	Sherman	MC	41.58511	-73.5108	NEHighlands	2.92
596	Shepaug River	6700	Washington	MC	41.68358	-73.3019	NECoastalZone	40.68
766	Stickney Hill Brook	3104	Union	MV	41.98333	-72.2179	NECoastalZone	5.87
908	Still Brook	3102	Stafford	MC	42.01921	-72.3127	NECoastalZone	1.77
357	West Branch Naugatuck River	6904	Torrington	MC, B	41.81814	-73.1441	NEHighlands	31.07
359	West Branch Salmon Brook	4319	Granby	MC	41.93717	-72.8215	NECoastalZone	23.76
605	Wyassup Brook	1001	N. Stonington	MV	41.45664	-71.8172	NECoastalZone	11.47

Table 2-3. Continued.

Site Code	Waterbody Name	Basin	Town	BCG, MMI Calibration Verification ¹	Latitude	Longitude	Ecoregion	Area Sq. Mi.
Severely Stressed Sites								
76	Five Mile River	7401	New Canaan	MC	41.14183	-73.4833	NECoastalZone	6.23
119	Hockanum River	4500	Manchester	MC	41.78828	-72.5503	NECoastalZone	55.48
110	Hockanum River	4500	East Hartford	MV	41.78218	-72.5912	NECoastalZone	74.40
159	Mad River	6914	Waterbury	MC, B	41.54393	-73.0384	NECoastalZone	25.93
233	Noroton River	7403	Stamford	MC, B	41.08984	-73.5152	NECoastalZone	9.58
236	Norwalk River	7300	Norwalk	MC, B	41.13587	-73.426	NECoastalZone	27.61
267	Pequabuck River	4315	Bristol	MC, B	41.67381	-72.8977	NECoastalZone	45.64
269	Pequonnock River	7105	Trumbull	MC	41.2343	-73.1838	NECoastalZone	22.08
272	Piper Brook	4402	Newington	MC	41.71861	-72.7274	NECoastalZone	17.21
289	Quinnipiac River	5200	Wallingford	MC, B	41.45008	-72.8407	NECoastalZone	110.98
514	Steele Brook	6912	Waterbury		41.56869	-73.0574	NECoastalZone	17.04
331	Steele Brook	6912	Waterbury	MV	41.58051	-73.0703	NECoastalZone	17.04
339	Still River	6600	Danbury	MC	41.40633	-73.4253	NECoastalZone	38.05
333	Still River	6600	Brookfield	MC, B	41.4389	-73.401	NECoastalZone	52.33
338	Still River	6600	Danbury	MV	41.38981	-73.4637	NECoastalZone	14.43
342	Sympaug Brook	6604	Danbury	MC, B	41.39229	-73.4284	NECoastalZone	7.25
354	Trout Brook	4403	West Hartford	MV	41.73135	-72.7231	NECoastalZone	17.75

¹ MC: MMI calibration site; MV: MMI verification site; B: BCG panel calibration site; Blank: Not used for calibration - met criteria for reference or stressed but too close to another site used in index development; would have represented redundant data.

2.2.2 Taxa List and Site Gradient

Prior to calibrating BCG levels, the workgroup assigned Connecticut taxa to the taxonomic attribute groups (Attributes I to VI; Section 1.1.1). Assignments of taxa to attributes relied on a combination of empirical examination of taxon occurrences at sites in the different stress classes, as well as professional experience of field biologists who had sampled the streams of Connecticut. The empirical analyses and professional opinions tended to agree, but in cases of disagreement, the group relied on consensus professional opinion, unless contradicted by an overwhelming response in the data analysis. As a group, participants discussed each taxon in the calibration data set, and developed a consensus assignment (Appendix A).

Biologists have long observed that taxa differ in their sensitivity to pollution and disturbance. While biologists largely agree on the relative sensitivity of taxa, there may be subtle differences among stream types (high vs. low gradient) or among geographic regions. The workgroup empirically examined the sensitivities of the benthic macroinvertebrates to the generalized stressor gradient classes described above.

2.3 Development of the BCG

Calibrating a regional BCG requires adjustment of the generalized conceptual model to regional conditions (Davies and Jackson, 2006; EPA, 2005; summarized in the Introduction). This includes components that construct a coherent ecological description of response to stressors in keeping with ecological theory and empirical observation:

- Describe the native aquatic assemblages under natural, undisturbed conditions. The description of natural conditions requires biological knowledge of the region, a natural classification of the assemblages, and, if available, historical descriptions of the habitats and assemblages.
- Identify regional stressors. A description of regionally dominant stressors will help define expectations for biological responses that are likely to occur. This step considers sources of physical and chemical stressors and causes of land use disturbance.
- Quantitative description of BCG levels that are the system responses to anthropogenic stressors.

2.3.1 Classification

Bioassessment is based on developing expectations for natural conditions where there are many natural variables (such as stream size, slope, dominant natural substrate, etc.) which may affect species composition of undisturbed streams. Accordingly, a critical step in any bioassessment development is to classify the natural conditions to the extent that they affect the biological indicators (Gerritsen and Paul, 2006).

Strata of biologically similar groups can be identified among reference sites through examination of biological gradients or assemblage types and association of the biological gradient with natural variables. We used non-metric multidimensional scaling (NMS) of the taxonomic data to

examine potential groupings. Additional supporting analyses included indicator species analysis, correlations, cluster analysis, metric distribution plots, and regression analysis. Stratification into distinct site classes is useful when each of the resulting classes is represented by sufficient numbers of samples to allow meaningful analysis within or among site classes.

NMS allows a comparison of taxa within each sample and an arrangement of the samples so that similar samples plot closer together than dissimilar samples in multiple dimensions. Natural environmental variables can be associated with the biological gradient through correlations with the biologically defined axes of the NMS diagram. NMS is a robust method for detecting similarity and differences among ecological community samples and works as well using presence/absence data as relative abundance data (McCune and Grace, 2002).

A site by taxa matrix was compiled. Similarity among reference biological samples was made using the Bray-Curtis (BC) similarity measure. The BC formula is sometimes written in shorthand as

$$BC = 1 - 2W / (A + B)$$

where W is the sum of shared abundances and A and B are the sums of abundances in individual sample units. The ordination software (PC-Ord, McCune and Grace, 2002) calculates a site by site matrix of BC similarity from which the arrangement of samples in the ordination diagram is derived. Multiple dimensions are compressed into two or three dimensions that we can perceive.

Rare and ambiguous taxa are not useful in the NMS ordination. Rare taxa were defined as those that occurred in less than three reference samples. Ambiguous taxa are those that are identified at higher taxonomic levels because of damaged or undeveloped specimens. The site by taxa matrix was therefore reduced to retain as much information as possible while excluding rare and ambiguous taxa. When several rare genera occurred within one family or when several identifications were at the family level, then all individuals were counted at the family level. When most identifications within a family were made at genus level, then the fewer identifications made at family level were excluded from the analysis. The site by environmental variable matrix included location information and catchment characteristics.

The NMS ordination methods used in classification of natural strata were also used in distinguishing taxa responses to the stressor gradient.

2.3.2 BCG levels

BCG level descriptions in the conceptual model tend to be rather general (e.g., “reduced richness”). To allow for consistent assignments of sites to BCG levels, it is necessary to operationalize, or codify, the general BCG level descriptions into a set of rules that anyone can follow and obtain the same BCG level assignments as the group of experts.

Operational rules codify the BCG level descriptions (“as naturally occur”, “reduced”, “greatly reduced”, etc.) to quantitative or semi-quantitative rules for each attribute (“Attribute 2 taxa > 50% of any other attribute”). These rules preserve the collective professional judgment of the

expert group and set the stage for the development of models that reliably assign sites to BCG levels without having to reconvene the same group. In essence, the rules and the models capture the group's collective decision criteria.

Rules are logic statements that experts use to make their decisions, for example: "If taxon richness is high, then biological condition is high." Rules on attributes can be combined, for example: "If the number of highly sensitive taxa (Attribute II) is high, and the number of tolerant individuals (Attribute V) is low, then the BCG level is 2." In questioning individuals on how decisions are made in assigning sites to BCG levels, it became evident that rules are not sharply defined ("crisp"). For example, there is no distinct number of highly sensitive taxa that would always distinguish BCG level 2 from BCG level 3. Rather, people use strength of evidence in allowing some deviation from their ideal for any individual attributes, as long as most attributes are in or near the desired range. Clearly, the definitions of "high," "moderate," "low," etc., are uncertain. These rules preserve the collective professional judgment of the expert group and set the stage for the development of models that reliably assign sites to BCG levels without having to reconvene the same group. In essence, the rules and the models capture the group's collective decision criteria.

Rule development required discussion and documentation of BCG level assignment decisions and the reasoning behind the decisions. During this discussion, we recorded

- each participant's decision ("vote") for the site;
- the critical or most important information for the decision—for example, the number of taxa of a certain attribute, the abundance of an attribute, the presence of indicator taxa, etc.; and
- any confounding or conflicting information and how this was resolved for the eventual decision.

Rule development was iterative. Following the initial development phase, the draft rules were tested by the panel to ensure that new data and new sites are assessed in the same way. The test sites had not been used in the initial rule development and also spanned the range of anthropogenic stress. Any remaining ambiguities and inconsistencies from the first iterations were also resolved. Rules can be used directly for assessments, for calibrating other assessment methods (e.g., multimetric index or discriminant model), or as the basis of an expert system.

2.4 Decision Criteria Model

Consensus professional judgment used to describe the BCG levels can take into account nonlinear responses, uncommon stressors, masking of responses, and unequal weighting of attributes. This is in contrast to the commonly used biological indexes, which are typically unweighted sums of attributes (e.g., multimetric indexes; Karr and Chu, 1999; Barbour et al., 1999), or a single attribute, observed to expected taxa (e.g., Wright, 2000; Simpson and Norris, 2000). Consensus assessments built from the professional judgment of many experts result in a high degree of confidence in the assessments, but the assessments are labor-intensive (several experts must rate each site). It is also not practical to reconvene the same group of experts for every site monitored in a long-term program for assessment and management. Since individuals

may be replaced on a panel over time, assessments may in turn “drift” due to individual differences of new panelists. Management and regulation, however, require clear and consistent methods and rules for assessment, which do not drift unless deliberately reset.

Use of the BCG in operational monitoring, management, and regulation thus requires a way to automate the consensus expert judgment so that the assessments are consistent until such time that they are explicitly altered due to new knowledge becoming available. Two options have been used in the past: the Maine DEP developed a set of multivariate linear discriminant models to imitate the expert consensus and predict a site assessment (Davies et al., 1995); and the UK Environmental Agency defined ranges of scores of two indexes (their RIVPACS index and a tolerance index) that corresponded to the expert consensus (Hemsley-Flint, 2000). Both of these approaches require one or more multivariate statistical models to statistically predict the expert judgment in assessments.

Instead of a statistical prediction of expert judgment, we have chosen to use a methodology that directly, explicitly and transparently converts the expert consensus to automated site assessment. The method uses fuzzy set theory applied to rules developed by the group of experts. Fuzzy sets and fuzzy logic are directly applicable to environmental assessment; they have been used extensively in engineering applications worldwide (e.g., Demicco and Klir, 2004) and environmental applications have been explored in Europe and Asia (e.g., Castella and Speight, 1996; Ibelings et al., 2003). We applied the approach for a fuzzy-set model developed for BCG assessment in New Jersey (Gerritsen et al., submitted), modified for the rules developed specifically for Connecticut streams.

Fuzzy sets and fuzzy logic allow degrees of membership (in sets) and degrees of truth (in logic), compared to all-or-nothing in classical set theory and logic. This has immediate advantages in scientific classification, for example, “sand” and “gravel,” where a particle with diameter of 1.999 mm is classified as “sand” in classical set theory, and one with 2.001 mm diameter is classified as “gravel.” In fuzzy set theory, both particles may have nearly equal membership in both classes (Demicco, 2004). Demicco and Klir (2004) proposed four reasons why fuzzy sets and fuzzy logic enhance scientific methodology

- fuzzy sets can capture “irreducible measurement uncertainty”, as in the sand/gravel example above;
- fuzzy logic captures vagueness of linguistic terms, such as “many,” “large” or “few”;
- fuzzy sets and logic can be used to manage complexity and computational costs of control and decision systems; and
- fuzzy logic enhances the ability to model human reasoning and decision-making.

2.5 Multimetric Index (MMI) Development

MMIs incorporate several signals of biological response by combining multiple measurements of a sample. In this analysis, the MMI was used to confirm results of the BCG calibration and to provide another tool for its application. The premise of the index development process is that physical and chemical disturbances are reflected by changes in the benthic macroinvertebrate community. Physical and chemical characteristics can first be used to distinguish minimally

disturbed (reference) sites from sites disturbed through human activity. Meaningful biological signals of disturbance are summarized in a multimetric index that can be used to evaluate biological integrity in sites of unknown quality. The development of a multimetric index calibrated on the benthic macroinvertebrate and environmental data collected in wadeable Connecticut streams followed a series of steps, as follows:

1. Collect and organize the data;
2. Define reference and stressed sites;
3. Stratify natural biological conditions;
4. Calculate biological metrics and determine metric sensitivity to stresses;
5. Combine appropriate metrics into index alternatives;
6. Select the most appropriate index for application in high gradient streams based on sensitivity and variability, and;
7. Assess performance of the index.

3.0 STREAMS AND BENTHOS OF CONNECTICUT-DESCRIPTIVE RESULTS

3.1 Regional Description

Connecticut is similar to most of the rest of the eastern US in the historical patterns of settlement and land use. Initial clearing of native forest was primarily for agriculture, towns, and lumber needs in the Colonial and early Federal eras. By the late 1700s, most of the native forest had been cleared (Bell, 1985). Connecticut's abundant water power from high gradient streams fostered early industrial development from the late Colonial era until the advent of steam power. Dominant industries of the 19th century were iron, copper, and textiles. The iron industry used local areas and charcoal for smelting, resulting in severe deforestation and firewood shortages in the mid-1800s (Bell, 1985). Connecticut's iron industry disappeared by the early 20th century because it could not compete against Midwestern large-scale steel mill. The copper industry and textiles remained important until the mid-20th century, after which they also declined. Light and medium industrial development continued through the middle of the 20th century, but the large-scale basic industries of steel and refining were limited by distance from raw materials. Industrialization also left a legacy on the landscape and in the state's waterbodies, often as buried or exposed contamination, which may leach or run off into streams.

Agriculture was more extensive in the uplands and hills outside of major river valleys prior to the US Civil War (1861-65) than it is today. The growth of railroad transportation during the 19th century led to the importation of cheaper food from the Midwest. Economically marginal farms established on rocky upland soils were largely abandoned in the period between 1870 and 1930, and reverted to forest (e.g., Bell, 1985). A legacy of 18th and 19th century agriculture is the stone fences now seen throughout New England's forests.

Connecticut has been urbanized since the peak of 19th century industrial development. Major urban areas today include the southwestern counties that form suburbs and exurbs of New York, New Haven and Hartford. Increasing suburban development extending from urban centers has led to extensive conversion of mixed agriculture and regrown wooded land to urban and suburban uses (e.g., Bell, 1985; Maizel et al., 1998).

3.2 Stream Classification

Ordination

In the NMS ordination of reference samples, the strongest classification variable was catchment size category. In ordinations of sites in taxa space (both presence/absence and relative abundance), the small and large sites (cutoff 10 square miles) are on opposite ends of the first axis (Figure 3-1). The smaller sites have more representation by the flies (Diptera), especially midges (Chironomidae). Mayflies, stoneflies, and caddisflies (Ephemeroptera, Plecoptera, and Trichoptera; EPT) were more dominant in larger sites (especially mayflies).

Cluster analysis

A cluster analysis revealed obvious groupings based on two or three clusters. With only two clusters, sample similarities based on relative abundance and on presence/absence give identical results. Discriminant function analysis (DFA) was used to identify natural determinants of the

distinctive biological groupings. The DFA was efficient at identifying two clusters based on catchment size alone. After entering catchment size in the models, other variables (e.g., ecoregion, latitude, longitude) have insignificant discriminatory power. Ecoregion was coded as a binary variable (NE Coastal Zone = 0, NE Highlands = 1). Classification errors increased when models identifying three groups were attempted.

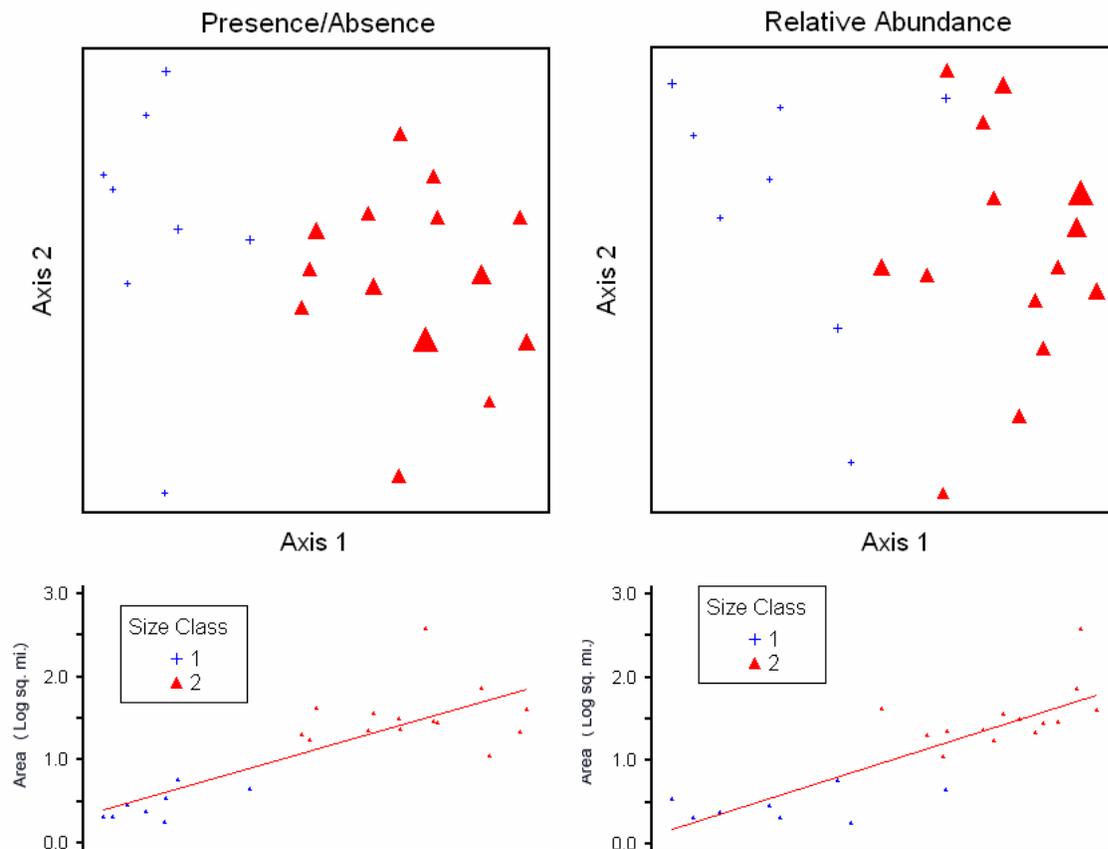


Figure 3-1. NMS ordination of reference data by presence/absence and relative abundance of benthic macroinvertebrates.

Correlation and Regression Analysis

Seventeen (17) reference metrics were significantly correlated with catchment area and two with ecoregion. Only two of the metrics that showed significant correlations with catchment area were considered in index development. The other metrics correlated with catchment area did not discriminate between reference and stressed sites. Small streams tend to have fewer mayfly taxa and a smaller proportion of sensitive EPT individuals than larger streams. Midges and Diptera (taxa and relative abundance of individuals) can be dominant in small reference streams (displacing the sensitive EPTs). The pattern described agrees with CT DEP biologist's observations that small streams can be dominated by midges. Regression analysis was used to derive an adequate adjustment for the metrics so that differences in catchment size would not affect metric and index scoring (Table 3-1, Figures 3-2–3-3). Because of the asymptotic form of the relationship, a regression equation was fit in the form of $b-m(1/x)$ where x is the log of

catchment area. The adjusted metric values are the mean of reference values plus the residual around the predicted value (observed-predicted).

Classification Conclusions

There is evidence that stream catchment size affects benthic macroinvertebrate community composition and metrics in reference streams of Connecticut. No other variables were identified as useful for site classification, though minor effects may be associated with ecoregion and sample collection date. This is confirmed by cluster analysis and DFA. NMS ordination showed that sites greater than 10 square miles are biologically distinct from smaller sites.

Table 3-1. Adjustments to metric values to account for catchment size.

Metric adjustment formula ^a	r ² b
Ephemeroptera Taxa _{adj} = 5.8 + Metric - (7.78-1.54*1/log10(area_sqmi))	0.66
% sensitive EPT _{adj} = 52.1 + Metric - (65.6-10.37*(1/log10(area_sqmi)))	0.26

^a Adjusted metric value = Mean_{Reference} + Metric_{Observed} - Metric_{Predicted}, where predictions are based on regression analysis of reference metric values on catchment size.

^b The r² value reflects the variability accounted for in the predictive, non-linear equation.

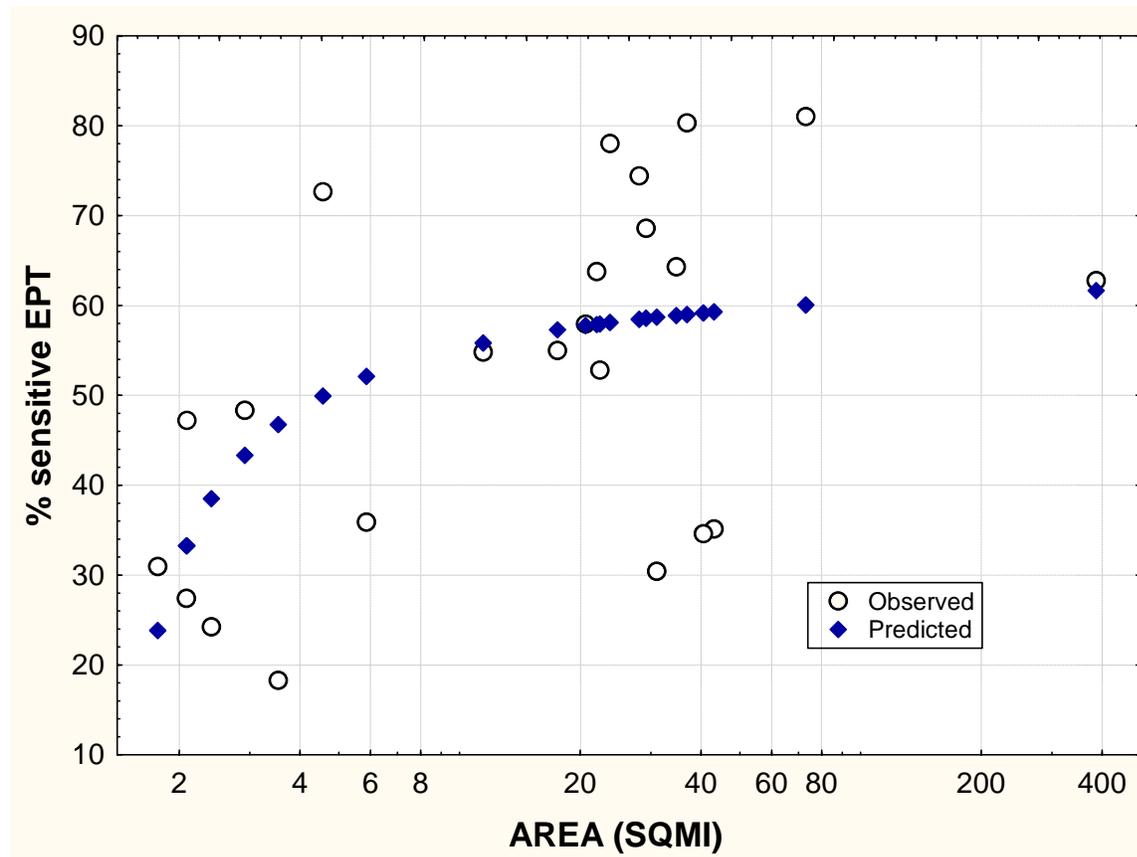


Figure 3-2. Observed and predicted values for the % sensitive EPT metric in reference sites.

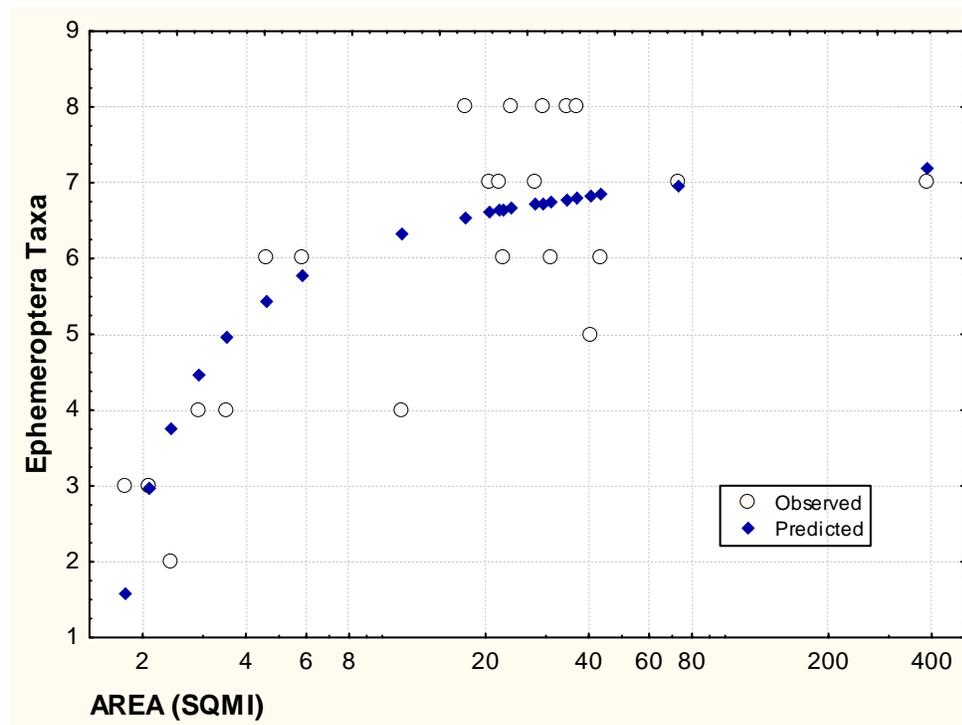


Figure 3-3. Observed and predicted values for the Ephemeroptera taxa metric in reference sites.

Correlation analysis reveals that several metrics are related to catchment area. However, many of the correlated metrics are not the strongest indicators of environmental stress (see Section 6 below). Distinct site classification introduces an artificial “breakpoint” to define the different classes, such as, sites above, or below, 10 square miles in catchment area. However, a site that is 9 square miles may be very similar to a site that is 11 square miles.

A continuous classification system adjusts individual metrics on a site specific basis, as demonstrated in the regression analysis. It is more appropriate than discrete classification in this analytical data set because: continuous classification avoids the artificial threshold between site categories, and it allows application of a single index in all sites, as opposed to applying different indices or different metric scoring scales depending on site class. The classification adjustments are applied only in the limited number of metrics that show effects of natural environmental gradients. It allows analysis of all data combined, which increases the robustness of the index development results compared to separate development processes for a smaller number of samples in each site class.

3.3 BCG Taxa Attributes

Graphical analysis of individual taxa on ordination plots was deemed to be the most useful for identifying attribute groups. The response gradient was defined by ordination analysis, where the least stressed and the most stressed sites were identified in the ordination, and where the least and most stressed sites were well-separated in the ordination (Figure 3-4). These sites thus represent the endpoints of the stressor gradient in ordination space, and the stressor gradient was

parallel to Axis 1 of the NMS ordination. The assumption here is that once natural classification and variability are accounted for, that remaining differences in taxonomic composition are caused by stressors.

As can be seen in Figure 3-5, the Connecticut sites were arranged along the first ordination axis, from least stressed (blue circles), to heavily stressed (black triangles). Slightly stressed sites (light green triangles) overlapped the least stressed, and heavily stressed (red triangles) were adjacent to the severely stressed. The moderately stressed sites covered the entire range from least to severely stressed, showing that responses to intermediate stresses are highly variable.

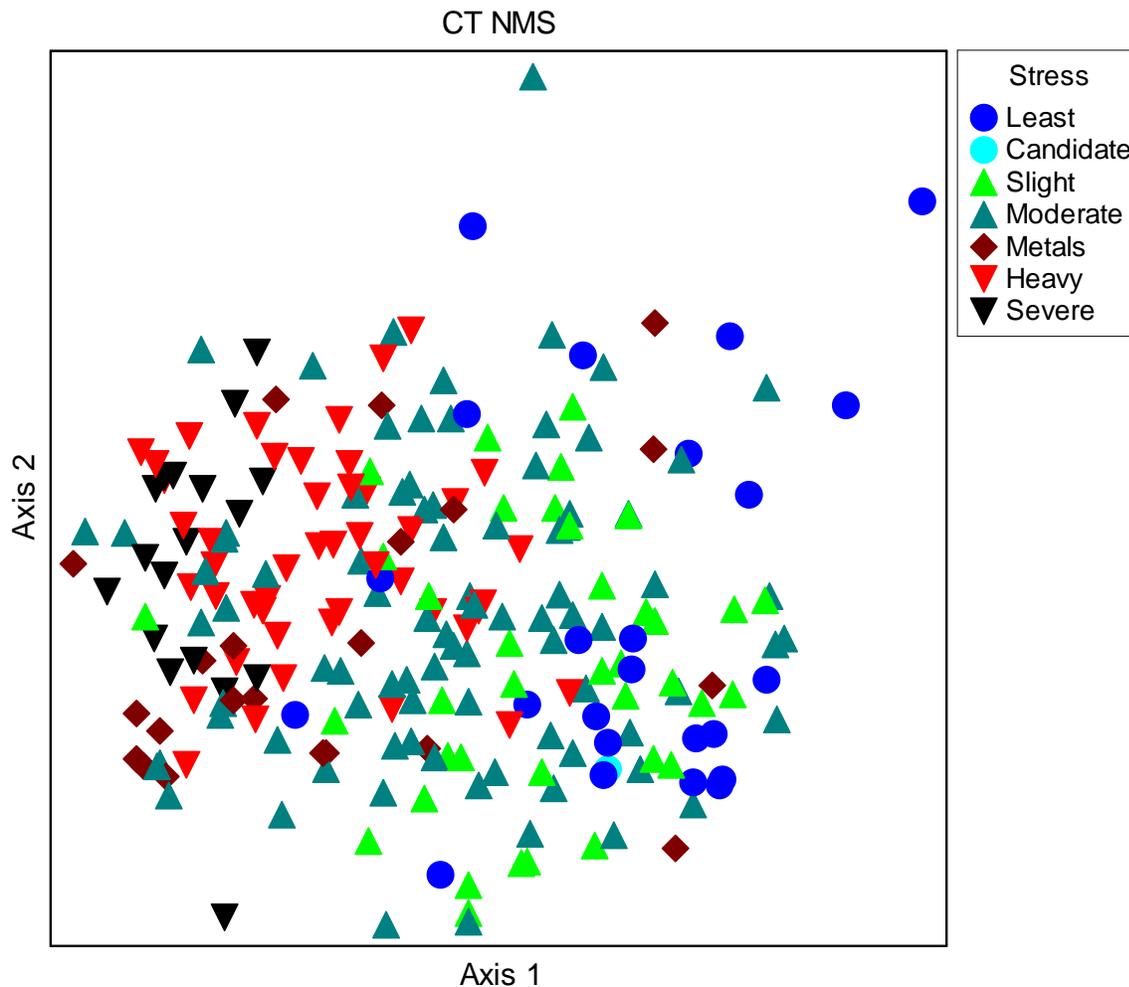


Figure 3-4. Ordination of Connecticut macroinvertebrate assemblages. Symbols identify stress gradient classes, from least stressed to severe stress (see text).

To estimate sensitivity of the taxa to the generalized stressor gradient, the abundance of each taxon was overlaid on the ordination plot, to show whether the taxon occurred in less-stressed or more-stressed sites (Figures 3-5–3-8).

For each taxon shown in Figures 3-5-3-8, symbol size is controlled by the relative abundance of the taxon at each site, so that taxa that are more abundant, or occurred more frequently in any of the *a priori* groups could be readily identified. The graphical analysis was combined with correlation of each taxon abundance with site scores on each ordination axis. Examples of the attribute assignments are shown in Figures 3-5-3-8.

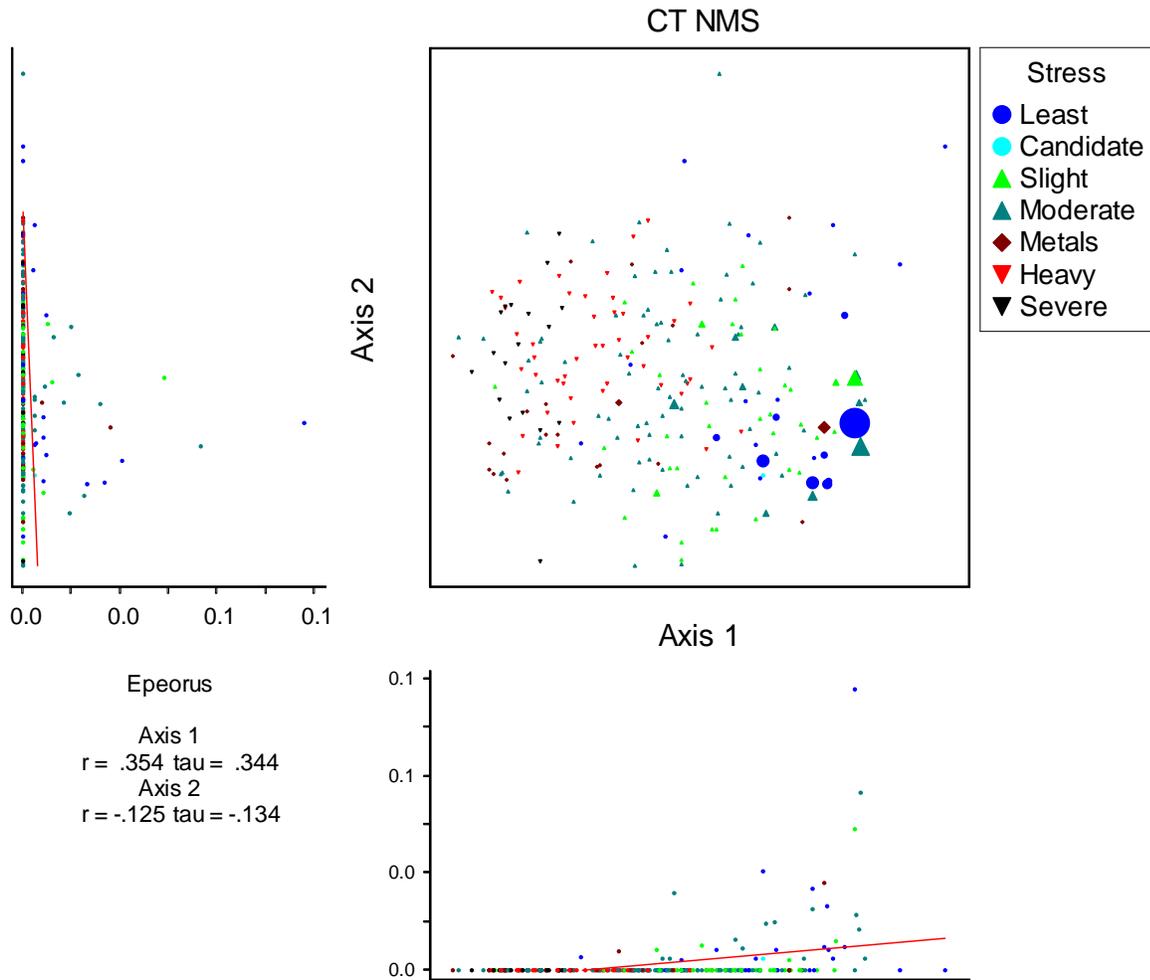


Figure 3-5. Occurrence and relative abundance of *Epeorus* in ordination space. The large plot with large symbols is the NMS ordination plot, with size of the symbols indicating relative abundance of *Epeorus*. The 2 smaller scatterplots to the left and below the large plot are relative abundance of *Epeorus* on each of the ordination axes. Correlation coefficients for *Epeorus* relative abundance are shown for each axis: r is the Pearson correlation coefficient; tau is the Spearman rank-order correlation coefficient. Note that *Epeorus* is most abundant at right-hand values of Axis 1, but has no relationship with Axis 2. This is also reflected in the correlation coefficients. In this figure, Axis 1 defines the stressor gradient, with least stressed sites occurring to the right on axis 1. *Epeorus* occurs in least-stressed (blue) to moderately stressed (dark green) sites, and is never dominant (maximum relative abundance 15%). *Epeorus* is an example of a typical Attribute II taxon (highly sensitive).

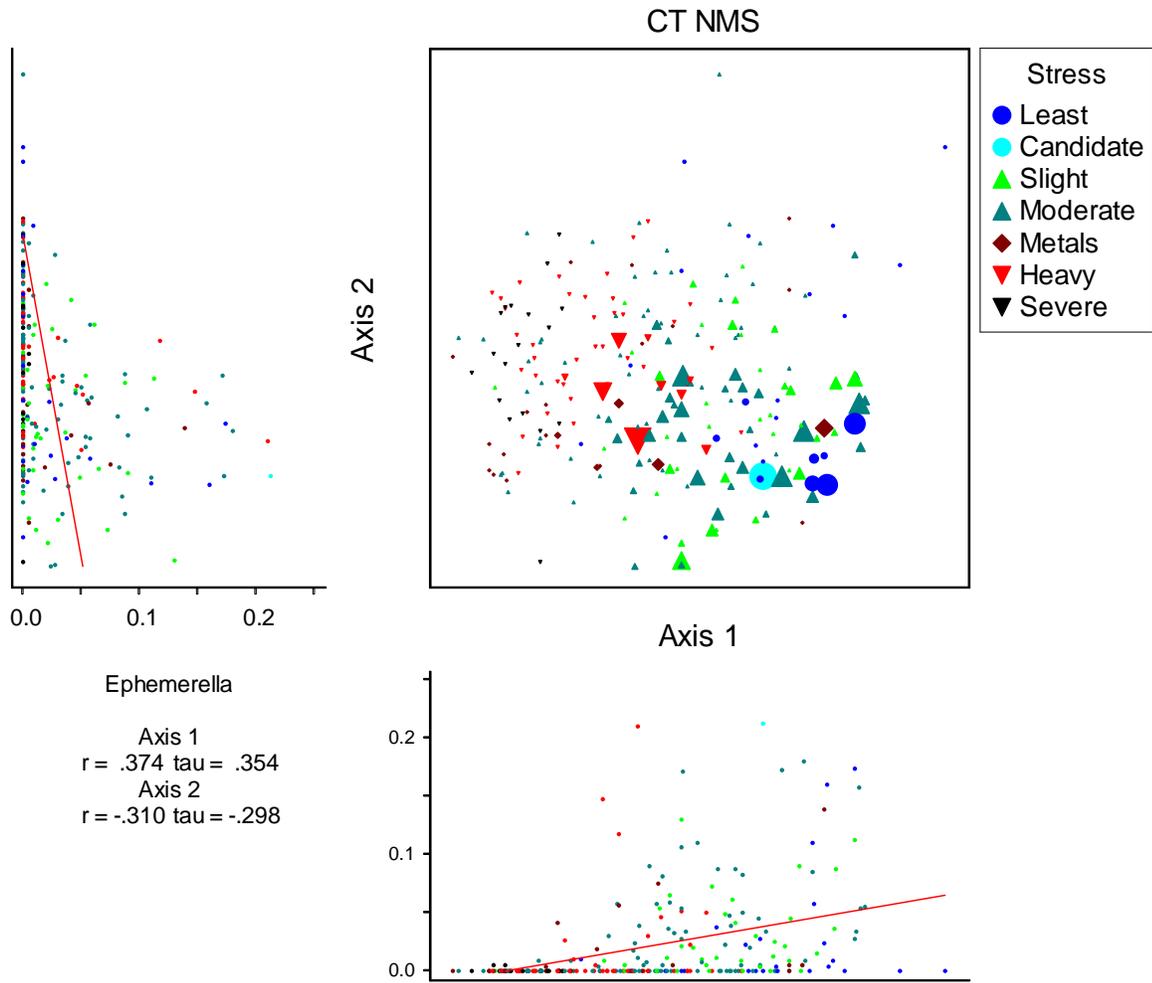


Figure 3-6. Occurrence and relative abundance of *Ephemerebella* in ordination space. See Fig. 8 for explanation. Note that *Ephemerebella* tends to be most abundant and most commonly found in least and slightly stressed sites, but it does occur in a small number of heavily stressed sites. *Ephemerebella* is an example of a typical Attribute III taxon (intermediate sensitive).

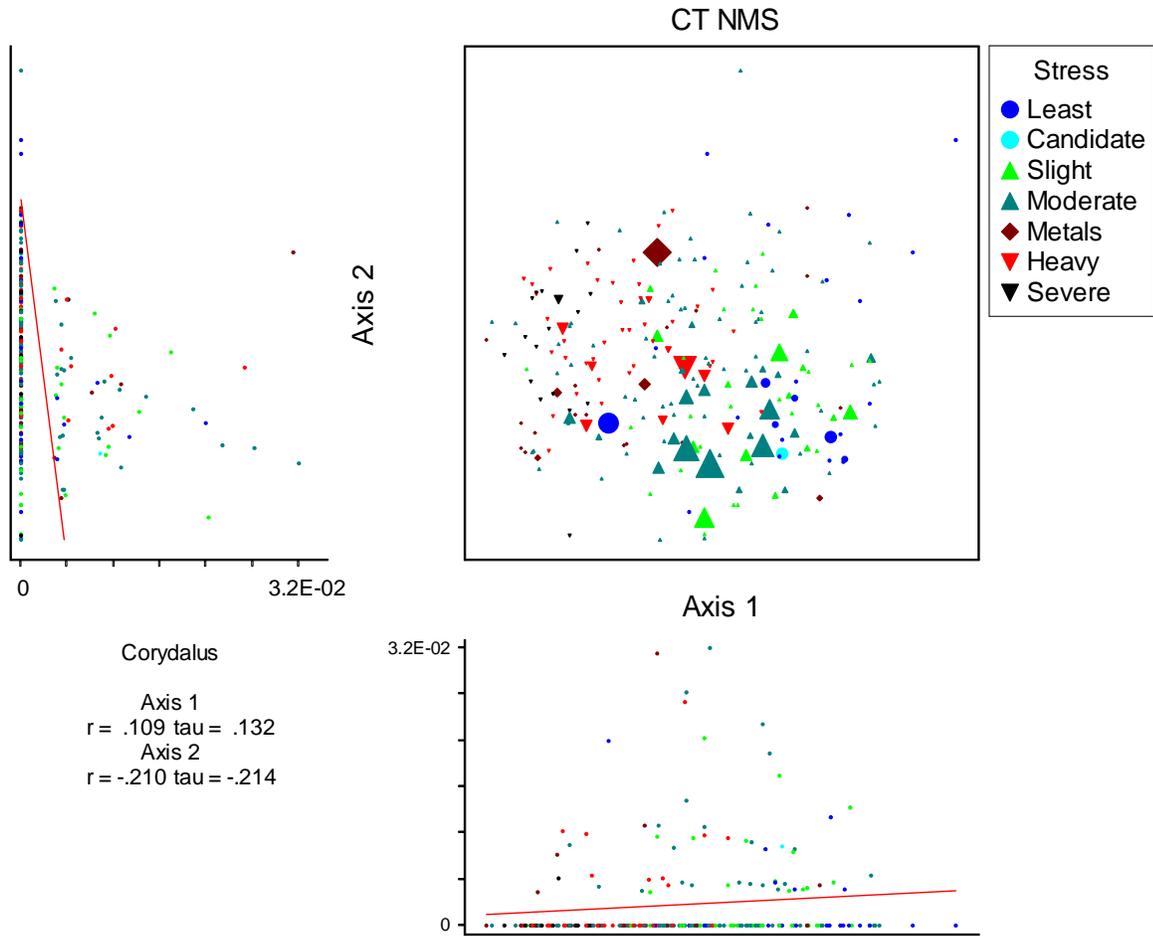


Figure 3-7. Occurrence and relative abundance of *Corydalus* in ordination space. See Fig. 8 for explanation. *Corydalus* is most abundant in moderate stressed sites, but occurs in all stress categories from least-stressed to severe. *Corydalus* is an example of a typical Attribute IV taxon (intermediate-tolerant).

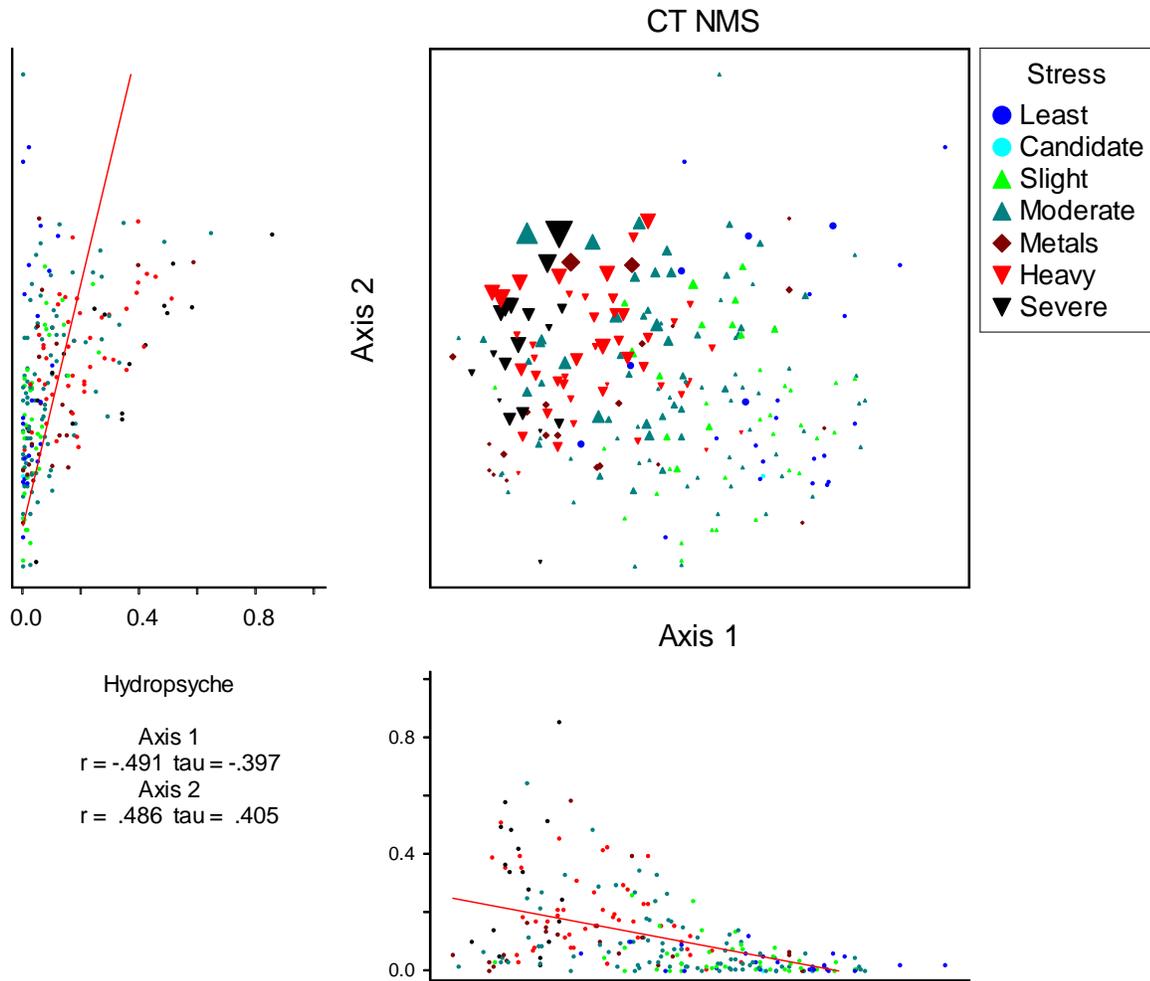


Figure 3-8. Occurrence and relative abundance of *Hydropsyche* in ordination space. See Fig. 8 for explanation. *Hydropsyche* is most abundant in heavily stressed and severely-stressed sites, and is an example of a typical Attribute V taxon (tolerant). The genus occurs at reduced abundance, and reduced frequency, in least-stressed sites.

Breakdown of taxa by attribute group is shown in Table 3-2. The Connecticut taxa list and final attribute assignments are given in Appendix A. Thirty six taxa were left unassigned because participants felt there was insufficient information on the taxa, or they were too rare in the dataset.

Table 3-2. Breakdown of taxa in macroinvertebrate taxa list by attribute group.

Ecological Attribute	Number of taxa	Example Taxa
I Endemic, rare	0	
II Highly Sensitive	30	<i>Hexatoma, Epeorus, Tallaperla, Lepidostoma</i>
III Intermediate Sensitive	68	<i>Psephenus, Nanocladius, Ephemerella, Acroneuria, Taeniopteryx, Dolophilodes, Rhyacophila</i>
IV Intermediate Tolerant	100	<i>Optioservus, Rheotanytarsus, Stenonema, Glossosoma, Chimarra, Physa</i>
V Tolerant	31	<i>Tubificidae, Cricotopus, Ceratopsyche, Cheumatopsyche, Hydropsyche, Gammarus</i>
VI Exotic, Invasive	1	<i>Corbicula</i>
X Unassigned	36	Mostly family identifications; <i>Pseudorthocladius, Nemata</i>

4.0 BIOLOGICAL CONDITION GRADIENT (BCG)

4.1 Site Assignments and BCG level Descriptions

The workgroup examined macroinvertebrate data from 48 high gradient sites and was able to reach consensus on the BCG level assignments for all sites reviewed. Data for each site, notes and decisions are in Appendix B. In some cases, there was discussion and some disagreement on which of two adjacent BCG levels a site should be assigned to. These sites were apparently intermediate, with characteristics of both of the adjacent BCG levels.

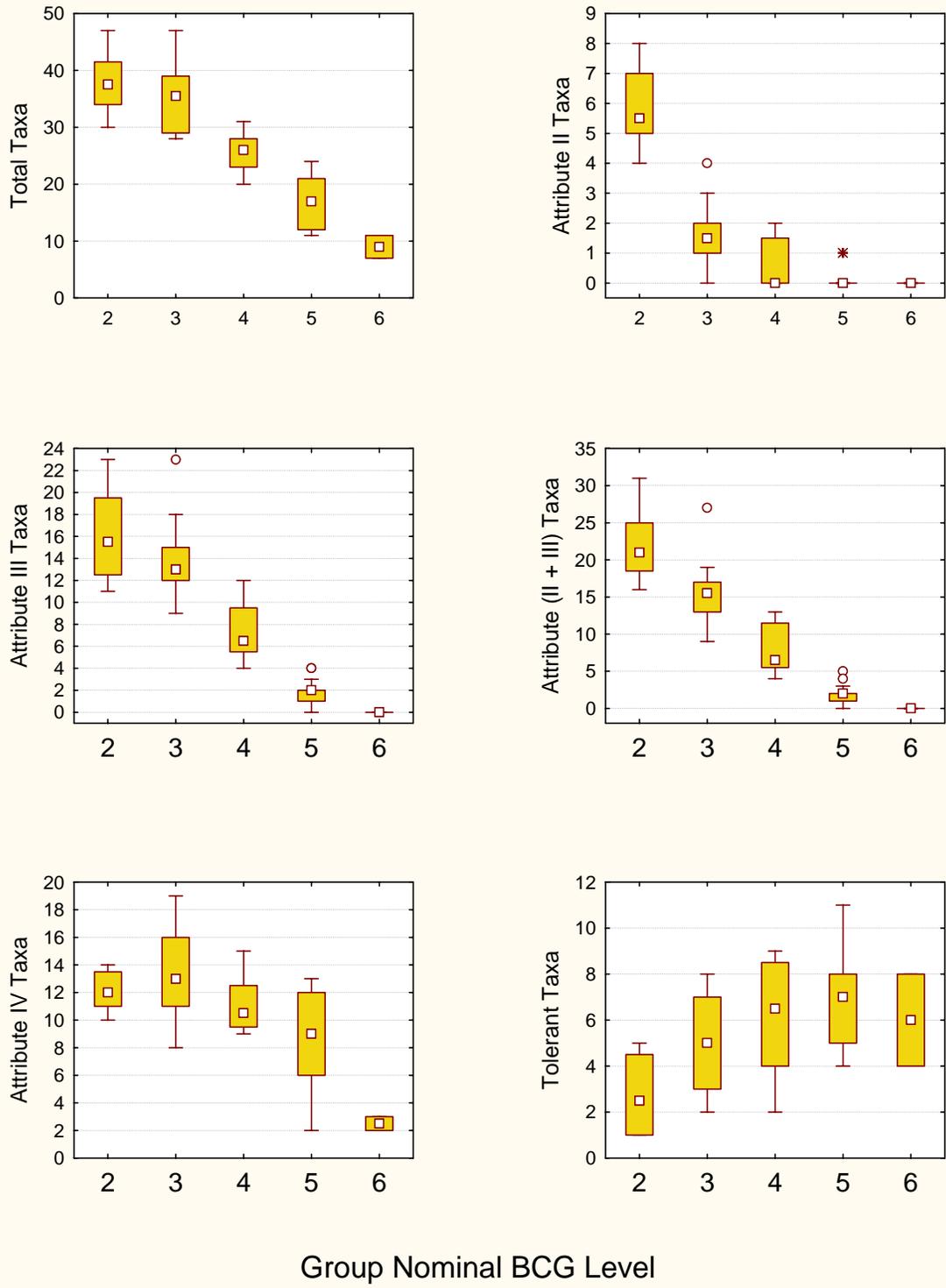
The group was able to distinguish 5 separate BCG levels (BCG levels 2-6). The first BCG level described in Davies and Jackson (2006) consists of entirely pristine sites, and was not included because there was no clear consensus whether BCG level 1 (pristine) sites actually occur in Connecticut. Nevertheless, two of the least stressed sites examined appeared to be candidates for BCG level 1 status. Further examination may be necessary to determine if these sites meet criteria for “minimally disturbed” (Stoddard et al., 2006).

4.2 Operational Rule Development

Examinations of taxonomic attributes among the BCG levels determined by the panel showed that several of the attributes are useful in distinguishing levels, and indeed, were used by the panel’s biologists for decision criteria. Statistical summaries of each attribute and BCG level are given in Table 6, and are shown graphically in Figures 4-1 and 4-2.

The group had developed preliminary decision rules for Connecticut data during the earlier NEWS data analysis (43 sites); however, the NEWS rules were calibrated to the NEWS multihabitat sampling methodology, and not to Connecticut’s riffle-only samples. Using the data distributions shown in Table 4-1 and Figures 4-1 and 4-2, we modified the NEWS rules to reflect the decisions made for sites using Connecticut’s sampling method (Table 4-2). Differences between the current rules and the earlier NEWS rules were minor, consisting of a slight increase of standards for BCG level 2 (one additional Attribute II taxon required, and the proportion of sensitive taxa increased by 15%), and a slight relaxation of standards for levels 3 and 4.

We found that most biologists preferred to use taxon richness within the two sensitive attributes as the first and most important criterion for determining site BCG level assignments. Thus, the number of highly sensitive taxa was most often used to distinguish between BCG level 2 and level 3 sites. BCG level 2 should have several highly sensitive taxa (Attribute II), but their richness may be reduced in level 3. For example, a rule for BCG level 2 is that highly sensitive taxon richness (Attribute II taxon richness) should be more than three to five taxa (Figure 4-1; Table 4-2). BCG level 3 is also discriminated from level 4 by total number of sensitive taxa, and by % sensitive individuals. BCG levels 4 and 5 are discriminated by the almost complete loss of sensitive taxa in level 5 (richness and relative abundance); and concomitant increase in relative abundance of tolerant taxa. The workgroup identified only two BCG level 6 sites in the calibration data set. The level 6 sites were similar to level 6 decisions from the historical data set (analyzed in the NEWS project): few total taxa or extremely low abundance (< 50% of target of 200 individuals in subsample), or extreme dominance by highly tolerant taxa (Attribute V).



Group Nominal BCG Level

Figure 4-1. Box plots of BCG metrics, by nominal BCG level (group majority choice) for 48 assigned sites. Taxon richness metrics. Sensitive taxa is the sum of both Attribute II and Attribute III taxa.

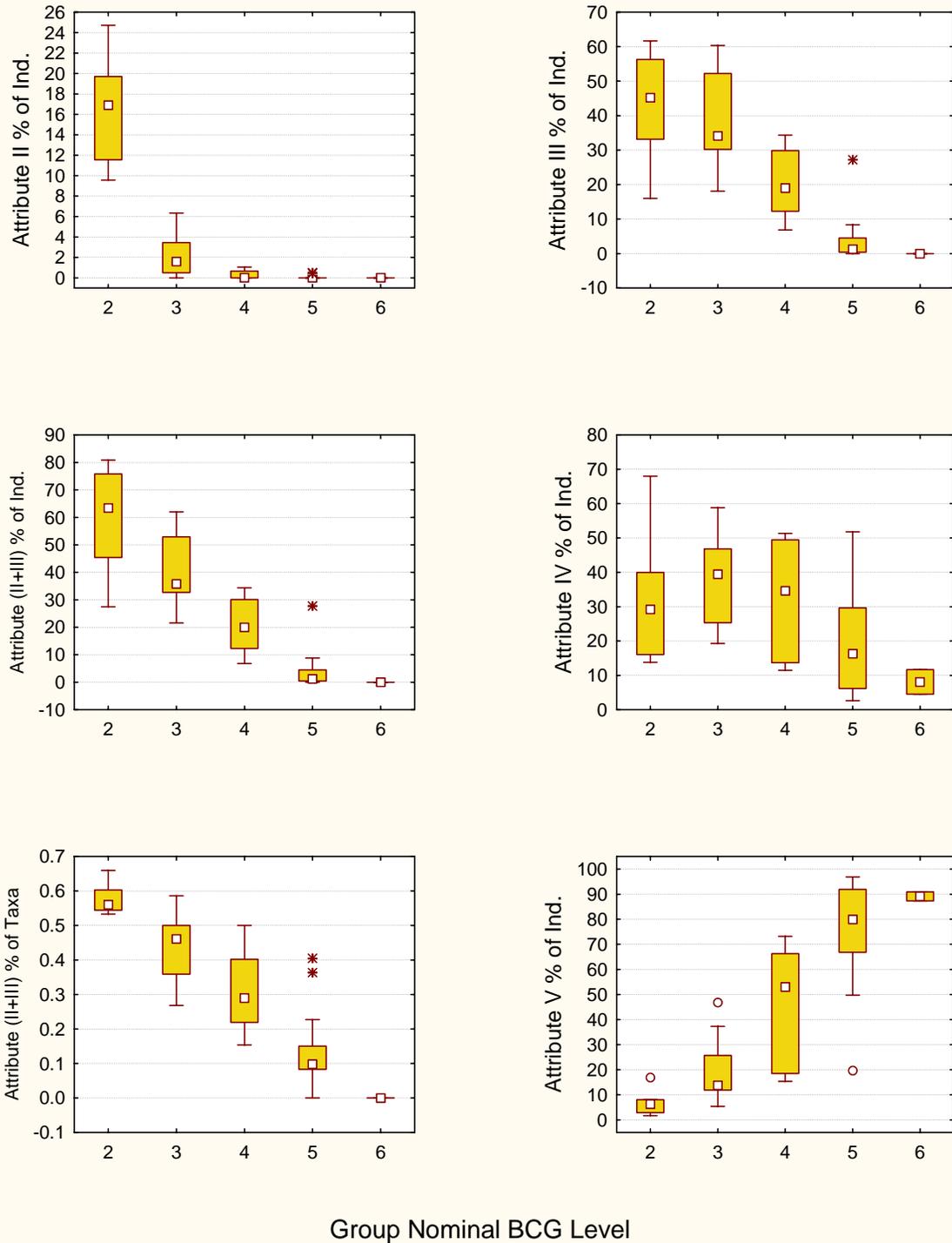


Figure 4-2. Box plots of BCG metrics, by nominal BCG level. Percent metrics.

Table 4-1. Ranges of attribute metrics in Connecticut high gradient samples by group assigned BCG levels.

Attributes	Group assigned BCG level (nominal)					
	1	2 (n=8)	3 (n=15)	4 (n=8)	5 (n=15)	6 (n=2)
0 General		rich 30–47	rich 28–47	rich 20–31	rich 11–24	Rich 7–11 Number: 44–214
I Endemics						
II Highly sensitive taxa		4–8 taxa 10–25%	0–4 taxa 0–6%	0–2 taxa 0–1%	0–1 taxa 0–1%	0 taxa
III Intermediate Sensitive taxa		11–23 taxa 16–61% of indiv.	9–23 taxa 18–60% of indiv.	4–12 taxa 7–34% of indiv.	0–4 taxa 0–8% of indiv.	0 taxa
		II + III: 16–31 taxa II + III: 53–66% of taxa II + III: 27–80% of indiv	II+III: 9–27 taxa II+III: 27–59% of taxa II+III: 22–62% of indiv	II+III: 4–13 taxa II+III: 15–50% of taxa II+III: 7–34% of indiv	II+III: 0–5 taxa II+III: 0–36% of taxa II+III: 0–9% of indiv	0
IV Intermediate Tolerant taxa		10–14 taxa, 14–68%	8–21 taxa, 19–59%	9–15 taxa, 11–51%	2–13 taxa, 3–45%	2–3 taxa, 4–5%
V Tolerant taxa		1–5 taxa, 2–17% Va+Vb: 1–5 taxa, V+Va 2–17% of indiv	2–7 taxa, 5–47% Va+Vb: 2–8 taxa, Va+Vb: 5–47% of indiv.	2–7 taxa, 15–72% Va+Vb: 2–9 taxa, Va+Vb: 15–73% of indiv.	3–7 taxa, 47–96% of indiv. Va+Vb: 4–11 taxa, Va+Vb: 50–97% of indiv.	4–7 taxa, 88–95% of indiv. Va+Vb: 4–8 taxa, Va+Vb: 91–95% of indiv
V.a Highly tolerant taxa		0 taxa IV + V + Vb: 20–73%	0–2 taxa IV + V + Vb: 38–78%	0–4 taxa, 0–8% IV + V + Vb: 66–93%	0–4 taxa, 0–32% IV + V + Vb: 91–100%	0–1 taxa, 0–2% IV + V + Vb: 100%
Indicator Taxa¹		E rich: 3–8 P rich: 2–7 T rich: 6–13 EPT: 15–24 E %: 2–41 Hydro 3–15% Tubi: 0–1% Nonins: 0.5–3%	E rich: 2–7 P rich: 1–6 T rich: 5–13 EPT: 9–20 E %: 1–41 Hydro 5–43% Tubi: 0–12% Nonins: 0–18%	E rich: 2–6 P rich: 1–3 T rich: 6–9 EPT: 10–17 E %: 2–25 Hydro 16–67 Tubi: 0–0.44% Nonins: 2–17%	E rich: 0–3 P rich: 0–1 T rich: 2–7 EPT: 2–11 E %: 0–16% Hydro 2–94% Tubi: 0–0.6% Nonins: 0–43%	E rich: 1 P rich: 0 T rich: 3–4 EPT: 4–5 E %: 2% Hydro 68–89% Tubi: 0–7% Nonins: 3–14%

1. E = Ephemeroptera; P = Plecoptera; T = Trichoptera; Hydro = Hydropsychidae; Tubi = Tubificidae; Nonins = non-insects

Table 4-2. Candidate decision rules for Connecticut High Gradient Streams. Ranges in parentheses denote fuzzy membership function (see 4.4).

Attributes	BCG level					
	1	2	3	4	5	6
0 General		2.1 Total taxa > (25–30) 2.2 count > (50–60%) of target	3.1 Total taxa > (19–23) 3.2 count > (50–60%) of target	4.1 Total taxa > (17–21) 4.2 count > (50–60%) of target	5.1 Total taxa > (8–12) 5.2 count > (50–60%) of target	Total taxa < (8–12) count < (45–55%) of target
I Endemics						
II Highly sensitive taxa		2.3 Taxa II > (3–5)				
III Sensitive taxa		2.4 % Taxa (II+III) > (45–55%) 2.5 % Indiv (II + III) > (30–40%)	3.3 Taxa (II+III) > (8–10) 3.4 % Indiv (II+III) > (30–40%)	4.3 Taxa (II+III) > (3–5) 4.4 % Indiv (II+III) > (10–20%)		
IV Intermediate tolerant taxa		(no rule)	(no rule)	(no rule)	(no rule)	
V Tolerant taxa (all)		2.6 % Indiv V < (10–15%)	3.5 % Indiv V < (40–50%)	4.5 % Indiv V < (65–75%)		
Indicator Taxa		[E taxa > 2]		[E taxa > 0]		
Combining Rule		2.1, 2.2, 2.3, 2.4 and (2.5 or 2.6)	Fails any level 2 rules 2.2-2.6, and 3.1, 3.2, 3.3, and (3.4 or 3.5)	Fails any level 2 rules 2.2–2.6 and fails level 3 rules 3.3–3.5 and 4.1, 4.2, 4.3, and (4.4 or 4.5)	Fails level 2 rules 2.2–2.6, and level 3 rules 3.2–3.5 and level 4 rules 4.2–4.5, and 5.1 and 5.2	Fails all higher levels

4.3 BCG Level Descriptions

Based on the characterization of sites identified as belonging to different BCG levels, we developed a set of linguistic rules for distinguishing levels. More complete description of the levels are in Appendix C.

BCG level 2

- Total Taxon Richness: high
- Total abundance: near target for subsample (200)
- Highly sensitive taxa: At least some taxa are present
- All sensitive taxa (highly sensitive + intermediate sensitive): comprise half or more of all
- All sensitive individuals: comprise 30% or more of all organisms
- Tolerant individuals (tolerant + highly tolerant): a small fraction of all organisms

Based on the group decisions, the BCG level 2 rules 1–4 are “all or nothing”, that is, all must be met for a sample to be considered level 2. Logically, they are combined with AND operators. Rules 5 and 6 are less stringent: one of rules 5 and 6 must be met, but not both (joined with OR operator). The BCG level 2 rules discriminate level 2 from level 3 (and lower).

BCG level 3

- Total Taxon Richness: moderate to high
- Total abundance: near target for subsample

Highly sensitive taxa: may be absent (**no rule**)

- Richness of all sensitive taxa (highly sensitive + intermediate sensitive) is moderate
- All sensitive individuals: comprise 30% or more of all organisms
- Tolerant individuals (tolerant + highly tolerant): less than half of all individuals

As with BCG level 2, the relative abundance rules 4 and 5 are less stringent: one of rules 4 or 5 must be met, but not both (joined with OR operator). The BCG level 3 rules discriminate level 3 from level 4 (and lower).

BCG level 4

- Total Taxon Richness: moderate to high
- Total abundance: near target for subsample

Highly sensitive taxa: may be absent (**no rule**)

- All sensitive taxa (highly sensitive + intermediate sensitive) present but may be low richness
- All sensitive individuals: more than an insignificant fraction

- Tolerant individuals (tolerant + highly tolerant): do not dominate completely

As with BCG levels 2 and 3, the relative abundance rules 4 and 5 are less stringent: one of rules 4 or 5 must be met, but not both (joined with OR operator). The BCG level 4 rules discriminate level 4 from level 5 (and lower). To qualify as BCG level 4, sensitive taxa had to be present in low diversity or higher, and comprise a low proportion, but more than negligible, of all organisms. The range of 10 to 15% relative abundance was deemed the lower bound to qualify as “a functional part of the community” and “more than negligible”.

BCG level 5

BCG level 5 was discriminated from level 4 by a significant reduction of sensitive taxa (Attributes II and III) to the point where they are merely incidental if present and are not a functional part of the community.

- Total Taxon Richness: Not low
- Total abundance: near target for subsample

BCG level 6 was discriminated from level 5 by increasing loss of all taxa or extremely low numbers. The two rules for BCG level 5 discriminate level 5 from level 6: failure of one of these rules means that a sample is assigned to level 6.

The rules are applied as a downward cascade: for a site to be rated as BCG level 2 (the highest described BCG level), all attributes must meet the level 2 condition (Table 4-2). A BCG level 3 rating requires one or more failures of level 2 rules, but the site must meet all minimum level 3 rules. The quantitative rules that follow from the linguistic rules are shown in Table 4-2.

4.4 Automated Decision Criteria Model

In order to develop the decision criteria inference model, each linguistic variable (e.g., “high taxon richness”) must be defined quantitatively as a fuzzy set (e.g., Klir, 2004). A fuzzy set has a membership function, and the membership functions of different classes of taxon richness are shown in Figure 4-3. We used piecewise linear functions to assign membership of a sample to the fuzzy sets shown (Figure 4-3). Numbers below a lower threshold have membership of 0, and numbers above an upper threshold have membership of one, and membership is a straight line between the lower and upper thresholds. For example, in Figure 2-1, a sample with 20 taxa would have a membership of 0.50 in the set “Low-moderate Taxa” and a membership of 0.50 in the set “Moderate Taxa.”

Inference uses the logic statements developed by expert consensus. In “crisp” logic, an AND statement is the same as “Intersection” in crisp set theory, and logical OR is equivalent to set theory “Union”. These are the same in fuzzy logic, however, a fuzzy AND uses the minimum membership of the two sets, and a fuzzy OR uses the maximum (Klir, 2004). For example, we may have a rule “If Highly Sensitive taxa are Moderate AND Sensitive Taxa are High, THEN level is 2.” To illustrate this rule, suppose a sample has membership of 0.25 in

the set: “Highly Sensitive taxa are Moderate” and membership of 0.75 in “Sensitive Taxa are High;” then its membership in level 2 is $\min(0.75, 0.25) = 0.25$. Output of the inference model may include membership of a sample in a single level only, ties between levels, and varying memberships among two or more levels. The level with the highest membership value is taken as the nominal level.

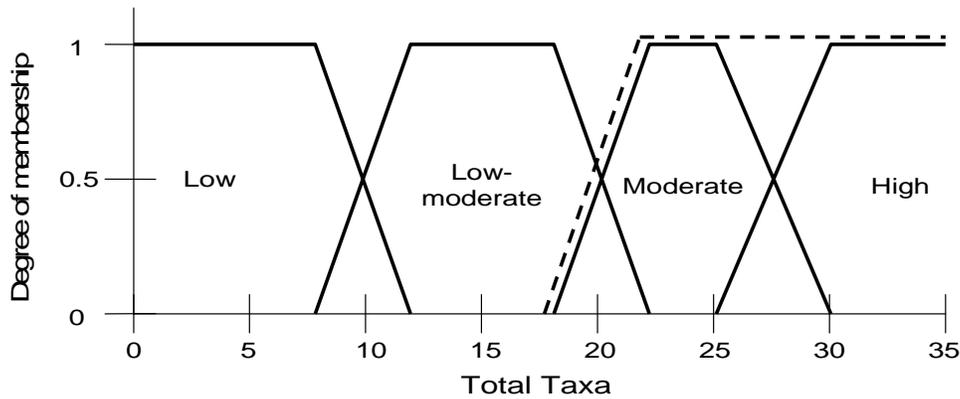


Figure 4-3. Fuzzy set membership functions assigning linguistic values of Total Taxa to defined quantitative ranges. Heavy dashed line shows membership of fuzzy set defined by “Total taxa are moderate to high.”

4.4.1 Model Performance

Model output is membership of a sample in each of the levels described in Table 3-2. In most cases, a single level is given a membership value of 1, and all the rest 0. Often, a single level will have a highest value and one or more other levels will be given a lower, non-zero membership value. We considered any two membership values within 0.1 as a tied decision between the levels.

The fuzzy decision model was developed and calibrated with 48 high-gradient samples rated by the group. The final model treats the levels as a logical cascade from level 2 to level 6: failure of a rule for any level is considered a “success” for the next lower level.

Overall, the decision model matched the panel decisions almost exactly: the nominal level of the fuzzy model exactly matched the workgroup nominal level assignments in 45 of 48 samples (94%). In all 3 cases where the model and the group did not agree, the respective minority and nominal choices were “flipped”. For example, for site CT 40-01, Clark Creek,

the panel's mean BCG level was 3.32, or a high-quality level 3, with a minority selection of level 2. The decision model was exactly tied between level 2 and level 3.

Examination of the disagreements may also reveal inconsistencies by the human assessors; for example, the group may have assessed a sample as level 5 because of a single sensitive taxon among only 7 taxa total, while the rule had required more taxa to qualify for level 5. In other instances, the comparisons revealed the need for refining model calibration.

5.0 MULTIMETRIC INDEX (MMI)

A biological metric is a numerical expression of a biological community attribute that responds to human disturbance in a predictable fashion. Metrics were considered for inclusion in this multimetric index on the basis of discrimination efficiency, low inter-annual or seasonal variability, ecological meaningfulness, contribution of representative and unique information, and sufficient range of values. They were organized into seven categories: richness, composition, evenness, pollution tolerance, BCG attributes (Section 3.3), functional feeding group, and habit (mode of locomotion).

5.1 Metric Methods

A suite of commonly applied, empirically proven, and theoretically responsive metrics was calculated for possible inclusion in a multimetric index. Tolerance metrics were based on both Hilsenhoff tolerance values and BCG taxa attribute groups (Davies and Jackson, 2006). Hilsenhoff tolerance values are on a 0 to 10 scale (most sensitive to most tolerant). The Hilsenhoff scale was derived primarily to address taxa tolerance to organic pollutants (Hilsenhoff, 1987). Attributes associated with taxa for BCG analysis range from sensitive-endemic to pollution tolerant. BCG attributes were assigned to taxa by consensus of a core group of agency biologists during a workshop conducted at DEP on September 6 and 7, 2006.

All richness metrics (e.g., insect taxa and non-insect taxa) were calculated such that only unique taxa are counted. Those taxa that were identified at higher taxonomic levels because of damage or under-developed features were not counted as unique taxa if other individuals in the sample were identified to a lower taxonomic level within the same sample. Genus level taxonomy was expected to provide responsive metrics, so all metrics were calculated at the level. Metrics were calculated in a modified version of the Ecological Data Application System (EDAS), a Microsoft Access application. Once calculated, the metrics were imported into the statistical package Statistica for further analysis.

Discrimination efficiency

Discrimination efficiency (DE) is the capacity of the biological metric or index to detect stressed conditions. It is measured as the percentage of stressed sites that have values lower than the 25th percentile of reference values (Stribling et al., 2000). For metrics that increase with increasing stress, DE is the percentage of stressed sites that have values higher than the 75th percentile of reference values. DE can be visualized on box plots of reference and stressed metric or index values with the inter-quartile range plotted as the box (Figure 5-1). When there is no overlap of boxes representing reference and stressed sites, the DE is greater than 75%. A metric with a high DE thus has a greater ability to detect stress than a metric with a low DE. Metrics with DEs <25% do not discriminate and were not considered for inclusion in the index.

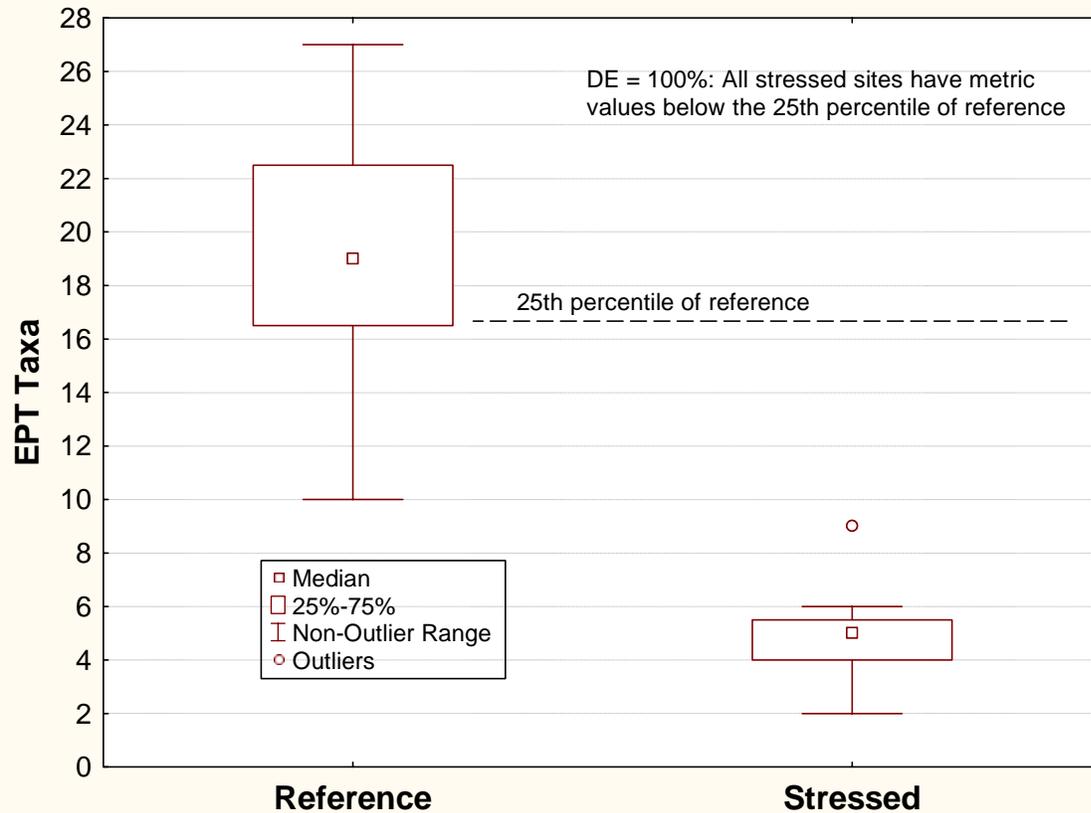


Figure 5-1. Illustration of metric discrimination efficiency (DE) between reference and stressed sites.

Metric variability

When comparing metrics, those with lower variability in the reference sites are preferable to those with higher variability. Variability was measured as the coefficient of variability (CV) in reference sites, calculated as the metric standard deviation over the mean, expressed as a percentage. Lower CVs indicate greater precision of metrics.

Other metric considerations

Ecologically meaningful metrics are those for which the assemblage response mechanisms are understandable and are represented by the calculated value. Ecological meaningfulness is a professional judgment based on theoretical or observed response mechanisms. Those metrics that respond according to expectations established in other studies are more defensible.

Metrics contribute information representative of integrity if they are from diverse metric categories. As many metric categories as practical should be represented in an index so that signals of various stressors can be integrated into the index. While several metrics should be included to represent biological integrity, those that are included should not be redundant with each other. Redundancy was evaluated using Pearson product-moment correlation analysis.

For metrics to discriminate on a gradient of stress, they must have a sufficient range of values. Metrics with limited ranges (e.g., richness of taxa poor groups or percentages of rare taxa) may

have good discrimination efficiency. However, small metric value changes will result in large and perhaps meaningless metric scoring changes.

Comprehensive metrics of sample composition were calculated in addition to the categorical metrics described above. RIVPACS provides a metric which compares the number of taxa expected to occur in reference sites with the number observed in test sites. After adjusting taxa counts for natural site classification variables, the observed to expected ratio (O/E) gives a measure of the degree to which the benthic sample reaches its potential richness. RIVPACS relies on cluster analysis of reference samples to identify naturally distinct community types; discriminant function analysis (DFA) to identify natural determinants of sites in the distinctive biological groupings; and models of expected and observed taxa; based on the predicted membership of a site in a group and of the probabilities of capturing each taxon in each site type. In the analysis, a null model was tested as a baseline for comparison of predictive precision in the final model. The null model does not account for natural variability and the final model does.

5.2 Metric Results

Eighty (80) metrics were calculated in the seven metric categories. Within calibration samples, 51 metrics responded with at least 75 percent of stressed sites worse than the 25th or 75th percentile of reference (Appendix D). Metrics were excluded from consideration in possible index alternatives if they discriminated weakly between reference and stressed sites ($DE < 75\%$), were redundant with more discriminating metrics, had limited ranges of values, or were not representative of the benthic community.

RIVPACS Model

The null RIVPACS model had an O/E mean and standard deviation of 1.00 and 0.21. The final model, accounting for catchment size, had an O/E mean and standard deviation of 1.07 and 0.19. The best RIVPACS models have means near 1.00 and standard deviations less than 0.20, optimally lower than 0.18. The model accounting for catchment size is slightly better than the null model because it has lower standard deviation (better precision). The discrimination efficiency of the final O/E index is 100%. Use of the O/E as an indicator has potential in Connecticut. However, it is not currently applicable because of computational complexities; calculation of O/E for new sites requires substantial software development and technology transfer to DEP. If O/E indices are pursued in the future, it would be worth exploring additional predictor variables, many of which can be obtained through remote (GIS) analysis.

5.3 Multimetric Index Composition

A multimetric index is a combination of metric scores that indicates a degree of biological stress in the stream community (Barbour et al., 1999). Individual metrics are candidate for inclusion in the index if they

- discriminate well between reference and stressed sites;
- are ecologically meaningful (mechanisms of responses can be explained);
- represent diverse types of community information (multiple metric categories); and

- are not redundant with other metrics in the index.

Metrics are scored on a common scale prior to combination in an index. The scale ranges from 0 to 100 and the optimal score is determined by the distribution of data. For metrics that decrease with increasing stress, the 95th percentile of all high gradient data was considered optimal and scored as 100 points. All other metric values were scored as a percentage of the 95th percentile value (Figure 5-2) except those that exceeded 100, which were assigned a score of 100. The 95th percentile value was selected as optimal instead of the maximum so that outlying values would not skew the scoring scale.

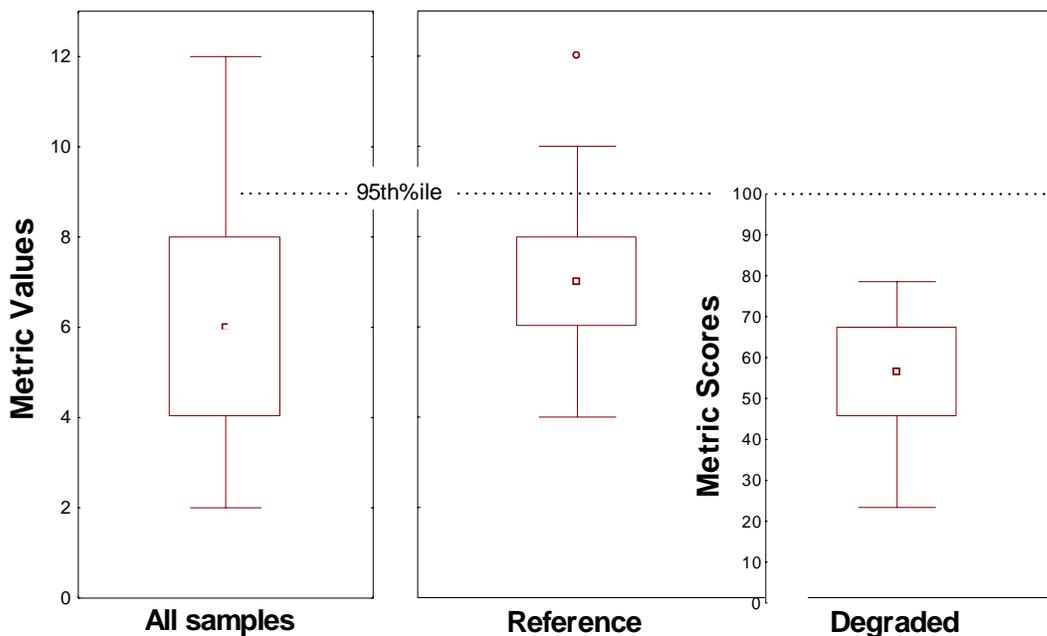


Figure 5-2. Metric scoring schematic for metrics that decrease with increasing stress. For metrics that increased with increasing stress (not shown), the 5th percentile of the data was considered optimal and assigned a value of 100 points, with increasing values scaled down to 0.

5.4 Index Results

Eleven index alternatives were calculated using an iterative process of adding and removing metrics, calculating the index, and evaluating index responsiveness and variability (Table 5-1). The first index alternatives included those metrics that had the highest DEs within each metric category. Subsequent index alternatives were formulated by adding, removing, or replacing one metric at a time from the initial index alternatives that performed well.

The index alternatives did not exhaust the possible combinations of metrics. The few alternatives tested all discriminated reference and stressed sites completely—there was no overlap of index value distributions. Therefore the selection of metrics was more subjective than if there were extreme (or even meaningfully discernible) differences in index performance among index alternatives. The bias in metric selection was for those metrics that performed best

in each metric category, that have precedent in regional bioassessment indices, and that are not redundant either statistically or conceptually. An example of conceptually redundant metrics are the several measures based on BCG attributes (counts of attribute 3 taxa are conceptually redundant with % of attribute 3 taxa). Also, the number of metrics was limited to simplify index application. None of the community model metrics (O/E and PMA) were used in index alternatives for two reasons: to simplify application of the multimetric index and because metrics of this type are usually considered as stand-alone indicators.

The final index alternative is one that met the criteria listed above and that performed best (high DE, low variability) among the alternatives tested. Each alternative index was evaluated based on discrimination efficiency (DE, calculated as for individual metrics), separation of reference and stressed index means as a multiple of the standard deviation reference scores (Z score), variability in reference sites, and verification results.

Three indexes had Z scores above 5. The Z score is an expression of the statistical separation of the reference and stressed sites: the difference between reference mean and stressed mean, divided by the standard deviation of the reference sites. The selected index (#13 in Table 5-1) had a slightly higher Z-score and slightly lower coefficient of variation (CV) than the nearest contenders.

The final index includes the following metrics:

- Ephemeroptera taxa (scoring adjusted for watershed area)
- Plecoptera taxa
- Trichoptera taxa
- Percent sensitive EPT (scoring adjusted for watershed area)
- Scraper taxa
- BCG Taxa Biotic Index
- Percent dominant genus

The selected index has a discrimination efficiency of 100%; all of the highly degraded sites have index values less than the 25th percentile of reference sites (Table 5-1). In fact, all of the degraded index values fall below the minimum of reference values (Figure 5-3). The verification data were comparable to calibration data.

Table 5-1. Index alternatives 1–14, with metrics included in each metric and evaluation statistics.

Metric	DE	1	2	3	4	5	6	7	8	9	10	11	12	13*	14
Total Taxa	100		2	3	4	5	6	7	8	9					
Non-Insect Taxa Percent	83.3	1													
EPT Taxa	100	1							8			11			
Ephemeroptera Taxa	100		2												
Ephemeroptera Taxa (adjusted for watershed area)	100			3	4	5	6	7		9	10		12	13	14
Plecoptera Taxa	100		2	3	4	5	6	7		9	10		12	13	14
Trichoptera Taxa	83.3		2	3	4	5	6	7		9	10		12	13	14
% EPT excluding Hydropsychidae and Baetidae	100	1	2												
% EPT excluding Hydropsychidae and Baetidae (adjusted for watershed area)	100			3	4	5	6	7	8	9	10	11	12	13	14
% Filterer	100	1													
% Predator	100	1	2	3	4			7							
Collector Taxa	100									9					
Scraper Taxa	100		2	3	4	5	6	7	8		10	11	12	13	14
Biotic Index, Individual TALU Attributes	100		2	3	4	5	6	7	8	9	10	11			
Biotic Index, Taxa TALU Attributes	100													13	14
Simpson's Index	100				4										
% dominant 1	91.7		2	3			6			9	10	11	12	13	
% Intolerant	100	1													
DE calibration	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
mean, all ref	74.3	73.0	75.5	75.4	76.9	78.0	74.2	80.1	76.7	77.5	81.0	77.8	77.9	76.6	
stddev, all ref	16.1	12.3	12.1	12.5	12.1	11.5	12.6	12.8	11.4	11.4	12.1	10.7	10.5	11.2	
CV, ref	21.6	16.8	16.1	16.5	15.7	14.8	17.0	16.0	14.9	14.7	14.9	13.7	13.5	14.6	
Cal Ref 25th %ile	56.2	62.2	71.3	71.1	71.0	72.7	69.2	71.7	71.7	72.7	73.0	73.7	72.2	70.0	
mean, all deg	18.8	19.3	20.5	17.9	18.2	21.8	17.2	19.5	22.9	20.5	22.8	22.7	20.9	16.6	
Z-score ((Mean Ref - Mean Deg) / Stdev Ref)	3.5	4.4	4.5	4.6	4.8	4.9	4.5	4.7	4.7	5.0	4.8	5.2	5.4	5.3	

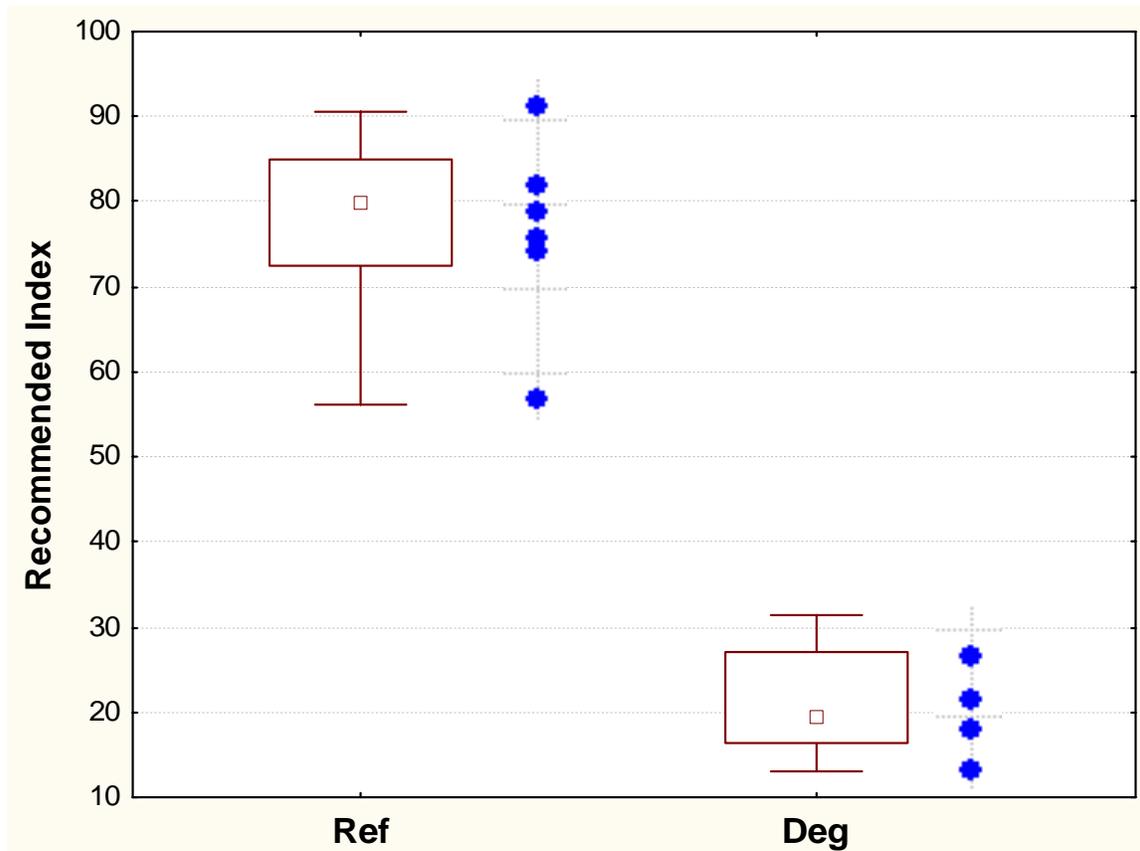


Figure 5-3. Index values in reference and degraded sites. Calibration data are shown in box plots, verification data are shown as individual points.

The BCG taxa biotic index is the average attribute value for all taxa in the sample (attribute 5a was given a value of 6 so that it could be averaged). Performance statistics and scoring formulas of the index metrics (Table 5-2) will allow application and interpretation of the index. Investigators should calculate scores from sample taxa lists (using genus level taxonomy) and average the scores to arrive at the appropriate index value.

Total taxa is in several of the index alternatives, but it was dropped from the final because it is borderline redundant (correlation coefficient near 0.80) with Trichoptera taxa, scraper taxa, and the BCG taxa biotic index. The strongest correlations among index metrics were between BCG taxa biotic index and both Ephemeroptera taxa and Plecoptera taxa, with a correlation coefficient of -0.76 (Table 5-3). This level of redundancy is acceptable.

Table 5-2. Performance statistics and scoring formulas for index metrics.

Metric	CV ¹	DE ²	Response ³	Scoring Formula ⁴
Ephemeroptera taxa ⁵	18.7	100	Dec	= 100*(X + 1.4) / 8.5
Plecoptera taxa	45.5	100	Dec	= 100* X / 6
Trichoptera taxa	33.8	83.3	Dec	= 100* X / 13
% sensitive EPT ⁵	31.3	100	Dec	= 100*(X + 9.2) / 75.2
Scraper taxa	30.1	100	Dec	= 100* X / 11
BCG Taxa Biotic Index	6.3	100	Inc	= 100*(4.6- X) / 1.5
% dominant genus	41.0	91.7	Inc	= 100*(85- X) / 73

¹ CV = Coefficient of Variability = 100*StdDev_{Ref} / Mean_{Ref}.

² DE = Discrimination Efficiency = percentage of degraded samples with metric values outside of the reference quartile range in the direction of response (calibration data only).

³ Direction of metric response with increasing stress, decreasing (Dec) or increasing (Inc).

⁴ The scoring range is between 0 and 100. If formula results in a value outside of the range, reset the score to the nearest extreme of the range.

⁵ See Table 2-3 for metric adjustment prior to scoring

Table 5-3. Correlations (Pearson r) among index metrics.

#	Metric	1	2	3	4	5	6	7
1	Ephemeroptera taxa (adj.)	•						
2	Plecoptera taxa	0.58	•					
3	Trichoptera taxa	0.57	0.50	•				
4	% sensitive EPT (adj.)	0.69	0.54	0.52	•			
5	Scraper taxa	0.67	0.50	0.75	0.52	•		
6	BCG Taxa Biotic Index	-0.76	-0.76	-0.68	-0.74	-0.69	•	
7	% dominant genus	-0.61	-0.54	-0.62	-0.59	-0.60	0.66	•

6.0 DISCUSSION AND CONCLUSIONS

6.1 BCG and MMI Concordance

The MMI uses metrics that are similar in objective to the BCG attributes but calculated somewhat differently (e.g., EPT taxa metrics in the MMI include taxa considered to be Attributes II, III, IV; and Attribute II includes taxa from the EPT orders, as well as a few dipteran and beetle taxa). The total MMI score is based on the average of all metrics, while BCG decisions are based on decision-specific critical attribute groups; e.g., Attributes II and III for the higher levels and Attribute V for lower levels. Concordance of the two assessment endpoints is strong (Figures 6-1–6-2). Figure 6-1 shows the BCG calibration data as rated by the panel, and Figure 6-2 shows the predicted results of the BCG model.

In spite of these differences, MMI scores could be used to separate levels (Figures 6-1, 6-2). Potential MMI scoring thresholds could be as follows:

BCG level	MMI Scoring Range
levels 1, 2	MMI > 75
level 3	75 ≥ MMI > 60
level 4	60 ≥ MMI > 43
level 5	43 ≥ MMI > 20
level 6	20 ≥ MMI

6.2 Variability

Data from a set of 20 sites that had been sampled in multiple years were analyzed for variability. The sites were selected on the basis of greater land use, land cover, and disturbance stability over the sampling period, that is, these sites had no major construction, development, restoration, or discharge upgrades in the period between samples. Some of the sites were protected, and some were in heavily disturbed areas. Stations were sampled 2–8 times over time periods of 1 to 10 years; consecutive samples were separated by 1 to 8 years

The data collected by Connecticut, and the indexes derived from them, show remarkable stability when samples from the same sites are compared among years (Figure 6-3, Table 6-1). The maximum difference within sites was 21 points of the MMI, and 1 level of the BCG. With respect to BCG levels, 16 of the 20 sites were stable within a single level with at most a single observation rated in an adjacent level. Two sites appeared to be intermediate between adjacent levels, with 2 or more observations rated in each level (Eightmile River and Roaring Brook).

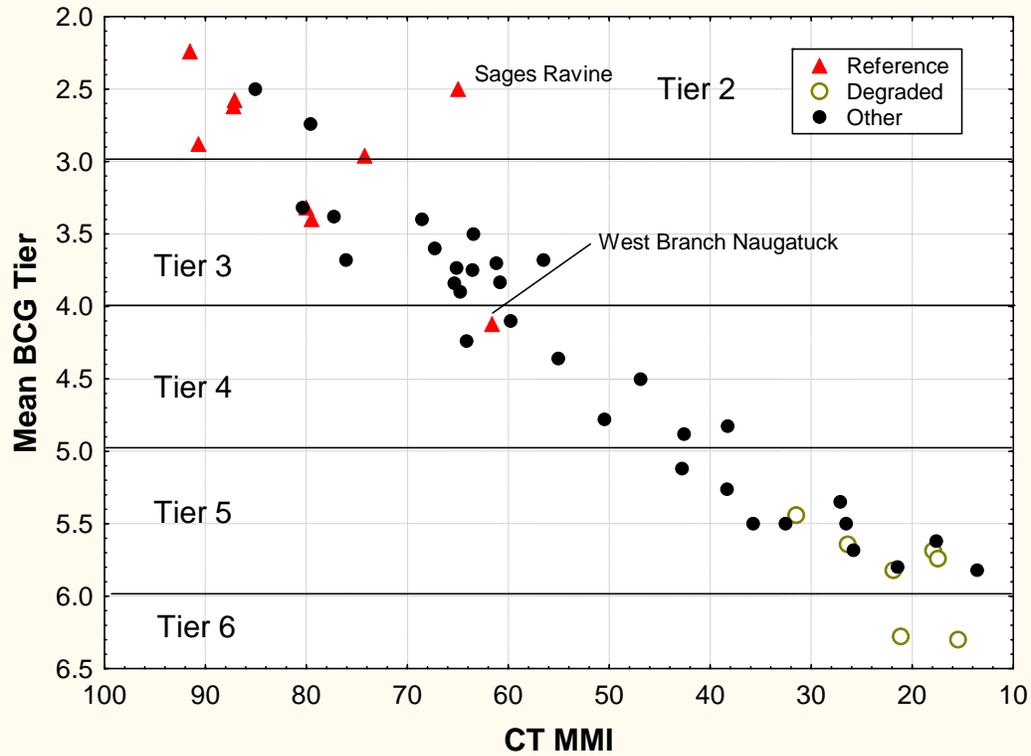


Figure 6-1. Comparison of index values and mean BCG levels (from group development), with outliers labeled. Horizontal lines indicate nominal BCG levels.

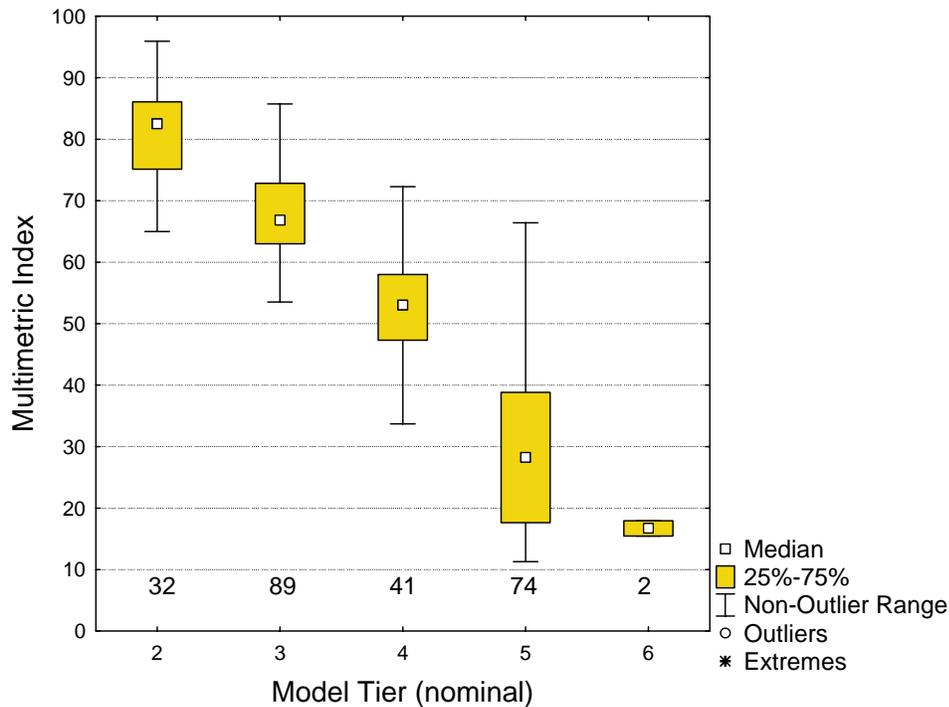


Figure 6-2. Connecticut Multimeric index by BCG levels, estimated from decision analysis model. Number of samples given below boxes.

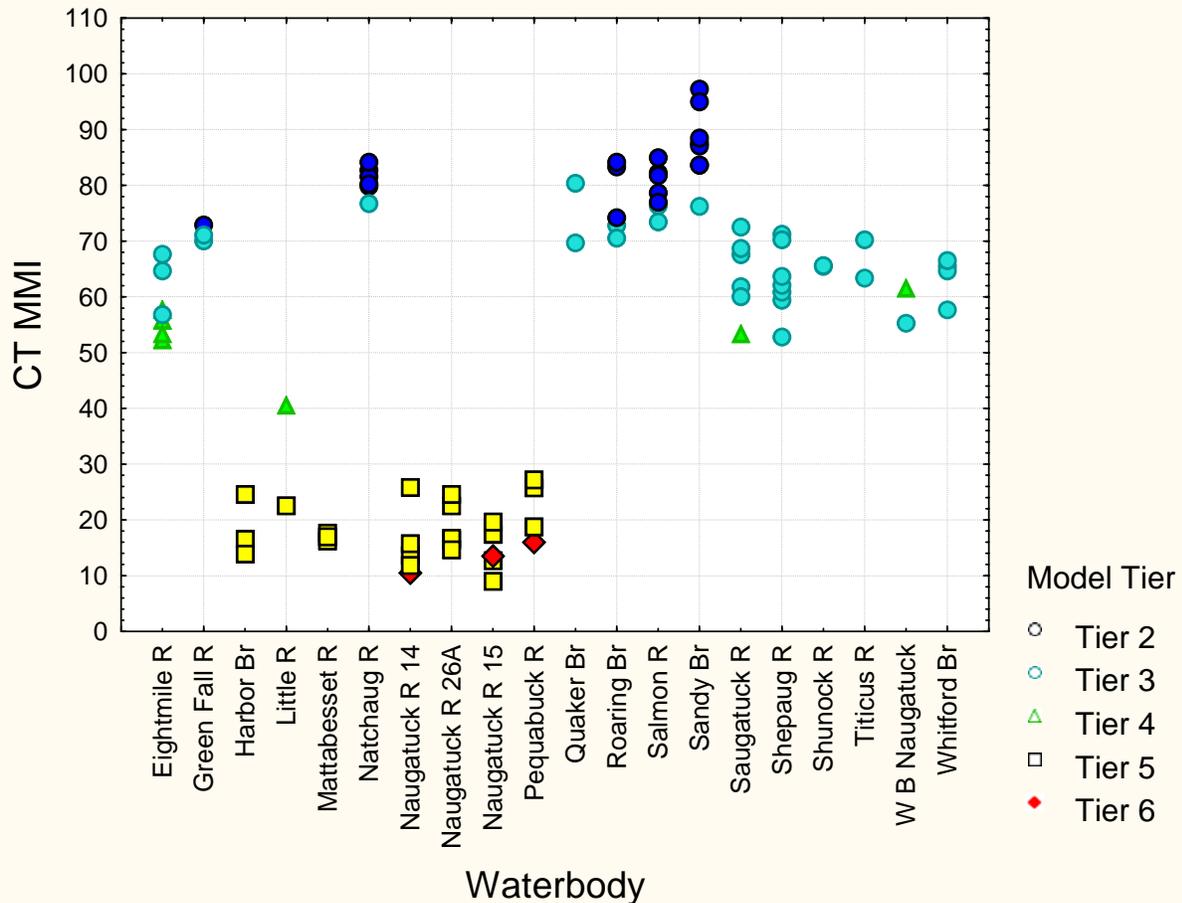


Figure 6-3. Nominal BCG assessments and IBI scores for 20 sites with repeated samples.

Table 6-1. Variability of indexes and metrics from multiyear observations

Quantity	Range	s.d.	Approx. 95% C.I.
BCG Nominal level	2 - 6	0.396	0.78
BCG Average level	2 - 6	0.382	0.75
Multimetric Index	0 - 100	5.41	10.6
% Dominant taxon	0 - 100	11	21.56
Plecoptera taxa	0 - 8	1.08	2.12
Scraper taxa	0 - 14	1.24	2.43
Ephemeroptera taxa	0 - 12	0.994	1.94
% Sensitive EPT	0 - 90	10.2	19.99
Trichoptera taxa	2 - 18	1.426	2.79
BCG Attribute index	2 - 6	0.141	0.276

The repeated observations (Figure 6-3, Table 6-1) demonstrate that the Connecticut methodology has low inherent variability in measurements taken among years. Each of the metrics has a standard deviation that is approximately 7–10% of the expected range of the metric. Such stability among years, at sites with no known changes in anthropogenic sources and stressors,

indicates that the methods and analysis are stable over time. Natural variability is confined to a single BCG level, or about 20 points on the MMI scale. Demonstration that a site has consistently shifted a single BCG level, or 20 MMI points, is positive indication of a change in stress level at the site (increase or decrease), beyond that expected due to natural variability.

We also examined whether there was an association between the biological scores and potential drought stress in the streams. We used the Palmer hydrological drought Index (PHDI), obtained from the National Climatic Data Center (www.ncdc.noaa.gov). There were no severe drought years in the data set, and there was no association between biological scores (MMI or BCG level) and the PHDI, within the range of the PHDI observed in these streams (-1.5 to + 3.5; or mild drought to moderately wet).

6.3 The BCG as an Assessment Tool

The BCG, as developed conceptually in Davies and Jackson (2006), addresses several limitations of existing biotic indexes. Advantages of the BCG include (Gerritsen et al., submitted):

- The BCG is based on ecological considerations with wide expert agreement, rather than on empirical analysis of a particular data set. It is calibrated using a data set, but the result is intended to be more general than a regression analysis of biological response to stressors.
- The BCG uses universal attributes (Attributes I to VI) that are intended to apply in all regions. Specifics of the attributes (taxon membership, attribute levels indicating good, fair, poor, etc.) do vary across regions and stream types, but the attributes themselves and their importance are consistent.
- The BCG requires descriptions of the classes or levels, from pristine to degraded. While requiring extra work, this ensures that future information and discoveries can be related back to the baseline level descriptions. Levels are not perfect or static—they will be altered by increase in knowledge.

The BCG may be more robust than current indexes because it allows, in some cases, for nonlinear responses. The BCG is not conceptually tied to “best available” sites as a reference condition: although best available sites are used as a practical ground truth, it is recognized at the outset that these sites are typically less than pristine, and may be a lower level (e.g., 2, 3, 4).

Implementation will be made more consistent with automated methods to assign sites to levels. At the simplest level, an IBI score range or metric, or a RIVPACs-based observed/expected score range (e.g., Barbour et al., 1999) can be divided into classes corresponding to levels developed here. The IBI and RIVPACs models characterize a gradient, but they do not necessarily reflect the professional consensus that goes into the level descriptions. An alternative is to develop a scoring model that replicates the professional consensus in the level descriptions, either by statistical inference (e.g., Davies et al. 1993) or by direct replication of the rules.

6.4 The BCG and Aquatic Life Use

The terms “Use”, “Designated Uses”, and “Aquatic Life Use” have specific meanings for water quality management in the context of the Clean Water Act. A state defines the uses for its waters, and develops physical, chemical and biological criteria to protect those uses. TALUs are aquatic life uses that are matched more closely to the Designated Uses, rather than a single one-size-fits-all aquatic life use (EPA, 2005).

The BCG is a scientific model of biological condition of waters, set on a universal scale from natural and undisturbed (BCG level 1) to completely biologically and ecologically disrupted (BCG level 6). The BCG, as a universal yardstick, is intended to be used in setting biological criteria to match specific TALUs. It is important to note that levels of the BCG are NOT equivalent to TALUs. The BCG is a scientific measurement yardstick only; it does not express policy decisions and breakpoints for designated uses.

Designated Uses are intended to be set at the highest attainable use for a water body, taking into account natural limitations or irreversible physical (infrastructure) alterations to the habitat or watershed (e.g., existing urban infrastructure, flood control, harbor facilities, irrigation, etc.). Infrastructure is not always irreversible: roads can be rerouted, many older dams and obstructions are being removed from streams, habitat can be restored, etc. Designated uses thus also include potential quality or condition that may not currently be attained, but could be attained with appropriate controls or restoration. Thus, Aquatic Life Uses must be set according to the biological potential of waterbodies, not according to their current condition.

The BCG, as a scientific measurement tool, does not explicitly include assessment of potential condition. The restoration potential of an impaired water body can only be predicted scientifically if the cause of impairment is known, that is, if we know what to do to restore the waterbody, and hence, we know which stressors will be reduced. Setting of designated uses, which implies a restoration goal, does not require causal analysis, because the designated uses are societal decisions on the desired state of a waterbody, and are determined by existing highest uses and watershed/habitat alterations deemed irreversible by the state (and existing as of 1976).

The levels of the BCG are biologically recognizable stages in condition of stream waterbodies. As such, they can form a biological basis for criteria and regulation of a state’s waterbodies. Current thresholds of biocriteria in many states (usually an IBI score, or something similar) are relatively low (e.g., level 4-level 5), and fail to protect outstanding condition waters (levels 1 and 2), or even good condition waters (level 3). Thus, biocriteria set at a lower BCG level will allow incremental degradation of waterbodies to the regulatory level.

The BCG provides a powerful approach for an operational monitoring and assessment program, for communicating resource condition to the public and for management decisions to protect or remediate water resources. The BCG and the calibrated decision system allow practical and operational implementation of multiple aquatic life uses in a state’s water quality criteria and standards. Adoption of the BCG as an assessment tool in the context of multiple Aquatic Life Uses (Tiered Uses) yields the technical tools for protecting the state’s highest quality waters, as well as developing realistic restoration goals for urban and agricultural waters.

Connecticut could use the BCG, or an IBI index calibrated to the BCG, to identify biological expectations for tiered aquatic life uses. Several of the stream sites in least-stressed catchments in this report were rated a BCG level 2 by the panel of biologists. The least-stressed catchments may also correspond to Outstanding or Exceptional waters (this would need to be confirmed). Accordingly, the biological criterion for these waters could be BCG level 2 or better.

6.5 Technical Recommendations

The Connecticut BCG is promising as a basis for decision criteria for Tiered Aquatic Life Use development. Results reported here should be confirmed and reviewed to address discrepancies between the BCG and MMI, and to examine outlier sites. The BCG description and rules can be tested and recalibrated, and Connecticut's monitoring program can be strengthened to support use of the BCG:

Develop BCG for low gradient streams. Connecticut's sampling and assessment has been developed and calibrated for high-gradient streams, because these are by far the dominant stream resource. Nevertheless, low gradient streams are common in coastal areas, and occur throughout the state. They are dominated by sandy stream bottom, and principal invertebrate habitat consists of snags, root wads, undercut banks, and emergent vegetation. Standardized high-gradient methods (mid-stream sampling) applied to low-gradient streams often results in low richness and dominance of organisms adapted to fine sediment. Development of a BCG for the low gradient streams would require a data set sampled with low-gradient methods (Barbour et al., 1999), with similar numbers of samples in reference and highly-stressed sites.

Develop BCG for a second assemblage. Two assemblages do not respond to stressors in exactly the same way and would not always agree in assessments. Fish may not be sampled (nor need to be) at all locations where benthic macroinvertebrates are sampled – assessments can be made from benthic macroinvertebrates alone, but the fish (or other assemblages) may indicate problems or issues that the invertebrates do not. Assessments based on fish are less reliable in small headwater streams (Yoder and Rankin, 1995). In Ohio, full attainment requires that both assemblages and all indexes meet the biocriteria, nonattainment of one but not both assemblages results in a rating of “partial attainment” (Yoder and Rankin, 1995).

Search for level 1 sites. No sites sampled met the NEWS criteria for Minimally Disturbed (Snook et al., 2007; Stoddard et al., 2006). Biological and ecological attributes for level 1 should also include a second assemblage of the spatial attributes, (IX and X), and the endemic-rare species attributes (Attribute I). Candidate minimally disturbed sites could be found in neighboring states (Massachusetts, New York, northern Pennsylvania).

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APPENDIX A

CONNECTICUT INVERTEBRATE TAXA LIST AND ATTRIBUTE ASSIGNMENTS

CT Data

Class or Order	Family	Genus	CT Data			
			BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Hirudinea	Erpobdellidae		5.5	8	1	1
Hirudinea	Erpobdellidae	Erpobdella	5.5	8	89	18
Hirudinea	Erpobdellidae	Nephelopsis	5.5	8	1	1
Hirudinea	Glossiphoniidae	Glossiphonia	5.5	6	1	1
Hirudinea	Glossiphoniidae	Helobdella	5.5	10	1	1
Oligochaeta			x	9	4	3
Oligochaeta	Enchytraeidae		3	8	24	14
Oligochaeta	Lumbricidae		x	10	2	1
Oligochaeta	Lumbricidae	Lumbricina	4	8	51	5
Oligochaeta	Naididae		4	8	23	4
Oligochaeta	Naididae	Dero	x	10	1	1
Oligochaeta	Naididae	Nais	4	8	19	12
Oligochaeta	Naididae	Pristinella	x		7	1
Oligochaeta	Tubificidae		5	10	362	84
Oligochaeta	Tubificidae	Aulodrilus	5	8	8	1
Oligochaeta	Tubificidae	Limnodrilus	5	10	20	1
Oligochaeta	Lumbriculidae		4	8	326	55
Acari			4	4	28	20
Acari	Lebertiidae	Lebertia	4	4	6	1
Acari	Limnocharidae	Rhyncholimnochares	4	4	3	3
Acari	Sperchonidae	Sperchon	4	4	6	5
Acari	Sperchonidae	Sperchonopsis	4	4	9	6
Acari	Torrenticolidae	Torrenticola	4	4	6	6
Collembola			x	5	2	1
Coleoptera	Dryopidae	Helichus	4	5	6	5
Coleoptera	Dytiscidae	Dytiscidae	4	5	1	1
Coleoptera	Elmidae	Ancyronyx	4	6	12	12
Coleoptera	Elmidae	Dubiraphia	4	6	58	24
Coleoptera	Elmidae	Elmidae	4	4	67	23
Coleoptera	Elmidae	Macronychus	4	4	40	29
Coleoptera	Elmidae	Microcylloepus	4	3	49	23
Coleoptera	Elmidae	Optioservus	4	3	1913	210
Coleoptera	Elmidae	Oulimnius	3	4	425	110
Coleoptera	Elmidae	Promoresia	3	3	549	99
Coleoptera	Elmidae	Stenelmis	4	5	1239	174
Coleoptera	Hydrochidae	Hydrochus	5		1	1
Coleoptera	Hydrophilidae	Berosus	5		22	11
Coleoptera	Psephenidae	Ectopria	3	5	99	50
Coleoptera	Psephenidae	Psephenus	3	4	1748	177
Coleoptera	Ptilodactylidae	Anchytarsus	3	2	76	23
Diptera			x		8	4
Diptera	Athericidae	Atherix	3	2	131	50
Diptera	Ceratopogonidae	Bezzia	3	6	17	16
Diptera	Ceratopogonidae	Ceratopogoninae	3	6	1	1
Diptera	Chironomidae		x	7	342	69
Diptera	Chironomidae	Brillia	4	5	39	31
Diptera	Chironomidae	Cardiocladius	5.5	5	54	26
Diptera	Chironomidae	Chaetocladius	x		1	1
Diptera	Chironomidae	Chironomini	4	7	6	4
Diptera	Chironomidae	Chironomus	5	10	8	6

CT Data

Class or Order	Family	Genus	BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Diptera	Chironomidae	Cladotanytarsus	5	7	10	6
Diptera	Chironomidae	Corynoneura	3	7	5	5
Diptera	Chironomidae	Cricotopus	5.5	7	321	90
Diptera	Chironomidae	Diamesa	4	5	88	31
Diptera	Chironomidae	Diamesinae	4	5	2	1
Diptera	Chironomidae	Dicotendipes	5.5	8	26	14
Diptera	Chironomidae	Diplocladius	4	8	63	18
Diptera	Chironomidae	Eukiefferiella	3	8	83	36
Diptera	Chironomidae	Glyptotendipes	5	10	2	1
Diptera	Chironomidae	Heterotrissocladius	3	0	21	5
Diptera	Chironomidae	Labrundinia	5	7	1	1
Diptera	Chironomidae	Limnophyes	5	8	2	2
Diptera	Chironomidae	Lopescladius	x		1	1
Diptera	Chironomidae	Microchironomus	x	8	1	1
Diptera	Chironomidae	Micropsectra	3	7	98	20
Diptera	Chironomidae	Microtendipes	4	6	345	94
Diptera	Chironomidae	Nanocladius	3	3	175	71
Diptera	Chironomidae	Neostempellina	x	8	7	6
Diptera	Chironomidae	Orthoclaadiinae	4	6	26	16
Diptera	Chironomidae	Orthoccladius	4	6	293	99
Diptera	Chironomidae	Parachaetoccladius	3	2	116	26
Diptera	Chironomidae	Parachironomus	5	10	1	1
Diptera	Chironomidae	Paracricotopus	x	6	2	2
Diptera	Chironomidae	Parakiefferiella	4	7	5	4
Diptera	Chironomidae	Parametriocnemus	4	5	158	68
Diptera	Chironomidae	Paraphaenoccladius	4	4	20	3
Diptera	Chironomidae	Paratanytarsus	4	6	10	8
Diptera	Chironomidae	Paratendipes	4	8	13	6
Diptera	Chironomidae	Phaenopsectra	4	7	4	3
Diptera	Chironomidae	Polypedilum	4	6	1004	123
Diptera	Chironomidae	Potthastia	4	2	10	10
Diptera	Chironomidae	Procladius	5	9	2	2
Diptera	Chironomidae	Pseudochironomus	4	5	1	1
Diptera	Chironomidae	Pseudorthoccladius	x	0	32	12
Diptera	Chironomidae	Psilometriocnemus	4	4	10	2
Diptera	Chironomidae	Rheocricotopus	3	6	7	7
Diptera	Chironomidae	Rheotanytarsus	4	6	929	153
Diptera	Chironomidae	Stempellinella	3	4	6	4
Diptera	Chironomidae	Stenochironomus	3	5	20	13
Diptera	Chironomidae	Stictochironomus	4	9	27	3
Diptera	Chironomidae	Stiloccladius	3	6	21	12
Diptera	Chironomidae	Sublettea	4	6	3	3
Diptera	Chironomidae	Sympotthastia	x	2	1	1
Diptera	Chironomidae	Synorthoccladius	5.5	2	8	6
Diptera	Chironomidae	Tanypodinae	5	7	2	2
Diptera	Chironomidae	Tanytarsus	4	6	132	39
Diptera	Chironomidae	Thienemanniella	4	6	8	8
Diptera	Chironomidae	Thienemannimyia	4	7	105	66
Diptera	Chironomidae	Tribelos	4	5	2	2
Diptera	Chironomidae	Trissocladius	x	5	1	1

CT Data

Class or Order	Family	Genus	BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Diptera	Chironomidae	Tvetenia	4	5	303	101
Diptera	Chironomidae	Unniella	4	4	2	2
Diptera	Chironomidae	Xenochironomus	2	0	7	6
Diptera	Chironomidae	Xestochironomus	x	7	4	3
Diptera	Dolichopodidae		x	4	4	4
Diptera	Empididae		4	6	6	4
Diptera	Empididae	Chelifera	4	6	5	5
Diptera	Empididae	Hemerodromia	5	6	237	93
Diptera	Empididae	Neoplasta	x	6	1	1
Diptera	Empididae	Oreogeton	x	6	1	1
Diptera	Empididae	Wiedemannia	x		1	1
Diptera	Muscidae	Limnophora	x	5	1	1
Diptera	Psychodidae	Pericoma	5.5	4	1	1
Diptera	Psychodidae	Psychoda	5.5		1	1
Diptera	Simuliidae		x	6	50	25
Diptera	Simuliidae	Prosimulium	4	4	5	3
Diptera	Simuliidae	Simulium	5	5	331	90
Diptera	Tabanidae		4	6	4	3
Diptera	Tabanidae	Chrysops	4	6	5	2
Diptera	Tabanidae	Tabanus	4	5	3	3
Diptera	Tipulidae		x	3	4	2
Diptera	Tipulidae	Antocha	5	3	458	144
Diptera	Tipulidae	Cryptolabis	2	3	1	1
Diptera	Tipulidae	Dicranota	3	3	70	33
Diptera	Tipulidae	Hexatoma	2	2	137	48
Diptera	Tipulidae	Limnophila	2	3	2	2
Diptera	Tipulidae	Limonia	x		4	1
Diptera	Tipulidae	Molophilus	4	3	2	1
Diptera	Tipulidae	Pedicia	x	6	1	1
Diptera	Tipulidae	Tipula	4	4	202	101
Ephemeroptera	Baetidae		4	4	7	5
Ephemeroptera	Baetidae	Acentrella	4	4	376	106
Ephemeroptera	Baetidae	Baetis	4	5	619	102
Ephemeroptera	Baetidae	Plauditus	3	4	16	5
Ephemeroptera	Baetiscidae	Baetisca	3	3	3	2
Ephemeroptera	Caenidae	Caenis	4	7	6	4
Ephemeroptera	Ephemerellidae		3	1	26	11
Ephemeroptera	Ephemerellidae	Dannella	2	2	1	1
Ephemeroptera	Ephemerellidae	Ephemerella	3	1	1600	145
Ephemeroptera	Ephemerellidae	Eurylophella	4	3	213	74
Ephemeroptera	Ephemerellidae	Serratella	3	2	476	86
Ephemeroptera	Ephemeridae	Ephemera	2	1	12	4
Ephemeroptera	Heptageniidae		x	4	63	19
Ephemeroptera	Heptageniidae	Cinygmula	x	4	2	2
Ephemeroptera	Heptageniidae	Epeorus	2	0	272	51
Ephemeroptera	Heptageniidae	Heptagenia	x		5	3
Ephemeroptera	Heptageniidae	Leucrocuta	2	1	74	33
Ephemeroptera	Heptageniidae	Nixe	x	2	1	1
Ephemeroptera	Heptageniidae	Rhithrogena	2	0	10	5
Ephemeroptera	Heptageniidae	Stenacron	4	7	51	24

CT Data

Class or Order	Family	Genus	CT Data			
			BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Ephemeroptera	Heptageniidae	Stenonema	4	3	3294	235
Ephemeroptera	Isonychiidae	Isonychia	3	2	3108	173
Ephemeroptera	Leptophlebiidae		2	2	331	57
Ephemeroptera	Leptophlebiidae	Habrophlebiodes	4	6	1	1
Ephemeroptera	Leptophlebiidae	Leptophlebia	2	4	8	4
Ephemeroptera	Leptophlebiidae	Paraleptophlebia	2	1	191	23
Ephemeroptera	Potamanthidae	Anthopotamus	2	4	9	7
Hemiptera	Sialidae	Sialis	3	4	38	27
Hemiptera	Veliidae	Microvelia	x		2	1
Hemiptera	Veliidae	Rhagovelia	x		2	2
Lepidoptera			x	5	2	2
Lepidoptera	Pyralidae		x	5	1	1
Lepidoptera	Pyralidae	Paraponyx	x	5	1	1
Lepidoptera	Pyralidae	Petrophila	x	5	16	10
Megaloptera	Corydalidae	Corydalus	3	6	197	87
Megaloptera	Corydalidae	Nigronia	3	4	792	186
Odonata	Aeshnidae	Boyeria	4	2	45	31
Odonata	Calopterygidae	Calopteryx	4	5	26	18
Odonata	Coenagrionidae		4	9	2	2
Odonata	Coenagrionidae	Argia	4	7	51	27
Odonata	Coenagrionidae	Enallagma	4	9	2	2
Odonata	Cordulegastridae	Cordulegaster	3	3	6	5
Odonata	Gomphidae		3	1	118	40
Odonata	Gomphidae	Dromogomphus	3	5	2	2
Odonata	Gomphidae	Hagenius	2	1	1	1
Odonata	Gomphidae	Lanthus	3	5	14	10
Odonata	Gomphidae	Ophiogomphus	2	1	107	41
Odonata	Gomphidae	Stylogomphus	3	0	50	31
Plecoptera			x		9	3
Plecoptera	Capniidae		4	1	99	51
Plecoptera	Capniidae	Allocapnia	4	3	184	13
Plecoptera	Capniidae	Paracapnia	3	1	6	5
Plecoptera	Chloroperlidae		3	1	3	3
Plecoptera	Chloroperlidae	Haploperla	3	1	12	1
Plecoptera	Chloroperlidae	Rasvena	3	0	2	2
Plecoptera	Chloroperlidae	Suwallia	3	0	1	1
Plecoptera	Chloroperlidae	Sweltsa	3	0	7	5
Plecoptera	Chloroperlidae	Utaperla	3		1	1
Plecoptera	Leuctridae		2	0	5	5
Plecoptera	Leuctridae	Leuctra	2	0	28	11
Plecoptera	Leuctridae	Paraleuctra	2		1	1
Plecoptera	Nemouridae		3	2	4	4
Plecoptera	Nemouridae	Prostoia	3	2	3	2
Plecoptera	Nemouridae	Shipsa	3	2	1	1
Plecoptera	Nemouridae	Soyedina	3	0	4	4
Plecoptera	Peltoperlidae	Tallaperla	2	0	132	42
Plecoptera	Perlidae		x	1	51	15
Plecoptera	Perlidae	Acroneuria	3	0	843	154
Plecoptera	Perlidae	Agnetina	3	2	36	17
Plecoptera	Perlidae	Eccoptura	3	0	16	7

CT Data

Class or Order	Family	Genus	CT Data			
			BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Plecoptera	Perlidae	Neoperla	2	1	7	4
Plecoptera	Perlidae	Paragnetina	3	1	275	92
Plecoptera	Perlodidae		2	2	11	8
Plecoptera	Perlodidae	Helopicus	x	2	5	2
Plecoptera	Perlodidae	Isogenoides	2	0	12	8
Plecoptera	Perlodidae	Isoperla	3	2	7	7
Plecoptera	Pteronarcyidae	Pteronarcys	2	0	13	7
Plecoptera	Taeniopterygidae		3	2	14	9
Plecoptera	Taeniopterygidae	Oemopteryx	3	1	17	6
Plecoptera	Taeniopterygidae	Taenionema	3	2	13	7
Plecoptera	Taeniopterygidae	Taeniopteryx	3	2	2859	197
Trichoptera			x		6	3
Trichoptera	Apataniidae	Apatania	3	3	1037	146
Trichoptera	Brachycentridae	Adicrophleps	2	2	6	3
Trichoptera	Brachycentridae	Brachycentrus	3	1	95	24
Trichoptera	Brachycentridae	Micrasema	3	2	391	63
Trichoptera	Glossosomatidae		4	0	10	8
Trichoptera	Glossosomatidae	Agapetus	3	0	10	5
Trichoptera	Glossosomatidae	Culoptila	x		2	2
Trichoptera	Glossosomatidae	Glossosoma	4	0	1015	167
Trichoptera	Glossosomatidae	Protoptila	3	1	13	7
Trichoptera	Goeridae	Goera	2	0	19	16
Trichoptera	Helicopsychidae	Helicopsyche	2	3	155	25
Trichoptera	Hydropsychidae		x	4	82	27
Trichoptera	Hydropsychidae	Ceratopsyche	5	3	6596	247
Trichoptera	Hydropsychidae	Cheumatopsyche	5	5	8446	278
Trichoptera	Hydropsychidae	Diplectrona	3	0	292	35
Trichoptera	Hydropsychidae	Hydropsyche	5	6	7433	268
Trichoptera	Hydropsychidae	Macrostemum	4	3	272	36
Trichoptera	Hydroptilidae		4	4	5	2
Trichoptera	Hydroptilidae	Hydroptila	4	6	85	45
Trichoptera	Hydroptilidae	Leucotrichia	5.5	4	197	34
Trichoptera	Hydroptilidae	Ochrotrichia	4	4	1	1
Trichoptera	Hydroptilidae	Oxyethira	4	3	1	1
Trichoptera	Hydroptilidae	Palaeagapetus	x	4	9	1
Trichoptera	Lepidostomatidae	Lepidostoma	2	1	503	90
Trichoptera	Leptoceridae	Ceraclea	2	3	4	4
Trichoptera	Leptoceridae	Mystacides	4	4	42	23
Trichoptera	Leptoceridae	Nectopsyche	x	3	1	1
Trichoptera	Leptoceridae	Oecetis	4	8	84	45
Trichoptera	Leptoceridae	Setodes	2	2	6	5
Trichoptera	Leptoceridae	Triaenodes	4	6	2	2
Trichoptera	Limnephilidae		x	4	49	17
Trichoptera	Limnephilidae	Hydatophylax	3	2	51	23
Trichoptera	Limnephilidae	Pycnopsyche	3	4	91	29
Trichoptera	Molannidae	Molanna	4	6	1	1
Trichoptera	Odontoceridae	Psilotreta	2	0	64	31
Trichoptera	Philopotamidae		x	3	11	6
Trichoptera	Philopotamidae	Chimarra	4	4	3272	202
Trichoptera	Philopotamidae	Dolophilodes	3	0	1060	111

CT Data

Class or Order	Family	Genus	BCG Attribute	HBI Tolerance	No. of Individuals	No. of Samples
Trichoptera	Phryganeidae	Hagenella	x	0	1	1
Trichoptera	Phryganeidae	Ptilostomis	4	5	1	1
Trichoptera	Polycentropodidae		3	6	8	8
Trichoptera	Polycentropodidae	Cernotina	3	6	6	5
Trichoptera	Polycentropodidae	Neureclipsis	4	7	37	19
Trichoptera	Polycentropodidae	Nyctiophylax	3	5	4	4
Trichoptera	Polycentropodidae	Polycentropus	3	6	33	25
Trichoptera	Psychomyiidae		4	2	1	1
Trichoptera	Psychomyiidae	Lype	4	2	15	14
Trichoptera	Psychomyiidae	Psychomyia	4	2	43	21
Trichoptera	Rhyacophilidae	Rhyacophila	3	0	631	147
Trichoptera	Uenoidae	Neophylax	3	3	86	38
Amphipoda	Crangonyctidae	Crangonyx	5	8	2	2
Amphipoda	Gammaridae	Gammarus	5.5	6	794	70
Amphipoda	Talitridae	Hyalella	4	8	23	10
Decapoda	Astacidae	Astacidae	x	6	5	4
Decapoda	Cambaridae		x	6	2	2
Decapoda	Cambaridae	Cambarus	4		3	3
Decapoda	Cambaridae	Orconectes	x	6	19	5
Isopoda	Asellidae		x	8	4	1
Isopoda	Asellidae	Caecidotea	4	8	8	5
Isopoda	Asellidae	Conasellus	5	8	228	45
Bivalvia	Corbiculidae	Corbicula	6	8	1	1
Bivalvia	Pisidiidae		4	8	621	96
Bivalvia	Pisidiidae	Pisidium	4	8	19	5
Bivalvia	Pisidiidae	Sphaerium	4	8	32	5
Gastropoda			x	6	3	3
Gastropoda	Ancylidae		4	6	29	12
Gastropoda	Ancylidae	Ferrissia	4	6	113	38
Gastropoda	Ancylidae	Laevapex	4	5	408	80
Gastropoda	Lymnaeidae		4	6	5	2
Gastropoda	Lymnaeidae	Fossaria	4	6	35	15
Gastropoda	Lymnaeidae	Pseudosuccinea	4	6	1	1
Gastropoda	Lymnaeidae	Stagnicola	4	6	7	3
Gastropoda	Physidae	Physa	4	8	195	53
Gastropoda	Planorbidae		4	6	2	2
Gastropoda	Planorbidae	Gyraulus	4	8	5	4
Gastropoda	Planorbidae	Helisoma	5	6	3	3
Gastropoda	Planorbidae	Micromenetus	4	5	53	25
Gastropoda	Planorbidae	Planorbula	4		1	1
Gastropoda	Valvatidae	Valvata	5	8	1	1
Gastropoda	Hydrobiidae		4	8	10	4
Gastropoda	Hydrobiidae	Amnicola	5	8	134	26
Nematoda			x		38	25
Enopla	Tetrastemmatidae	Prostoma	4	8	223	86
Turbellaria			4	4	47	26

APPENDIX B

BIOLOGICAL DATA AND BCG LEVEL ASSIGNMENTS

Stream name	date	EP	GH	Panel			median	mean	Nominal
				MB	JG	BJ			
Hockanum Brook	10/31/1996	4.2	3.5	3.5			3.5	3.7333	3
East Branch Byram River	10/03/2002	3.8	4.2	3.8		3.2	3.8	3.75	3
East Branch Byram River	10/03/2002	3.8	4.2	3.5			3.8	3.8333	3
Hockanum River	10/26/1998	5.5	5.5	5.8	5.5	5.2	5.5	5.5	5
Mad River	11/14/1996	5.8	6.5	6.5	6.5	6.2	6.5	6.3	6
Mattabesset River	10/16/1996	5.5	5.8	5.8	5.5	5.5	5.5	5.62	5
Mill River	11/13/1997	4.2	4.2	4.2	4.8	3.8	4.2	4.24	4
Mill River	10/14/1997	3.8	3.5	3.5			3.5	3.6	3
Branford River	10/17/1997	4.2	3.8	4.5	4.5	4.8	4.5	4.36	4
Naugatuck River	11/05/1996	5.5	5.2	5.2	5.5		5.35	5.35	5
Noroton River	10/10/2000	5.5	5.8	5.8	6.2	5.8	5.8	5.82	5
Norwalk River	10/30/1997	5.5	5.2	5.5	5.5	5.5	5.5	5.44	5
Norwalk River	10/28/1997	5.2	5.2	5.2	5.5	5.2	5.2	5.26	5
Byram River	10/20/1997	5.8	5.8	5.8	5.5	5.5	5.8	5.68	5
Pattaconk Brook	10/29/1998	3.5	3.5	3.5	3.2	3.2	3.5	3.38	3
Pequabuck River	10/01/1998	5.2	5.2	5.2	4.8	5.2	5.2	5.12	5
Pequabuck River	10/01/1998	5.5	5.8	5.8	5.8	5.8	5.8	5.74	5
Pequabuck River	10/02/1998	5.8	5.8	5.8			5.8	5.8	5
Quinnipiac River	10/14/1997	5.5	6.2	5.2	5.8	5.5	5.5	5.64	5
Rippowam River	10/31/1997	5.5	5.5	5.5	5.5	5.5	5.5	5.5	5
Salmon Brook	10/19/1998	4.5	3.5	3.5	3.8	4.2	3.8	3.9	3
Salmon River	10/27/1998	2.2	2.5	2.5	2.5	2.8	2.5	2.5	2
Sandy Brook	10/21/1998	2.5	2.5	2.8	2.5	2.8	2.5	2.62	2
Scantic River	10/22/1998	4.8	5.2	4.5	4.8		4.8	4.825	4
Shetucket River	10/28/1999	4.8	5.2	4.8	4.8	4.8	4.8	4.88	4
Still River	10/24/2000	6.2	6.2	6.2	6.5		6.2	6.275	6
West Branch Naugatuck Rive	11/05/1996	4.2	3.5	4.2	4.5	4.2	4.2	4.12	4
Willimantic River	10/14/1999	3.5	3.8	4.2	3.5	3.5	3.5	3.7	3
Willimantic River	10/03/2000	5.5	5.8	5.8	6.2	5.8	5.8	5.82	5
Mount Hope River	10/26/1999	3.5	3.2	3.5			3.5	3.4	3
Moosup River	10/28/1999	3.5	3.2	3.5			3.5	3.4	3
Blackwell Brook	10/22/1999	3.8	3.8	3.8	3.5	3.5	3.8	3.68	3
Broad Brook	10/06/1999	3.8	4.5	4.2	3.8	4.2	4.2	4.1	4
Merrick Brook	10/07/1999	2.8	2.8	2.8	2.5	2.8	2.8	2.74	2
Quinebaug River	10/27/1999	4.5	4.5	4.2	4.5	4.8	4.5	4.5	4
Shunock River	10/07/2003	3.8	4.2	3.5	3.5	4.2	3.8	3.84	3
Seth Williams Brook	10/19/2000	5.5	5.5	5.5	5.5	5.5	5.5	5.5	5
Quaker Brook	10/23/2000	3.5	3.2	3.5	3.2	3.2	3.2	3.32	3
Sages Ravine Brook	10/21/2002	2.5	2.5	2.5			2.5	2.5	2
Furnace Brook	10/13/1999	4.2	4.5	5.2	5.2	4.8	4.8	4.78	4
East Branch Salmon Brook	11/01/2004	2.5	2.2	1.8	2.2	2.5	2.2	2.24	2
Beach Brook	11/01/2004	2.5	2.5	2.8		2.5	2.5	2.575	2
Housatonic River	10/27/2004	3.5	3.8	3.2			3.5	3.5	3
Crooked Brook	10/27/2004	3.8	3.5	3.8	3.5	3.8	3.8	3.68	3
Clark Creek	10/28/2004	3.5	3.2	3.5	3.2	3.2	3.2	3.32	3
Hammonasset River	10/21/1997	3.2	2.8	2.8	2.8	3.2	2.8	2.96	2
Sympaug Brook	10/17/2000	5.5	5.8	5.5	5.8	6.2	5.8	5.6857	5
Mountain Brook	10/28/2002	2.8	2.8	2.8			2.8	2.88	2

APPENDIX C

DESCRIPTIONS OF BCG LEVELS IN CONNECTICUT STREAMS

Biological Condition Gradient: description of gradient and rules for cold-water streams of Connecticut

Resource Condition Tiers	Biological Condition Characteristics (Effects)
<hr style="border: 1px solid black;"/> <b style="font-size: 2em;">1 <hr style="border: 1px solid black;"/>	<p>I <i>Historically documented, sensitive, long-lived, or regionally endemic taxa</i></p> <p>→ Long-lived native species of fish-host specialist or long-term brooder mussels such as Brook floater- <i>Alasmodonta varicosa</i>; Triangle floater- <i>Alasmodonta undulata</i>; Yellow lampmussel- <i>Lampsilis cariosa</i> are present in naturally occurring densities</p> <p>II <i>Highly Sensitive taxa</i></p> <p>→ The proportion of total richness represented by rare, specialist and vulnerable taxa is high: Plecoptera: Peltoperlidae, <i>Amphinemura</i>, <i>Isogenoides</i>, <i>Neoperla</i>, <i>Pteronarcys</i>, <i>Leuctra</i>; Ephemeroptera: <i>Centroptilum</i>, <i>Heterocloeon</i>, <i>Brachycercus</i>, <i>Drunella</i>, <i>Rhithrogena</i>, <i>Epeorus</i>, <i>Leucrocuta</i>; Trichoptera: <i>Proptotila</i>; <i>Psilotreta</i>, <i>Lepidostoma</i>, <i>Ceraclea</i>; Diptera: Blephariceridae, <i>Stempellina</i>, <i>Limnophila</i></p> <p>III <i>Intermediate Sensitive taxa</i></p> <p>→ Densities of Intermediate Sensitive taxa are as naturally occur: Plecoptera: <i>Acroneuria</i>; Ephemeroptera: <i>Ephemerella</i>, <i>Baetisca</i>, <i>Proclaeon</i>; Coleoptera: <i>Psephenus</i> Diptera: <i>Rheocricotopus</i>, <i>Stempelinella</i></p> <p>IV <i>Taxa of Intermediate tolerance</i></p> <p>→ Densities of intermediate tolerance taxa are as naturally occur: Trichoptera: <i>Diplectrona</i>, <i>Hydroptila</i>, <i>Chimarra</i>, <i>Neureclipsis</i>; Diptera: <i>Tvetenia</i>, <i>Polypedilum</i>, <i>Microtendipes</i>, <i>Simulium</i>; Coleoptera: <i>Stenelmis</i>;</p> <p>V <i>Tolerant taxa</i></p> <p>→ Occurrence and densities of Tolerant taxa are as naturally occur: Diptera: <i>Cricotopus</i>, <i>Chironomus</i>, <i>Rheotanytarsus</i>, <i>Dicrotendipes</i>; Non-Insects: <i>Caecidotea</i>, Isopoda, Erpobdellidae, Tubificidae, Glossiphoniidae</p> <p>VI <i>Non native or intentionally introduced taxa</i></p> <p>→ Non native taxa such as Brown trout, Rainbow trout, Yellow perch, are absent or, if they occur, their presence does not displace native biota or alter native structure and function</p> <p>VII <i>Physiological condition of long-lived organisms</i></p> <p>→ Anomalies are absent or rare; any that occur are consistent with naturally occurring incidence and characteristics</p> <p>VIII <i>Ecosystem Function</i></p> <p>→ Rates and characteristics of <i>life history (e.g., reproduction, immigration, mortality, etc.)</i>, and materials exchange processes (<i>e.g., production, respiration, nutrient exchange, decomposition, etc.</i>) are comparable to that of "natural" systems</p> <p>→ The system is predominantly heterotrophic, sustained by leaf litter inputs from intact riparian areas, with low algal biomass; P/R<1 (Photosynthesis: Respiration ratio)</p> <p>IX <i>Spatial and temporal extent of detrimental effects</i></p> <p>→ Not applicable- disturbance is limited to natural events such as storms, droughts, fire, earth-flows. A natural flow regime is maintained.</p> <p>X <i>Ecosystem connectance</i></p> <p>→ Reach is highly connected with groundwater, its floodplain, and riparian zone, and other reaches in the basin, at least annually. Allows for access to habitats and maintenance of seasonal cycles that are necessary for life history requirements, colonization sources, migration and <i>refugia</i> for extreme events.</p>

2

Minimal changes in structure of the biotic community and minimal changes in ecosystem function

Virtually all native taxa are maintained with some changes in biomass and/or abundance; ecosystem functions are fully maintained within the range of natural variability

Whole assemblage and sample

- Overall taxa richness and density is as naturally occurs
- **RULE 1, 2:** Taxa richness is high and subsample density is near target
- **Quantitative Rule 1, 2:** Total taxa > (25-30) genera and Total individuals > (50-60% of target)

I Historically documented, sensitive, long-lived, regionally endemic taxa

- Rule not defined.

II Highly Sensitive taxa

- Richness of rare and/or specialist invertebrate taxa is low to moderate though densities may be low .
- **RULE 3:** At least some taxa are present
- **Quantitative Rule 3:** Taxa (II) > (3-5)

III Intermediate Sensitive taxa

- Richness and abundance of intermediate sensitive taxa is high. Some may have increased due to slightly elevated production
- **RULE 4:** All sensitive taxa (highly sensitive + intermediate sensitive): comprise half or more of all taxa
- **RULE 5 :** All sensitive individuals: comprise nearly half or more of all organisms
- **Quantitative Rule 4:** Taxa (II + III) > (45 – 55%) of all taxa
- **Quantitative Rule 5:** Individuals (II + III) > (30-40%)

IV Taxa of Intermediate tolerance

- May be slight increases in densities of macroinvertebrate taxa
- **RULE:** None

V Tolerant taxa (also includes taxa considered highly tolerant)

- Occurrence and densities of Tolerant taxa are as naturally occur. Typically present but a very small fraction of organisms.
- **RULE 6:** Tolerant individuals (tolerant + highly tolerant) comprise a small fraction or less of all organisms
- **Quantitative Rule 6:** Individuals (V) < (10-15%)

VI-IX Non-native taxa; Physiological condition; Ecosystem Function; Spatial and temporal extent

- Not addressed for macroinvertebrates

X Ecosystem connectance

- Connectance on a local scale (floodplain, tributaries) remains good but dams and other flow obstructions downstream impede migration of fish and mussels (eels, salmonids, migration-dependent unionids)

COMBINATORIAL RULE

- To be considered Tier 2, rules 1 - 4 must apply; combined with AND, and Rule OR Rule 6.
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3	<p>Whole assemblage and sample</p> <ul style="list-style-type: none"> → Overall taxa richness and density is as naturally occurs → RULE 1, 2: Taxa richness is moderately high and subsample density is near target → Quantitative Rules 1, 2: Total taxa > (18-22) and Total individuals > (50-60% of target)
<p>Evident changes in structure of the biotic community and minimal changes in ecosystem function</p> <p><i>Some changes in structure due to loss of some rare native taxa; shifts in relative abundance of taxa but sensitive-ubiquitous taxa are common and abundant; ecosystem functions are fully maintained through redundant attributes of the system</i></p>	<ul style="list-style-type: none"> I <i>Historically documented, sensitive, long-lived, or regionally endemic taxa</i> <ul style="list-style-type: none"> → Rule not defined II <i>Highly Sensitive taxa</i> <ul style="list-style-type: none"> → Some replacement of taxa having narrow or specialized environmental requirements, with functionally equivalent <i>intermediate-sensitive</i> taxa; coldwater obligate taxa are disadvantaged. Reduced richness; may be absent. → RULE: May be absent (no rule) III <i>Intermediate Sensitive taxa</i> <ul style="list-style-type: none"> → Intermediate sensitive or generalist taxa are common and abundant; taxa with broader temperature-tolerance range are favored → RULE 3: All sensitive taxa (highly sensitive + intermediate sensitive) are moderately diverse → Quantitative Rule 3: Taxa (II + III) > 8 - 10 → RULE 4: All sensitive individuals: comprise a substantial fraction of all organisms → Quantitative Rule 4: Individuals (II + III) > (30-50%) IV <i>Taxa of Intermediate (indifferent) tolerance</i> <ul style="list-style-type: none"> → Filter-feeders may show increased densities in response to nutrient enrichment, but relative abundance of all expected major groups is well-distributed → Increased temperature and increased available nutrients may result in increased algal productivity causing an increase in the thickness of the diatom mat. → RULE: None V <i>Tolerant taxa (also includes taxa considered highly tolerant)</i> <ul style="list-style-type: none"> → Richness of Chironomidae is increased; but overall relative abundance is well-distributed among taxa from Groups III, IV and V, with the majority of taxa represented from Groups III and IV. → RULE 5: Tolerant individuals (tolerant + highly tolerant) comprise a moderately small fraction or less of all organisms → Quantitative Rule 5: Individuals (V + Va) < (40 - 50%) VI-X <i>Non-native taxa; Physiological condition; Ecosystem Function; Spatial and temporal extent; Connectance</i> <ul style="list-style-type: none"> → Not addressed for macroinvertebrates. <p>COMBINATORIAL RULE Must fail Tier 2 and must meet Rules 1 – 3, and (4 OR 5)</p>

4	<p>Whole assemblage and sample</p> <ul style="list-style-type: none"> → Overall taxa richness is slightly reduced, and density may be high → RULE 1, 2: Taxa richness is moderately high and subsample density is near target → Quantitative Rule 1, 2: Total taxa > (18-22) and Total individuals > (50-60% of target) <p>I Historically documented, sensitive, long-lived, regionally endemic taxa</p> <ul style="list-style-type: none"> → Rule not defined. <p>II Highly Sensitive taxa</p> <ul style="list-style-type: none"> → Richness of specialist and vulnerable taxa is notably reduced; if present, densities are low → RULE: May be absent (no rule) <p>III Intermediate Sensitive taxa</p> <ul style="list-style-type: none"> → Densities of intermediate-sensitive taxa are sufficient to indicate that reproducing populations are present but relative abundance is reduced due to increased densities of opportunist invertebrate taxa (Group IV) → Overall mayfly taxonomic richness is reduced relative to the Tier 2 condition.; Predatory stoneflies are reduced → RULE 3: Sensitive taxa (highly sensitive + intermediate sensitive) are moderately diverse; may be less than Tier 3 → Quantitative Rule 3: Taxa (II + III) > (3-5) → RULE 4: All sensitive individuals comprise at least a moderate and functional fraction of all organisms → Quantitative Rule 4: Individuals (II + III) > (10-20%) <p>IV Taxa of Intermediate (indifferent) tolerance</p> <ul style="list-style-type: none"> → Increased loads of suspended particles favor collector-filterer invertebrates resulting in increased densities and relative abundance of filter-feeding caddisflies and chironomids → RULE: None <p>V Tolerant taxa (also includes taxa considered highly tolerant)</p> <ul style="list-style-type: none"> → There is an increase in the relative abundance of tolerant generalists but they do not exhibit significant dominance → Overall relative abundance is well distributed among taxa from Groups III, IV and V, with the majority of the total abundance represented from Group IV. → RULE 5: Tolerant individuals comprise less than half of all organisms → Quantitative Rule 5: Individuals (V) < (65 - 75%) <p>VI-X Non-native taxa; Physiological condition; Ecosystem Function; Spatial and temporal extent; Connectance</p> <ul style="list-style-type: none"> → Not addressed for macroinvertebrates. <p>COMBINATORIAL RULE</p> <p>Must fail Tier 2 and must meet Rules 1, 2, and 5, and either of Rules 3 or 4. To distinguish from Tier 3, an average of Rules 2, 3, 4, and 5 is used.</p>
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Major changes in structure of the biotic community and moderate changes in ecosystem function

Sensitive taxa are markedly diminished; conspicuously unbalanced distribution of major groups from that expected; organism condition shows signs of physiological stress; system function shows reduced complexity and redundancy; increased build-up or export of unused materials

Whole assemblage and sample

- Overall taxa richness is reduced, but density may be high
- **RULE 1, 2:** Taxa richness is moderate and subsample density is near target
- **Quantitative Rule 1, 2:** Total taxa > (8-12) and Total individuals > (50-60% of target)

I Historically documented, sensitive, long-lived, or regionally endemic taxa

- Rule not defined

II Highly Sensitive taxa

- Only the rare occurrence of individual representatives of specialist and vulnerable taxa with no evidence of successful reproduction
- **RULE:** May be absent (no rule)

III Intermediate Sensitive taxa

- Either absent or present in very low numbers, indicating impaired recruitment and/or reproduction
- **RULE:** May be absent
- **Quantitative Rule:** Failure of Tier 4 rules (complement)

IV Taxa of Intermediate (indifferent) tolerance

- Filter-feeding invertebrates such as Hydropsychid caddisflies (e.g., *Cheumatopsyche*) and filter-feeding midges (e.g., *Rheotanytarsus*, *Microtendipes*) may occur in very high numbers
- **RULE:** None

V Tolerant taxa (also includes taxa considered highly tolerant)

- Frequent occurrence of tolerant collector-gatherers
- Relative abundance of non-insects often equal to or higher than relative abundance of insects
- Deposit-feeders such as Oligochaeta are increased
- Numbers of tolerant predators are increased
- **RULE:** May be very abundant
- **Quantitative Rule:** Failure of Tier 4 rule (complement)

VI-X Non-native taxa; Physiological condition; Ecosystem Function; Spatial and temporal extent; Connectance

- Not addressed for macroinvertebrates.

COMBINATORIAL RULE

Failure of Tier 4 rules and must meet both Rules 1 and 2

6	<p>Whole assemblage and sample</p> <p>→ Overall taxa richness is greatly reduced, but density may be high, or greatly reduced (indicating toxicity)</p> <p>→ RULE: Taxa richness may be extremely low or subsample density may be below target</p> <p>→ Quantitative Rule: Total taxa < (8-12) or Total individuals < (45-55% of target) (fails Tier 5)</p>
<p>Severe changes in structure of the biotic community and major loss of ecosystem function</p> <p><i>Extreme changes in structure; wholesale changes in taxonomic composition; extreme alterations from normal densities and distributions; organism condition is often poor; ecosystem functions are severely altered</i></p>	<p>I <i>Historically documented, sensitive, long-lived, regionally endemic taxa</i></p> <p>→ Poor water quality, compaction of substrate, elevated temperature regime and absence of fish hosts for reproductive functions preclude the survival of any mussel fauna</p> <p>II <i>Highly Sensitive taxa</i></p> <p>→ These taxa are absent due to poor water quality, elevated temperature regime, alteration of habitat, loss of riparian zone, etc.</p> <p>III <i>Intermediate Sensitive taxa</i></p> <p>→ Absent due to above listed factors, though an occasional transient individual, usually in poor condition, may be collected.</p> <p>IV <i>Taxa of Intermediate (indifferent) tolerance</i></p> <p>→ Filter-feeding insects and other macroinvertebrate representatives of this group are severely reduced in density and richness, or are absent.</p> <p>V <i>Tolerant taxa (also includes taxa considered highly tolerant)</i></p> <p>→ Low dissolved oxygen conditions preclude survival of most insect taxa except those with special adaptations to deficient oxygen conditions</p> <p>→ The macroinvertebrate assemblage is dominated by tolerant non-insects</p> <p>VI-X <i>Non-native taxa; Physiological condition; Ecosystem Function; Spatial and temporal extent; Connectance</i></p> <p>→ Not addressed for macroinvertebrates.</p> <p>→</p> <p>COMBINATORIAL RULE</p> <p>Rule for Tier 6 is any failure of Tier 5 rule</p>

APPENDIX D

METRIC STATISTICS

Appendix D

Metric Statistics

Trend: Direction of metric response with increasing stress. The trends for unresponsive metrics were left blank.

Incr = increasing metric values with increasing stress.

Decr = decreasing metric values with increasing stress.

DE: Discrimination Efficiency = the percentage of degraded samples lower or higher than the quartile of the reference samples, in the direction of the trend. This appendix considers calibration data only.

CV_{ref}: Coefficient of Variability = the standard deviation of reference metric values over the mean of the values, expressed as a percentage.

Metric Name	Metric Code	Trend	DE	CV_{ref}
<u>Richness</u>				
Total Taxa	TotalTax	Decr	100	22.2
Insect Taxa	InsecTax	Decr	100	23.6
Non-Insect Taxa Percent	NonInsPT	Incr	83.3	83.1
EPT Taxa	EPTTax	Decr	100	22.9
Ephemeroptera Taxa	EphemTax	Decr	100	33.0
Ephemeroptera Taxa (adjusted for watershed area)	adjEphemTax	Decr	100	18.7
Plecoptera Taxa	PlecoTax	Decr	100	45.5
Trichoptera Taxa	TrichTax	Decr	83.3	33.8
Diptera Taxa	DipTax	Decr	41.7	55.0
Midge Taxa	ChiroTax	Decr	41.7	68.5
Orthoclaadiinae Taxa	OrthoTax			73.2
Tanytarsini Taxa	TanytTax			94.9
Coleoptera Taxa	ColeoTax	Decr	100	41.2
Crustacea & Mollusca Taxa	CrMolTax		8.3	101.0
Oligochaeta Taxa	OligoTax			135.6
<u>Composition</u>				
% EPT	EPTPct	Incr	50.0	28.2
% EPT excluding Hydropsychidae and Baetidae	EPTnHBpct	Decr	100	37.3
% EPT excluding Hydropsychidae and Baetidae (adjusted for watershed area)	adj%SEPT	Decr	100	31.3
% Ephemeroptera	EphemPct	Decr	66.7	71.4
% Plecoptera	PlecoPct	Decr	100	84.3
% Trichoptera	TrichPct	Incr	100	53.3
% Baetidae:Ephemeroptera	Baet2EphPct		16.7	217.9
% Hydropsychidae:EPT	Hyd2EPTPct	Incr	100	74.6
% Hydropsychidae:Trichoptera	Hyd2TriPct	Incr	100	52.8
% Diptera	DipPct			116.2
% Midge	ChiroPct	Decr	41.7	135.5
% Non-Insect	NonInPct	Incr	50.0	160.2
% Amphipoda	AmphPct		33.3	
% Coleoptera	ColeoPct	Decr	91.7	68.5
Cricotopus&Chironomus/Chironomidae	CrCh2ChiPct		33.3	361.8
% Bivalvia	BivalPct		8.3	212.1
% Crustacea & Mollusca	CrMolPct	Incr	41.7	237.9
% Gastropoda	GastrPct	Incr	41.7	270.7
% Isopoda	IsoPct		25.0	
% Odonata	OdonPct		8.3	126.9
% Oligochaeta	OligoPct		16.7	277.2
% Orthoclaadiinae:Midges	Orth2ChiPct	Incr	41.7	70.3
% Tanytarsini	TanytPct			179.6
% Tanytarsini:Midges	Tnyt2ChiPct		33.3	116.4
<u>Evenness</u>				
Shannon-Weiner Index (base e)	Shan_e	Decr	100	13.7
Evenness	Evenness	Decr	91.7	14.5
Margoleff's Diversity	D_Mg	Decr	100	23.8
Simpson's Index	D	Incr	100	49.3

Metric Name	Metric Code	Trend	DE	CV_{ref}
% dominant 1	Dom01Pct	Incr	91.7	41.0
<u>Feeding Group</u>				
% Collector	ClctPct	Decr	83.3	50.5
% Filterer	FiltrPct	Incr	100	74.6
% Predator	PredPct	Decr	100	50.7
% Scraper	ScrapPct	Decr	83.3	47.3
% Shredder	ShredPct	Decr	100	75.5
Collector Taxa	ClctTax	Decr	100	28.2
Filterer Taxa	FiltrTax	Decr	66.7	33.5
Predator Taxa	PredTax	Decr	100	37.9
Scraper Taxa	ScrapTax	Decr	100	30.1
Shredder Taxa	ShredTax	Decr	100	57.5
<u>Tolerance</u>				
Beck's Index	BeckBI	Decr	100	24.7
Hilsenhoff's Index	HBI	Incr	100	26.4
Biotic Index, Individual BCG Attributes	BCG_BI	Incr	100	11.4
Biotic Index, Taxa BCG Attributes	BCG_TBI	Incr	100	6.3
% Intolerant	IntolPct	Decr	100	38.5
% Tolerant	TolerPct		16.7	118.9
Intolerant Taxa	IntolTax	Decr	100	28.9
Tolerant Taxa	TolerTax		8.3	80.8
<u>BCG</u>				
BCG attribute 2 taxa	TotalAtt2Gen	Decr	100	59.1
BCG attribute 3 taxa	TotalAtt3Gen	Decr	100	32.4
BCG attribute 4 taxa	TotalAtt4Gen	Decr	91.7	25.1
BCG attribute 5 taxa	TotalAtt5Gen	Incr	66.7	45.5
BCG attribute 5a taxa	TotalAtt5aGen	Incr	75.0	264.0
BCG attribute 2 % taxa	At2PctGen	Decr	100	51.7
BCG attribute 3 % taxa	At3PctGen	Decr	100	18.9
BCG attribute 4 % taxa	At4PctGen	Incr	66.7	21.0
BCG attribute 5 % taxa	At5PctGen	Incr	100	55.7
BCG attribute 5a % taxa	At5aPctGen	Incr	75.0	280.2
% BCG attribute 2	Att2Pct	Decr	100	78.8
% BCG attribute 3	Att3Pct	Decr	100	39.0
% BCG attribute 4	Att4Pct	Decr	66.7	50.4
% BCG attribute 5	Att5Pct	Incr	100	104.1
% BCG attribute 5a	Att5aPct	Incr	75.0	364.8
<u>Community Models</u>				
Percent Model Affinity	PMA	Decr	100	48.7
Observed/Expected (p>half)	OE_p>half	Decr	100	17.5
Observed/Expected (p>0)	OE_p>0	Decr	100	19.5