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# TB208: Biological Water Quality Standards to Achieve Biological Condition Goals in Maine Rivers and Streams: Science and Policy

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Leonidas Tsomides, and Thomas J. Danielson**

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Cover: Machias River, near Beddington, Maine. Photograph by Susan Davies

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## Preface

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This publication describes the philosophy, history, methodology, and management applications of numeric biological criteria in water quality standards in Maine. The presentation describes the decision-making process used by the Maine Department of Environmental Protection (MDEP) for assessing attainment of aquatic life uses in water quality standards using benthic macroinvertebrates in Maine streams and rivers including eight case studies of management applications and the improved environmental outcomes that have resulted. The MDEP, University of Maine, and business and nonprofit stakeholders participated in the development and testing of Maine's numeric biological criteria. This publication further discusses the broader relevance of numeric biological criteria in water quality management at both the state and federal levels and considers parallels and differences between Maine's biological criteria and other biological assessment methods in the United States and the European Union.

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# Part 1: Evolution of Biologically Based Water Management In Maine— Merger of Policy and Scientific Needs

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## 1.1 INTRODUCTION

### 1.1.1 Historical policy context

In 1990, the U.S. Environmental Protection Agency (USEPA) issued a guidance document entitled “Biological Criteria: National Program Guidance for Surface Waters” (USEPA 1990). This document urged the U.S. states to develop narrative biological criteria to assess the biological integrity of aquatic communities, a goal of the federal Clean Water Act (CWA) not addressed by the physical and chemical water quality assessment approaches that had been practiced for decades (Yoder and Rankin 1998; Karr and Yoder 2004; Schleiger 2000; Adler 2003). With the release of four additional biological criteria documents the USEPA has further acknowledged the importance of biological information to assess attainment of the designated use “aquatic life support” (ALU)<sup>1</sup>, as required of states by the CWA (USEPA 2005, 2011, 2013, 2016).

In the early 1980s, the state of Maine had already recognized the inadequacy of assessing stream and river quality based solely on chemical and physical criteria (Courtemanch et al. 1989; Davies et al. 1991). In 1986, Maine passed a revised water quality classification law (MRSA Title 38 Article 4-A § 464-466) emphasizing its objective, in keeping with that of the CWA, “to restore and maintain the chemical, physical and biological integrity of the State’s waters” and “to establish a water quality classification system which will allow the State to manage its surface waters so as to protect the quality of those waters.” The prevailing practice in most state water quality standards in the United States has been to establish a single, pass–fail boundary for assessing attainment of ALU (Yoder and Barbour 2009; MBI 2011; USEPA 2011, 2013). Typically, the pass–fail boundary is associated with the CWA’s §101(a)(2) Interim Goal of “water quality which provides for the protection and propagation of fish, shellfish, and wildlife and provides for recreation in and on the water,” often referred to as the fishable–swimmable goal. In contrast, Maine’s 1986 law established ALUs for four water quality classifications for rivers and streams (AA, A, B, and C) to articulate goals that span the range from Maine’s interpretation of the CWA Interim Goal (Class C) to the ultimate objective of the CWA “to restore and maintain the chemical,

physical, and biological integrity” (Classes AA and A). The narrative criteria for Maine’s management classes and their associated aquatic life are further defined in ecological terms (Table 1 [narrative aquatic life and habitat criteria for Maine] and Table 2 [definitions of terms]). Maine’s higher classifications (AA, A, B) include aquatic life criteria more stringent than how states have typically implemented the CWA fishable–swimmable Interim Goal. Maine’s water classification law also establishes one class for lakes and ponds (GPA) and three classes for marine waters (SA, SB, and SC) that are similar in construction, but are not discussed further in this publication.

### 1.1.2 Maine’s water classification law and biological standards

The law assigns a class to every waterbody in the state<sup>2</sup> (MRSA Title 38 Article 4-A § 467-469), which establishes the target goal for its water quality and establishes narrative criteria for attainment of ALUs (referred to in this publication as the statutory goal). Some large waterbodies, such as the Kennebec and Penobscot Rivers, are divided into segments that have different statutory goals. Following 1986 passage of the water classification law, the statutory goals were assigned by the state legislature through a public process, balancing ecological, social, and economic needs and values. As illustrated in Case Studies 1 and 8, there is a public process to periodically revisit statutory goals and nominate waterbodies for change in their statutory goals, usually upgrades such as from Class C to Class B. For each water quality class, the law defines

1. designated uses that articulate environmental and social expectations (e.g., water supply, recreation in and on the water, hydropower, agriculture and industrial supply, and habitat for fish and other aquatic life)

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1 The CWA designated use of “aquatic life support” is commonly referred to as “aquatic life use” (ALU).

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2 Waters of the state are defined in MRSA Title 38 Article 3 §361-A as “any and all surface and subsurface waters that are contained within, flow through, or under or border upon this State or any portion of the State, including the marginal and high seas, except such waters as are confined and retained completely upon the property of one person and do not drain into or connect with any other waters of the State, but not excluding waters susceptible to use in interstate or foreign commerce, or whose use, degradation or destruction would affect interstate or foreign commerce.”

**Table 1.** Maine’s tiered narrative aquatic life and habitat criteria and numeric dissolved oxygen and bacteria criteria for rivers and streams; current EPA adopted ambient water quality criteria for toxic substances apply to all classes. (MRSA Title 38 Article 4-A § 464-466)

Class	Management Objectives	Dissolved Oxygen	Bacteria ( <i>E. coli</i> )	Biological Standards and Habitat Characteristics
AA*	Highest-quality water, minimal human interference; No discharges allowed; No impoundment allowed	As naturally occurs	As naturally occurs	Habitat shall be characterized as free-flowing and natural; Aquatic life shall be as naturally occurs
A*	High-quality water with limited human interference; Discharges limited to noncontact process water or highly treated wastewater of quality equal to, or better than, the receiving water; Impoundments allowed (see Management and Biological Standard)	7 ppm or 75% saturation	As naturally occurs	Habitat shall be characterized as natural; Aquatic life shall be as naturally occurs
B	Good-quality water; Discharge of well-treated effluent with ample dilution permitted; Impoundments allowed (see Management and Biological Standard)	7 ppm or 75% saturation  October 1–May 15: 9.5 ppm	May 15 to September 30– Geometric mean: 64/100 ml  Instantaneous (single sample): 236/100 ml	Habitat shall be characterized as unimpaired; Discharges shall not cause adverse impacts to aquatic life; Receiving water shall be of sufficient quality to support all aquatic species indigenous to the receiving water without detrimental changes in the resident biological community
C	Acceptable water quality, achieves the interim goals of the Clean Water Act (fishable–swimmable); Discharge of well-treated effluent permitted; Impoundments allowed	5 ppm or 60% saturation; D.O sufficient to support salmonid spawning, incubation, and survival in identified areas	May 15 to September 30– Geometric mean: 126/100 ml  Instantaneous (single sample): 236/100 ml	Habitat for fish and other aquatic life; Discharges may cause some changes to aquatic life provided that the receiving waters shall be of sufficient quality to support all species of fish indigenous to the receiving water and maintain the structure and function of the resident biological community
Impoundments in Classes A and B.	Riverine impoundments managed for hydropower generation and not classified as Great Ponds	Same as for the assigned class except where stratification occurs	Same as for the assigned class	Support all species of fish indigenous to those waters and maintain the structure and function of the resident biological community

\*The narrative aquatic life standard is the same for Class AA and Class A.

2. narrative and numeric criteria (physical, chemical, and biological) used to determine if water-bodies attain all their designated uses
3. technical terms used in the law (Tables 1 and 2)

For rivers, the classes range from Class AA with the highest expectations for water quality and greatest restrictions on human activity to Class C (Maine’s

interpretation of the CWA Interim Goal) having more opportunity for human activities (MDEP 2002). The state independently evaluates chemical, physical, and biological criteria results (including independent assessment of the results from different assemblages, e.g., algae and macroinvertebrates) to determine overall attainment of assigned class (MRSA Title 38 Article 4-A § 464-470). This publication addresses assessing attainment of biological

**Table 2.** Definitions of terms used in Maine’s narrative aquatic life criteria. (MRSA Title 38 Article 4-A § 464-466)

Term	Definition
Aquatic life	Any plants or animals that live at least part of their life cycle in fresh water
As naturally occurs	Conditions with essentially the same physical, chemical, and biological characteristics as found in situations with similar habitats, free of measurable effects of human activity
Community function	Mechanisms of uptake storage and transfer of life-sustaining materials available to a biological community that determine the efficiency of use and the amount of export of the materials from the community
Community structure	The organization of a biological community based on numbers of individuals within different taxonomic groups and the proportion each taxonomic group represents of the total community
Indigenous	Supported in a reach of water or known to have been supported according to historical records compiled by state and federal agencies or published in scientific literature
Natural	Living in, or as if in, a state of nature not measurably affected by human activity
Resident biological community	Aquatic life expected to exist in a habitat that is free from the influence of the discharge of any pollutant, which shall be established by accepted biomonitoring techniques
Unimpaired	Without a diminished capacity to support aquatic life
Without detrimental changes in the resident biological community	No significant loss of species or excessive dominance by any species or group of species attributable to human activity

criteria for macroinvertebrates only. Classes AA and A share the same narrative criteria for the support of aquatic life, but Class A allows more human activities (hereafter the biological criteria for Classes AA and A are referred to as A). If biological monitoring reveals that conditions in a waterbody are worse than the waterbody’s assigned statutory goal (i.e., Class A, B, or C), it is deemed unacceptable and is reported as impaired as required by the CWA §303(d) (MDEP 2002, 2010, 2012). If conditions are found to exceed the criteria of the assigned statutory goal, that waterbody must be considered a potential candidate for upgrade to the next higher classification to maintain the high-quality condition (MRSA Title 38 Article 4-A § 464.4). *Case Study 1* and its figures present a summary of water quality reclassification in Maine since implementation of the water classification law in 1985. Section 1.2.2 and the appendices provide methods and standard protocols.

Upon passage of the revised classification law, we set about to develop a system of numerical biological criteria (biocriteria) that could objectively assess biological integrity, satisfying both the objective of the Clean Water Act and the narrative aquatic life goals of Maine’s Water Quality Classification Law. We substantially altered the traditional approach and means of assessing water quality standards by establishing goals and

narrative and numeric criteria for biological condition (Courtemanch and Davies 1988; Courtemanch et al. 1989; Davies et al. 1991). Three management classes (i.e., A, B, C) delineate boundaries between levels of aquatic life condition (Courtemanch et al. 1989; Courtemanch 1995; Shelton and Blocksom 2004; USEPA 2005; Davies and Jackson 2006). Each water quality classification is designed to protect a qualitatively different level of biological condition (further explained in Table 1 and Figure 1). Terms are defined in statute to enable consistent ecological interpretation of current biological condition relative to statutory goal conditions assigned by the Maine Legislature (Table 2).

Maine’s narrative aquatic life criteria boundaries are based on the theoretical subsidy–stress gradient model of Riebesell (1974), which was further developed by Odum et al. (1979) and Odum (1985). These authors provided a conceptual model of expected patterns of biological change in response to aquatic stressors (Figure 1). This model describes a parabolic response to increasing inputs of usable resources and a negative, step-change response to toxic conditions. Odum’s model uses the term subsidy to refer to the commonly observed phenomenon of a positive response in biological attributes (e.g., higher density, richness, biomass) when a system is subjected to mild or moderate enrichment (e.g.,

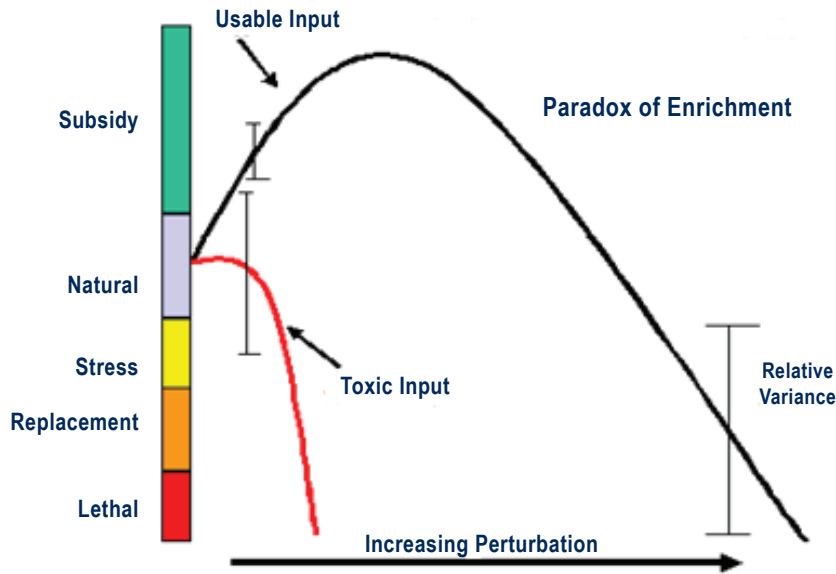


Figure 1. Odum subsidy–stress gradient provides the ecological theory basis for Maine’s aquatic life use descriptions (redrawn from Odum et al. 1979). Some disturbances have an enriching or subsidizing effect on biological assemblages because they provide more than normal usable resources (nutrients, organic matter). Inputs in excess of what can be processed by the resident community have a detrimental effect (increased biochemical oxygen demand, accumulation of unusable resources) and lead to negative community response. Toxic or poisonous inputs have a detrimental effect.

increased input of usable nutrients and organic matter). At some point, as inputs continue to increase, they begin to induce stress, resulting in a negative biological response. The negative response (e.g., loss of sensitive taxa, loss of ecological function, hyper-dominance expressed as extreme densities of opportunist taxa [Rabeni et al. 1985; Yoder and Rankin 1995b; Richardson et al. 2000]) is due to excessive accumulation of organic matter and consequent changes in the physio-chemical environment that increase stress on aquatic communities. Although Odum’s model may not be a good fit for highly productive, alkaline ecoregions, we found it provides a good basis to describe commonly observed stressor-response relationships in northern New England (Davies and Jackson 2006; Snook et al. 2007). Class AA and Class A represent Maine’s minimally disturbed streams and rivers that, with some exceptions, are naturally low in productivity, with low nutrient concentrations, ion concentrations, and acid-neutralizing capacity (Davis et al. 1978). Class B represents streams and rivers with low to moderate enrichment with attendant subsidy effects. Class C represents enrichment with some evidence of stress effects, but still meeting Maine’s interpretation

of the CWA §101(a)(2) of fishable–swimmable conditions (38 MRSA §465.4; §466).

While natural and ecoregional gradients influence biological communities in important ways, biological responses to human disturbance are frequently far more obvious and abrupt, often overwhelming ecological gradients (May 1977). Stress ecology emphasizes the recognition of biological changes that occur in response to human-caused disturbance (i.e., gradients of environmental quality), as distinct from biological responses to natural gradients such as elevation, climate, alkalinity, stream size, or geographic location. With this in mind, we further refined our expectations for stages of biological degradation by considering classical and applied studies in stress ecology (Margalef 1963, 1981; Gardner and Ashby 1970; May 1973; Cairns 1974 1977 1981a,

1981b; Karr 1981; Matthews et al. 1982; Minshall et al. 1985; Odum 1985; Hughes and Gammon 1987; Hughes 1994; Yoder and Rankin 1995b; Lorentz et al. 1997; Hughes et al. 1998). We concluded that the biological expectations described by these studies, especially the subsidy-stress gradient model, were a useful theoretical basis on which to set management boundaries for Maine stream biota. These broad categories of biological condition also fit well with the state’s other tiered criteria for dissolved oxygen, bacteria, and habitat described in Maine’s Water Quality Classification Law.

### 1.1.3 Biological assessment

The Maine Department of Environmental Protection (MDEP) uses biological assessments to determine if the conditions for aquatic life in rivers and streams attain the narrative and numeric criteria of their assigned statutory goals. The USEPA has defined the term biological assessment (or bioassessment) to mean “an evaluation of the biological condition of a waterbody using biological surveys and other direct measurements of resident biota in surface waters” (USEPA 1990). The assessment may be performed on any of a number of components of the

overall biological community, for example, on diversity of a biological assemblage (i.e., a taxonomic or assemblage group such as algae, invertebrates, fish), on a hierarchical level (individual, population, community), or on a trophic or food-web level (primary producers, secondary producers, decomposers). For the most part, bioassessment in the United States has focused on the fish community (Karr 1981; Karr et al. 1986; Halliwell et al. 1998; Meng et al. 2002), the benthic macroinvertebrate community (Hilsenhoff 1987; Rabeni et al. 1985; Lenat 1988; Heino et al. 2003), a combination of the two (Plafkin et al. 1989; Yoder and Rankin 1995a; Barbour et al. 1999; USEPA 2013), or more recently the benthic algal community (Stevenson and Bahls 1999; Fore 2003; Stevenson et al. 2008, Danielson et al. 2011, 2012). The chosen component is then used as a surrogate indicator of the entire interacting community of aquatic life in the ecosystem, with conclusions regarding the well-being of the assessed component being generalized to conclusions about the health of the entire aquatic community. The work of many water pollution scientists worldwide, beginning in the early 1900s through present day, has contributed to the current sophistication with which different community components are understood (Hynes 1960; Cairns 1974; Karr et al. 1986; Cairns et al. 1993; Wright 1995; Yoder and Rankin 1995a; 1995b; Chessman 1999; Moss et al.

1999; Barbour et al. 2000; Hawkins et al. 2000; Karr and Chu 2000; Llanso et al. 2002; Jenerette et al. 2002; Birk and Hering 2009; Birk et al. 2012).

The subject of this publication is Maine's promulgation in law of narrative aquatic life criteria and the development of a supporting numeric biocriteria rule, based on sample information and statistical analysis of the benthic macroinvertebrate community. Because of the importance and usefulness of assessing multiple assemblages, Maine has also established a statewide algal monitoring and assessment program (Danielson et al. 2011, 2012). Further, the Midwest Biodiversity Institute is developing indices of biotic integrity (IBI) for fish assemblages in large rivers in the major river catchments in Maine (Yoder et al. 2009).

Table 3 presents the developmental chronology of narrative and numeric biological criteria to assess attainment of statutory goals for aquatic life in Maine. Maine's numeric aquatic life criteria, the subject of this publication, were promulgated in 2003 (CMR 06-096, Chapter 579) and have been in continuous use since. Part 2 and the appendices contain details regarding development and validation of the statistical biocriteria model. Part 3 presents example applications of these numeric biocriteria, the management results, and the benefits derived from Maine's biological approach to water quality

**Table 3.** Chronology of biocriteria program development.

Dates	Major Activities
1983–1988	Standardization of field and lab methods for macroinvertebrates in 1987; collection of baseline dataset; methods subsequently refined as documented in Davies and Tsomides (1997, 2002) and MDEP (2014)
1986	Passage of revised ALUs and narrative biocriteria in Maine water quality standards law
1989–1990	Exploratory data analysis; development of electronic database management system (FoxPro) Formation of stakeholder technical advisory committee
1990–1992	Development and testing of macroinvertebrate Phase I linear discriminant model (LDM) using 145 sample dataset
1997–1998	Re-parameterization of Phase I LDM using 228 new samples (n=373) to create Phase II LDM
1999–2006	Standardization of stream algal and wetland macroinvertebrate and algal field methods and collection of baseline datasets
2000–2001	Macroinvertebrate database migration to Oracle and ArcInfo
2001–2007	Exploratory algal and wetland data analysis and database migration to Oracle and ArcInfo
2003	Numeric biocriteria for river and stream macroinvertebrates adopted in rule by the Maine Board of Environmental Protection and approved by the Maine Legislature
2004–present	Use of river and stream macroinvertebrate numeric biocriteria in regulatory decision making; development and testing of 3 new LDM's for stream algae and wetland macroinvertebrates and algae.
2010–present	Use of algal and wetland bioassessment in federal water quality reporting (§303d listing of impaired waters); Pending: adoption in rule of numeric biocriteria for algae and wetlands

decision-making, as recorded over the last 30 years. Part 4 contains the summary and conclusions.

## 1.2 BIOLOGICAL CRITERIA DEVELOPMENT

### 1.2.1 Development of a scientifically and legally defensible tool

In developing numeric biological criteria, our objective was to produce a biological assessment protocol that would fulfill a variety of needs, with the goal of institutionalizing a water management approach that was highly responsive to protection, maintenance, and restoration of high-quality aquatic biological conditions. We required that it would be a scientifically, as well as legally, defensible tool for water quality management for Maine, hence, our objective included promulgation of numeric biocriteria and assessment protocols into the environmental regulations of the state of Maine. We also required a scientifically sound technical approach that would accurately rank the biological condition of unknown sites relative to biota collected from sites minimally disturbed by human activity (Stoddard et al. 2006). As detailed in Part 2, the completed statistical assessment system uses a set of interrelated Gaussian linear discriminant models (Fisher 1936; Flury 1997) consisting of one four-way model to differentiate between four classification groups (A vs B vs C vs Nonattainment [NA]), followed by three additional two-way linear discriminant models for pairwise differentiation to increase predictive success (A vs B-C-NA; A or B vs C-NA; A or B or C vs NA; [Figure 2]).

### 1.2.2 Standard protocols

The MDEP uses standard sampling, quantitative analysis, and assessment protocols that are promulgated in state regulation (Appendices). The assessment procedure includes professional review of final results of the statistical model before issuance of final departmental determinations of water quality standards class attainment (CMR 06-096, Chapter 579; Appendices 4, 5, 6, and 7). The biocriteria rule (summarized in part in Appendix 7) specifies sample characteristics that are appropriate for analysis by the model and gives procedures for evaluating atypical sampling situations. Standard field, laboratory, subsampling, and data management methods are detailed in Appendix 1. Sampling design considerations are provided in Section 2.1.1.

### 1.2.3 Comparison of model outcome to statutory goal

The final step in Maine's regulatory process is to compare a sample's final determination (i.e., the assessment outcome from the statistical model system and professional review of all results) to the statutory goal assigned by the Maine Legislature for the sampled stream site. If the final determination matches or exceeds the statutory goal (i.e., equal to or better than the statutory goal and applicable aquatic life criteria), then the waterbody attains the aquatic life criteria of the statutory classification assignment. Statutory provisions require that waters confirmed to be attaining the standards of a higher classification (for example, due to strength of initial outcome and/or as confirmed by consistent attainment in repeated sampling) be considered for reclassification upgrade to the next higher class (MRSA Title 38 Article 4-A §464). If the final determination is of a lower water quality class, then the waterbody does not attain the aquatic life criteria of its statutory classification and the waterbody is considered either for restoration and inclusion on the Maine Impaired Waters list (U.S Clean Water Act §303(d)) or as a candidate for resampling to confirm nonattainment status. Procedural details are found in CMR 06-096 Chapter 579, MDEP 2012, and Appendices 1, 6, and 7.

These criteria are unique because they are based on a probability of fit for each class. Other water quality criteria, such as dissolved oxygen concentrations, have traditionally been based on fixed threshold values with little heed paid to either environmental or sample variance in making an attainment decision. The statistical assessment system (linear discriminant models-[LDM]) that we developed provides probability outcomes that add valuable information on the level of confidence of the predicted classification. Appendix 8 shows a site data report with taxonomic data and LDM results; Case Study 7 provides a link on Google Earth to all MDEP quality-assured site-specific bioassessment data summary reports.

## Part 2: Model Building and Results

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### 2.1 INTRODUCTION

The development of a statistical model for predicting stream quality classification to be used as a regulatory tool was dependent upon several preexisting conditions within the Maine DEP. The most fundamental prerequisite condition, as described in [Part 1](#), was a legal framework for biologically based water classification, its interpretation, and the buy-in of stakeholders with the philosophy of statistical prediction of attainment of biological class. The second condition was specific biological measures and criteria that reflected actual stream quality. The selection and justification of benthic macroinvertebrates is discussed in detail in [Appendix 1, Section A1.1](#). The third condition was a representative and standardized sampling method for reliably estimating the benthic macroinvertebrate community. A standardized rock substrate sampler was developed, tested, and refined in Maine streams starting in the 1970s (Rabeni and Gibbs 1977; Rabeni et al. 1985; Davies 1987; see [Appendix 1](#) for details). The fourth condition was an a priori classification assignment of sampled streams in the model-building database, a classification that was integrally related to Maine's water classification law (see [Section 1.1.2](#)). The last condition was a sampling design and a database that could be used for predictive model development (see [Section 2.1.1](#)).

With these conditions in place, development of a statistical predictive model was initiated in 1989. A summary of the approach to the model development is outlined in this section. A more detailed presentation, along with the test statistics and model coefficients, is covered in [Appendices 1](#) (data collection and management), [2](#) (modeling approach and structure), [3](#) (assignment of biologists' classification), [4](#) (calculation of indices), [5](#) (use of professional judgment), and [6](#) (determination of class attainment).

#### 2.1.1 Sampling design for data used in model construction

Beginning in 1983, MDEP aquatic biologists collected benthic macroinvertebrate samples from upstream and downstream of most major wastewater discharges, from rivers and streams affected by urban or agricultural land use, from impounded rivers, and from a large number of minimally disturbed (i.e., reference quality) water bodies. The dataset included all

geographic regions in Maine and stream sizes ranging from first to seventh order (see [Appendix 1, Figure A1](#)). The sampling locations were chosen to establish a baseline dataset that reflected a complete condition gradient representing the range of water quality in Maine. When evaluating the effects of a presumed disturbance, upstream samples provided information about the expected local biological conditions in the absence of the source of disturbance. Sampling to assess disturbed locations was conducted in late summer to represent the typical warm, low-flow, low oxygen conditions that exist during times of maximal stress to aquatic macroinvertebrates. Typically, samples were collected from flowing areas such as riffles and runs with eroded substrates. Each sampler had a standardized amount and type of gravel substrate providing comparable colonizing material regardless of the parent substrate on which it was placed. Detailed descriptions of specific sampling methods can be found in [Appendix 1](#), and Davies and Tsomides (2002, 2014).

### 2.2 EXPLORATORY DATA ANALYSIS

#### 2.2.1 Data reduction

The benthic macroinvertebrate community data collected for model development originally included 145 stream samples (1983-1989, including multiple years at some sites); by the time the final model was constructed (1998), it included 373 samples. Benthic macroinvertebrates in the samples were identified to the lowest practical taxonomic level (genus (most), family, or class (Oligochaeta) (see taxonomic counting rules in [Appendix 1, section A1.4](#) and [Appendix 4](#)). This community data resulted in more than 500 potential predictor variables when both individual taxon occurrence and abundance were considered along with calculated community variables such as diversity and richness indices, biological stress tolerance indices, functional feeding groups, food web guilds, and community metrics such as predator/herbivore ratios. Because of the large number of potential variables, various exploratory summaries and data-reduction techniques were used to determine variable intercorrelations /independence and those that explained high percentages of the total benthic community variance. [Table 4](#) lists some of the statistical techniques employed during our early stages of data analysis.



**Table 4.** Statistical techniques used to establish subsets of potential predictor variables.

Exploratory Multivariate Analysis Performed	Literature Source
Hierarchical cluster analysis	Romesburg 1984; Krzanowski and Marriott 1995; Myers and Resh 2002; Rawlings et al. 2003
K-means cluster analysis	Hand 1981; Kaufman and Rousseeuw 1990; Jenerette et al. 2002
Multidimensional scaling	Schiffman 1981; Heino et al. 2003; Roy et al. 2003
Principal coordinate analysis	Anderson and Willis 2003
Principal components analysis	Pielou 1984; Jolliffe 1986; Jenerette et al. 2002
Multiple regression analysis	Rawlings 1988; Chessman 1999; Holtrop and Fischer 2002
Two-way indicator species analysis	Hill 1979; Wright et al. 1984; Gauch 1991; Joy and Death 2001, 2003; Brodersen and Anderson 2002
Log linear modeling	Agresti 1990
Logistic regression	Emmons et al. 1999
Detrended correspondence analysis	Gauch 1991; Jongman et al. 1995; Anchorena and Cingolani 2002
Variance component analysis	Hoaglin et al. 1991

We next applied a series of more rigorous exploratory multivariate analyses (Mosteller and Tukey 1977) to examine factors that might be useful in a natural classification (methods also listed in Table 4). These analyses were used to assess whether a stratified modeling approach might be necessary, i.e., whether separate models based on geographic location or stream order might be necessary. Overall, the analyses did not provide any evidence that a physically or geographically stratified model was warranted. Factor analysis and cluster analysis were used to assess collinearity among variables as a means of reducing the large matrix of potential predictor variables to a smaller set of independent, orthogonal predictors. Final assessment of collinearity was performed during the linear discriminant modeling using backward variable selection (Flury 1997).

## 2.3 STATISTICAL PREDICTIVE MODELING

### 2.3.1 Selection of a predictive classification methodology

In developing a stream and river classification system for Maine, we decided that the policy requirement to categorize samples into one of four a priori water quality groups (A, B, C, and NA for nonattainment if the community assemblage did not conform to the narrative criteria of any class) was compatible with empirical experience in Maine and with ecological theory (Section 1.1.2). Classes AA and A were grouped together because they share the same aquatic life goal of “as naturally occurs.” There are many approaches to predictive classification such as classification trees, logit and multinomial linear models, neural networks, fuzzy clustering, partial least squares regression, and classical discriminant function analysis (Anderson 1984; Greene 1993; Härdle and Simar 2007; Garson 2008; Gerritsen 2008; Abdi 2010). Most of the accessible multivariate methods that have been developed for classification require an a priori set of classes that represent distinct populations. This is especially true of those methods that provide a probabilistic estimate of class membership, a requisite deemed important at the outset of our project. Given this constraint, early in the development of this project we reasoned that construction of a Maine biocriteria discriminant model could serve the state’s regulatory needs to predict probabilities of water quality class membership (i.e., classification attainment) for new samples. The results of the exploratory analysis indicated that a model-building approach based upon either linear (Lachenbruch and Goldstein 1979; Hand 1981; Jobson 1992) or quadratic (James 1985) discriminant analysis could be advantageous. Discriminant analysis is based upon well-tested statistical theory, uses multiple measures of the biological community, estimates model parameters for class prediction that have ecological meaning, and provides probabilistic outcomes for a given water body’s class membership, given that various distributional assumptions are met (Manly 1991; Jobson 1992). Discriminant analyses have been used successfully by investigators to classify natural aquatic communities relative to habitat or environmental variables (Hill 1977; Joy and Death 2001; Myers and Resh 2002; Llanos et al. 2002; Brodersen and Anderson 2002; Hoberg and Feder 2002; Jenerette et al. 2002; Jowett and Richardson 2003; Danielson et al. 2012) or to determine if environmental disturbance has affected water quality or the benthic community

(Moman and Zehr 1998; Moss et al. 1999; Pusey et al. 2000). The experience of some researchers, though, indicates that discriminant analysis does not always lead to a satisfactory community classification (Emmons et al. 1999; Olden and Jackson 2002; Wheeler and Allen 2002; Rawlings et al. 2003) thus we were careful to assess the scientific validity of the final model for the state of Maine's purposes.

### 2.3.2 A priori classification of streams—The basis of prediction of class membership

Statistical classification of biological water classes was a challenge since the only a priori classes that existed initially in Maine were those defined in law by the legislature, i.e., the statutory classes assigned to rivers and streams in Maine's water quality standards (see Section 1.1.1). Maine's statutory classes are goal-based and thus did not always correspond to actual water quality or biological condition of streams in Maine. In addition, Maine's classification system is dynamic and meant to change and evolve over time as stream conditions improve. At first, we considered algorithmic-based approaches of defining a priori water quality groups (i.e., A, B, C, NA) based upon natural patterns in the data. We tested several alternative approaches using objective search algorithms based on combinations of macroinvertebrate community metrics. Objective search algorithms, such as ordination, have been successful in many parts of the world for defining a priori water quality groups (Rabeni et al. 1985; Moman and Zehr 1998; Chessman 1999; Joy and Death 2001; Brodersen and Anderson 2002; Jenerette et al. 2002; Anderson and Willis 2003; Rawlings et al. 2003). Alternative approaches included K-means clustering, two-way indicator species analysis (TWINSPAN), and several multivariate ordination techniques such as

CCA (canonical correspondence analysis) and NMDS (nonmetric multidimensional scaling) (Anderson 1984; Pielou 1984; Manly 1991;). In general, we found the objective search methods unsatisfactory because clusters depended upon the algorithm used and did not reflect differences in water quality (see Appendix 2, Figures A3 and A4)

As an alternative approach to objective statistical methods of defining stream classes, we used expert knowledge/prior experience to assign the samples to the four water quality groups, and we identified response signals (to different levels of human disturbance) for 31 quantifiable measures of macroinvertebrate community structure (Appendix 3 and Tables 5 and 6). This classification process was then followed by validation using objective methods to confirm that the a priori groupings were, in fact, statistically distinguishable. This approach has been well developed (Press 1980). Discriminant analysis and function derivation do not have to rely on classes that only occur in nature. As long as classes are statistically distinct and their members possess a Gaussian distribution within a class, then most assumptions are met (Anderson 1984). To establish a priori groups, MDEP biologists initially evaluated benthic macroinvertebrate community data for each stream sample (without knowing site locations) and assigned samples to an aquatic life condition category. The assignment exercise was followed by independent biologists from the private stakeholder sector who also evaluated a subset of the data. The methodology was based on the degree to which each biologist found that the sampled community conformed to one of the narrative aquatic life criteria (Class A, B, C, or NA) as described in the statute and accompanying definitions (Tables 1 and 2) (Shelton and Blocksom 2004) and as further described by a table of expected metric response characteristics across the water quality gradient represented by the

**Table 5.** Relationship between narrative aquatic life criteria, ecological values, and quantifiable measures.

Narrative ALU Criteria	Ecological Value	Quantifiable Measures
Class A—as naturally occurs	Taxonomic and numeric equality to natural, presence of indicator taxa	Similarity, richness, abundance, diversity, EPT, indicator taxa, biotic index
Class B—unimpaired, maintain indigenous taxa	Retention of taxa and numbers, absence of hyper-dominance, presence of sensitive taxa	Community loss, richness, abundance, diversity, equitability, evenness, EPT, indicator taxa, biotic index
Class C—maintain structure	Resistance, redundancy, resilience, balanced distribution	Richness, diversity, equitability, evenness
Class C—maintain function	Energy transfer, resource assimilation, reproduction	Trophic groups, richness, abundance, community loss, fecundity, colonization rate

**Table 6.** A priori expectations for variable responses across four biological stream condition classes (A, B, C, and NA).

Measure of Community Structure	Relative Findings			
	A	B	C	NA
Total Abundance of Individuals	Intermediate, sometimes low	High	Variable	Variable: often very low or very high
Abundance of Ephemeroptera	High	High	Low	Low to zero
Abundance of Plecoptera	Highest	Intermediate	Low to absent	Zero
Proportion of Ephemeroptera	Highest	Intermediate to high*	Low	Very low to zero
Proportion of Plecoptera	Highest	Variable*	Low	Zero
Proportion of Hydropsychidae	Intermediate	Highest	Variable	Variable, often low
Proportion of Ephemeroptera & Plecoptera	Highest	Variable	Low	Low to zero
Proportion of Glossosoma	Highest	Low to intermediate	Very low to absent	Zero
Proportion of Brachycentrus	Highest	Low to intermediate	Very low to absent	Zero
Proportion of Oligochaetes	Low	Low	Low to intermediate	Highest
Proportion of Hirudinea	Low	Variable	Variable	Variable to highest
Proportion of Gastropoda	Low	Low	Variable	Variable to highest
Proportion of Chironomidae	Lowest	Variable*	Highest	Variable
Proportion of Conchapelopia & Thienemannimyia	Lowest	Low	Variable	Variable to highest
Proportion of Tribelos	Low to absent	Low to absent	Low to intermediate	Variable to highest
Proportion of Chironomus	Low to absent	Low to absent	Low to intermediate	Variable to highest
Generic Richness	Variable	Highest	Variable	Lowest
Ephemeroptera Richness	High	High	Low	Very low to zero
Plecoptera Richness	Highest	High to intermediate	Low to absent	Zero
EPT Richness**	High	Highest	Variable	Low
Proportion Ephemeroptera Richness	Highest	High	Low	Low to zero
Proportion Plecoptera Richness	Highest	Variable	Low	Zero
Proportion Diptera Richness	Low	Variable	Highest	Variable to high
Proportion Ephemeroptera & Plecoptera Richness	Highest	High	Low to intermediate	Low to zero
EPT Richness divided by Diptera Richness	High	Highest	Low to intermediate	Lowest to zero
Proportion Non-EPT Richness	Lowest	Low	Intermediate to high	Highest
Percentage Predators	Low	Low	High to intermediate	Highest
Percentage Collector & Filterer-Gatherers divided by Percentage Predators & Shredders	High	Highest	Intermediate to low	Variable
Number of Functional Feeding Groups Represented	Variable	High***	Variable	Lowest
Shannon-Weiner Generic Diversity	Low to intermediate	Highest	Intermediate	Lowest
Hilsenhoff Biotic Index	Lowest	Low	Intermediate	Highest

\*depending on dominance of other groups

\*\*Ephemeroptera-Plecoptera-Trichoptera Richness

\*\*\*often with all present

water quality classes (Table 6). The panel of biologists received limited habitat data (e.g., depth, water velocity, substrate composition, temperature) in order to evaluate the intrinsic biotic potential of the sampled habitat, but biologists had no knowledge of the site locations or degree of human disturbance. Three of the authors (DLC, SPD, LT) made the assignments independently (Appendix 3). A Delphi technique was employed to reconcile any disagreements among the classifications of the biologists and to accomplish a final consensus assignment of samples into a priori classes (Bakus et al. 1982; Courtemanch 1993; Davies et al. 1995; Walley and O'Connor 2001). The consensus assignments that we developed are referred to as the “biologist classifications.” A similar approach has since been widely employed by U.S. states and Native American tribes to calibrate local biological condition gradients (Davies and Jackson 2006; USEPA 2016).

Because of the importance of the a priori classes as the basis for creating a predictive discriminant model, we sought confirmation of the reproducibility of the biologist classifications from two additional experienced Maine macroinvertebrate biologists not affiliated with MDEP. Overall concurrence with the consensus assignment we developed and then assessed by the independent biologists was 83% for one biologist and 90% for the other biologist (Table 7). We decided to use the biologist classifications as the a priori groups, because the purely algorithmic approaches, such as the K-means clustering, did not distinguish differences in biological response to differing levels of human disturbance (see Appendix 2, Figure A3). That is to say, they did not produce groups of samples that were related to a water quality gradient as clearly as did the biologists classifications.

**Table 7.** Summary of non-MDEP biologists’ percentage concurrence with MDEP biologists’ rankings of sites.

Class	Biologist 1 (n=40)	Biologist 2 (n=40)
	----- % -----	
A(10)	80	90
B(10)	60	80
C(10)	90	90
NA(10)	100	100
Total	83 (33/40)	90 (36/40)

### 2.3.3 Approach to the construction of a system of hierarchical discriminant models

We first attempted to build a single four-way linear discriminant model (LDM) based on the four a priori biologists’ water quality group assignments (A, B, C, and NA, see Appendix 2, section A2.4) to the sampled streams. We used the procedures of Wilkinson (1989), employing Fisher’s discriminant analysis (Fisher 1936; Hand 1981). We performed the initial modeling attempt on 145 samples, and after five years of use and additional data collection, we performed the model reparameterization on 373 samples (Table 3, “Phase I”). We used stepwise discriminant analysis (James 1985; Krzanowski and Marriott 1995; Horrocks and Ogden 2003), and iterative backward selection (Flury 1997) to select variables based upon their ability to maximize distinctions between water quality classes (Mahalanobis distance, Manly 1991). We applied multivariate analysis of variance (MANOVA) and univariate analysis of variance (ANOVA) to assess the significance of variables in detecting differences between the a priori biologist classifications (Appendix 2, section A2.4). ANOVA and MANOVA helped determine how significant a single variable was in discriminating at least one biological condition group from another. We used ANOVA only as an informal, subjective assessment since discriminant analysis is a multidimensional analysis that uses linear combinations of variables, not the individual variables acting independently (Kendall 1987). We eliminated highly correlated, redundant, or insignificant variables from consideration (Flury 1997). Thus, we caution that the exclusion of a variable does not mean that it is not an important predictor, but rather that it might be highly correlated with another variable that is also a good predictor. This iterative process included an ongoing evaluation of interim results for consistency with ecological goals described in Maine’s narrative water quality standards. All of the stepwise approaches, parameter estimation, ANOVA and MANOVA analyses were performed in the statistical analysis package SYSTAT (Wilkinson 1989).

Early on in the development of linear discriminant functions for the prediction of water quality class membership, one of us (FAD) investigated the use of probabilities derived from a Gaussian probability density function estimated by a linear discriminant function as a random variable input to subsequent linear discriminant functions. This practice had not been developed previously by statistical modelers, but was legitimately borne out by many theoretical treatments

regarding the Gaussian distribution (e.g., Lukacs and King 1954). As described later in this publication, we ultimately constructed a set of interrelated Gaussian linear discriminant models (Figure 2; Appendix 8) consisting of one model to differentiate between four classification groups (four-way model) and followed by three additional linear discriminant models that use results of the four-way model, but differentiate between only two groups at a time: “C or better,” “B or better,” and “A” (referred to as the two-way models).

Unlike the four-way linear discriminant model that attempted to place samples into one of four classes, the two-way discriminant models considered only two classes to distinguish at a time (Figure 2). The C-or-better model was designed to distinguish the NA group from an aggregate class formed by combining the A, B, and C classes. Thus, the C-or-better model determines whether a sample attains at least Class C aquatic life criteria. Similarly, the B-or-better model was designed to distinguish the A and B classes from the C and NA classes and determines if a sample attains at least Class B aquatic life criteria. Finally, the A model was designed to distinguish the A class from an aggregate class consisting of the B, C, and NA classes.

The three two-way predictive discriminant models include variables derived from the probabilities of group membership produced by the four-way model ( $p_{A1}$ ,  $p_{B1}$ ,  $p_{C1}$ , and  $p_{NA1}$ ; see Appendix 2, section A2.5, and Appendices 4, 5, and 6). The MDEP biocriteria rule (CMR 06-096, Chapter 579) refers to the four-way discriminant model as the “first-stage” model since it is run first; the rule refers to the three two-way discriminant models as “second-stage” models. The C-or-better model includes a variable that is the sum of  $p_{A1}$ ,  $p_{B1}$ , and  $p_{C1}$ . Similarly, the B-or-better model includes a variable that is the sum of  $p_{A1}$  and  $p_{B1}$ . Finally the A model includes  $p_{A1}$  as one of its variables. The use of a multivariate axis, such as a discriminant function or a principle component axis, as a predictor is not new (Hotelling 1957; Kendall 1966, 1987; Jolliffe 1986). The sums of normally distributed random variates are themselves normally distributed variates (Anderson 1984; Allen 1990).

The likelihood (probability) of each of the four biologist class memberships in the first-stage model was used as one of several additional predictors in each of the two-way linear discriminant models as follows: first, the likelihood of biologist class membership was estimated by one of four linear axes, a linear combination of each of the nine predictor coefficients and an intercept, each of whose values were dependent upon the class being

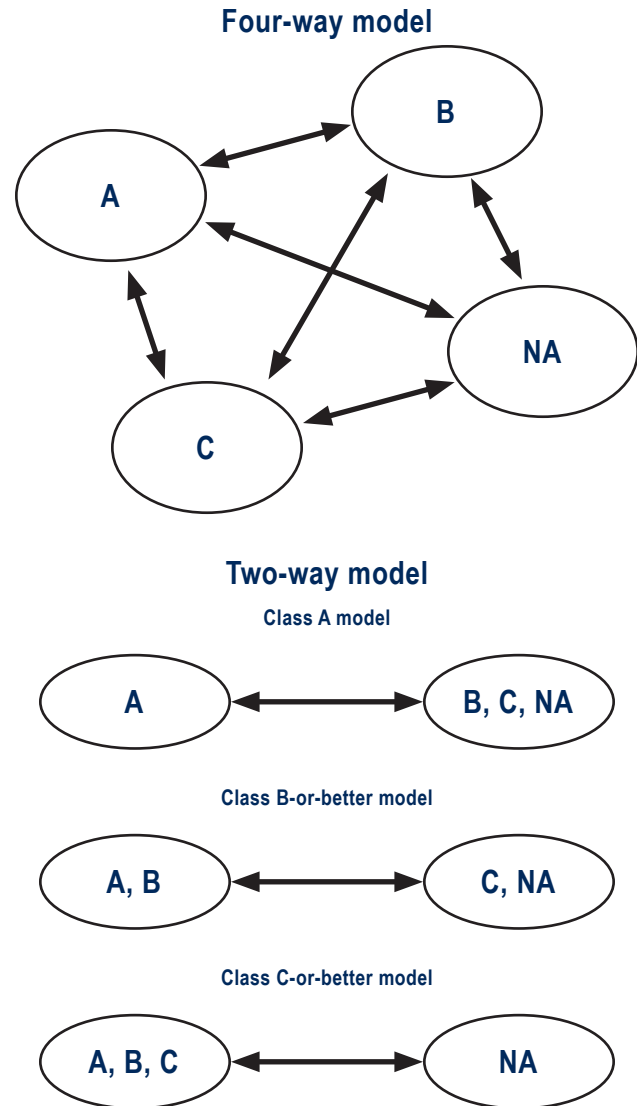


Figure 2. Schematic of four-way and two-way model relationships.

predicted (see Appendix 2, Table A5). To fit the Class A two-way model, the likelihood of Class A membership was used as an input or predictor variable for the two-way model for Class A prediction compared to the other three classes (B, C, and nonattainment) (Figure 2).

Backward stepwise discriminant analysis was used to select the best additional predictor variables for the new two-way Class A model. An identical methodology was used to select predictor variables and fit the B-or-better model and the C-or-better model. In these two models, the likelihood for B or better was derived by adding the probabilities estimated by the first-stage model for the A and B biologist class memberships, and the likelihood for C or better was derived by adding the probabilities estimated by the first-stage model for the

A, B, and C biologist class memberships (see Appendix 2, section A2.5). When used in a hierarchical sequence, the first-stage and second-stage models increased the accuracy of predicting the four biologist classes (Appendix 2, Tables A12–A14).

We used the two-way model approach for two reasons. First, the MDEP is primarily interested in a one-tailed prediction of classification. The pertinent question, in terms of identifying a need to initiate management action, is whether or not a site is attaining at least its statutory goal. For example, a stream with the statutory goal of Class B would attain its goal if the model placed it in either the A or B class. The second reason that pair-wise models are used is that they allow for greater statistical discrimination than models, such as the four-way linear discriminant model, that attempt to isolate each class from every other class. This is particularly true because Class A sites often exhibit low macroinvertebrate densities and low richness due to the naturally oligotrophic conditions of Maine waters. Class A sites therefore may have similarities to some NA sites with low macroinvertebrate densities due to pollution or habitat stressors, as well as to some Class B sites. This confounding characteristic makes accurate prediction difficult and probability scores less confident.

## 2.4 RESULTS—HIERARCHICAL PREDICTIVE MODELS

### 2.4.1 The biologist classification system

The prior experience of biologists (expert judgment) reflected in the biologist classifications and the results of statistical analysis were consistent with the characteristic responses predicted by Odum's subsidy-stress gradient model (Figure 1) and supported the biologists' experience that structurally and functionally distinct biological groups exist across a gradient of water quality. We commonly observed the parabolic pattern of biological change consistent with Odum's enrichment hump (Figure 1). For example, increased densities of some macroinvertebrate taxa (e.g., *Acroneturia* spp., *Stenonema* spp., *Maccaffertium* spp.) and overall increased richness are commonly observed at sites enriched by elevated total phosphorus, relative to nonenriched sites (Figure 3). Empirical data demonstrate the subsidy hump in Maine streams, shown in Figures 3 and 4 using conductivity (ionic strength) as a generalized surrogate for enrichment. Further, known toxic locations commonly show precipitous linear declines in many biological response variables relative to undisturbed locations. Such observations are consistent with Odum's predicted

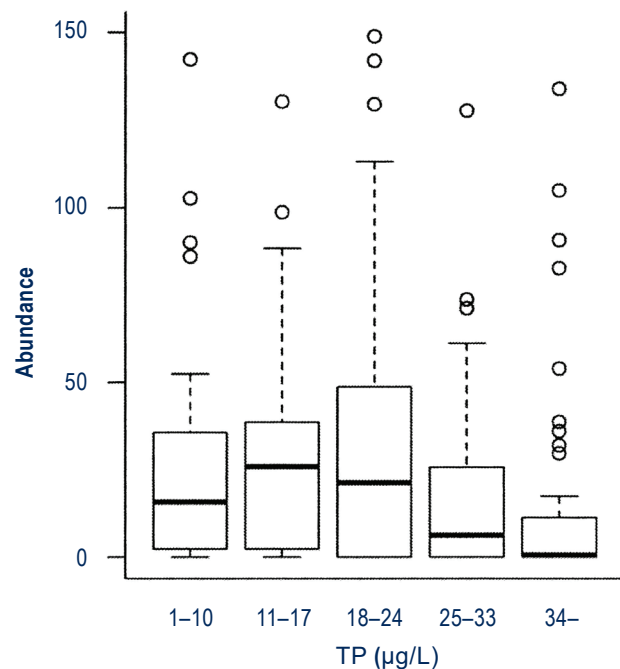


Figure 3. Empirical evidence supporting Odum's subsidy-stress gradient model: Response of summed abundance of *Acroneturia*, *Stenonema*, and *Maccaffertium* to increasing enrichment, as represented by different total phosphorus (TP) range (n=273).

stress response to toxic conditions (Figure 1; Figure 4, Conductivity > 200).

### 2.4.2 The hierarchical predictive model system

The development of the hierarchical system of discriminant models is described in detail in Appendix 2. The first-stage or four-way discriminant model consisted of nine predictor variables (Appendix 2, Tables A5 and A6). Variable definitions and computational algorithms are shown in Appendix 4. The prediction accuracy (correct classification vs misclassification of biologist-classified streams) was highest for Class A sites (72.5% correct) and lowest for Class C sites (62.3%). All possible permutations of accuracy in predictions of classifications are shown in Table A7 in Appendix 2. A jackknife approach was used to determine if large variation in prediction occurred if subsets of the data were used to fit the model. This was not the case. The model coefficients were fairly invariant to variation in the data used for estimation of coefficients and prediction of the classification of the sample sites (Appendix 2, Table A8).

However, because correct classification of the biologist stream class rankings ranged between 65.3% and

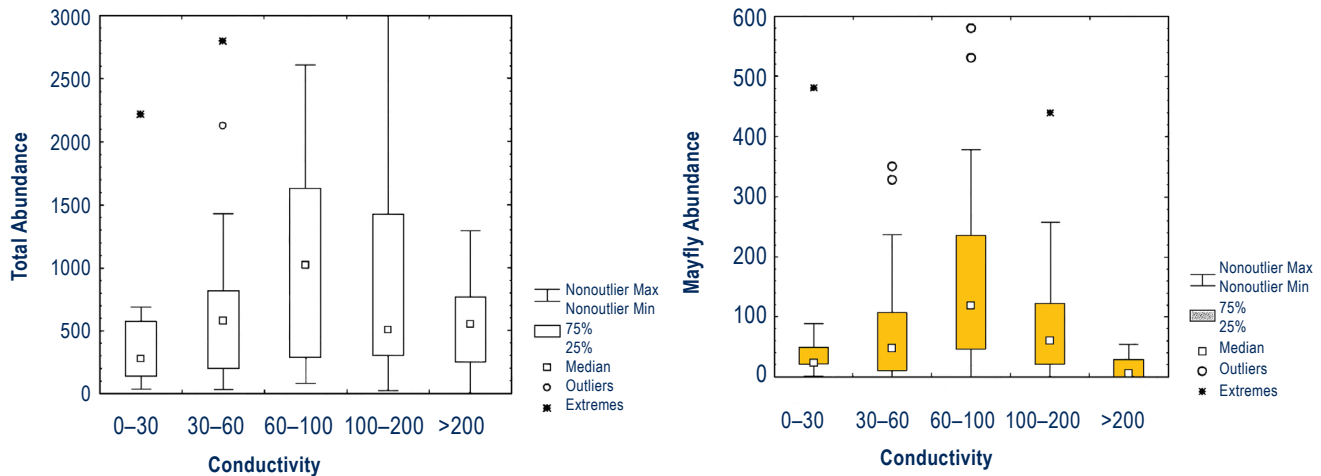


Figure 4. Empirical evidence supporting Odum's subsidy-stress gradient model: Response of taxonomic density and richness variables to increasing enrichment, as represented by different conductivity ranges in mmho/l, (n=285 samples).

72.5% in the four-way model (Table A7), a second tier of three independent two-way linear discriminant models was developed to be used in a hierarchical manner subsequent to the initial classification by the first-stage model (see Figure 2).

The three, two-way models, Class A, Class B or better, or Class C or better, were developed with 6, 7, and 4 predictor variables, respectively (Appendix 2, Tables A9–A11). In Appendix 2, Figures A6–A8 show the distribution of each of the significant predictor variables measured in the 373 sampled streams. These graphs show the overlap of the metrics as they are portioned out among the biologist-assigned classes and demonstrate that the predictors with the higher levels of significance tend to have less overlap in their populations between predicted groups of classes.

All of the metrics (n=23 total for first-stage and second-stage models) used in the hierarchical model system for prediction of the biologists' water quality classes are listed in Appendix 4. Coefficients for the three, two-way models are listed in Appendix 2, Table A15. These coefficients are used to estimate the probabilities of biologist class memberships that are used in the MDEP stream water quality regulation and licensing. Appendices 5 and 6 illustrate the hierarchical structure of the predictive models and the sequential process of estimating biologists' class membership probabilities. Appendix 7 provides a detailed overview of how the model prediction probabilities are incorporated into an integrative professional judgment in making a final regulatory decision on class attainment.

#### 2.4.3 Accuracy in predictions of the hierarchical predictive model system

The accuracy of the final hierarchical system of predictive models was 90.0% for Class A (Appendix 2, Table A12), 96.5% for Class B or better (Appendix 2, Table A13), and 96.1% for Class C or better (Appendix 2, Table A14). These high levels of accuracy in classification were considered well suited for regulatory management decisions along with the use of professional judgment criteria (see Appendix 7), especially since the majority of model results had predictive probabilities >0.90, indicating a strong association to a particular class.

#### 2.4.4 Validation and robustness in predictions of the hierarchical predictive model system

Robustness and quasi-validation of the final hierarchical model system was attempted by both jackknife analysis, and using an independent dataset, after the final first-stage model was built. The first method for validation of the discriminant functions was to perform a jackknife discriminant procedure (Jobson 1992: 278; Miller 1998: 220). This procedure iteratively estimates the discriminant function by using multiple subsets of the data (a small percentage of the data left out for each iteration). The jackknife procedure assesses the percentage correct classification of data not used to build the models and provides an estimate of sample error as it relates to coefficient estimates and misclassifications.

The jackknife results are listed in Appendix 2, Table A16. The procedure demonstrated that the level

of correct classification was as high or higher with the subsets of original data as with construction of final discriminant models with the entire data set ( $n=373$ ). This suggests that the hierarchical model system is robust to aberrations in the data and not sensitive to changes in the sample size of our model-building data set. This is important in two ways. First, it suggests that the sample size of the model-building data set is large enough for the development of a predictive regulatory model for Maine, and second, that the final model is probably robust enough to predict stream classification across the diversity of Maine's streams.

The independent validation was performed on a small data set of 34 streams (see Appendix 2, Table A17). The final first-stage model was used to predict the classification of 34 independent streams. It performed as well as for classification of the original model-building dataset, with an overall correct classification of class A streams of 75%, class B streams of 100%, class C streams of 71%, and nonattainment streams of 100%. Therefore, the predictive model has generality beyond the model-building data set.

#### 2.4.5 Comparison of hierarchical model results to Maine RIVPACS model

Following model building, further objective verification of the interpretive validity of the biologist classification was confirmed when the assignments of the final MDEP biocriteria model were compared to a RIVPACS (River Invertebrate Prediction and Classification System) model (Hawkins 2006) that was developed using Maine's initial stream sample data set. Maine's RIVPACS results indicate a biological gradient from high-quality to lower-quality condition and provides evidence to rebut criticism of circularity flaws in a model built from a priori biologist classification assignments (Figure 5). The distribution of observed/expected (O/E) scores for samples assigned to aquatic life management classes by the discriminant models demonstrates a decline in expected taxa. This decline parallels the narrative standards and biological expectations in the biological condition gradient defined by Maine's aquatic life use standards and biocriteria. Both Class A and Class B distributions fall within the expectations for reference-quality streams, affirming the narrative criteria for Class A ("as naturally occurs") and for Class B ("without detrimental changes to the biological community") (Tables 1 and 2). Figure 5 also shows the 10th percentile of reference falling somewhat

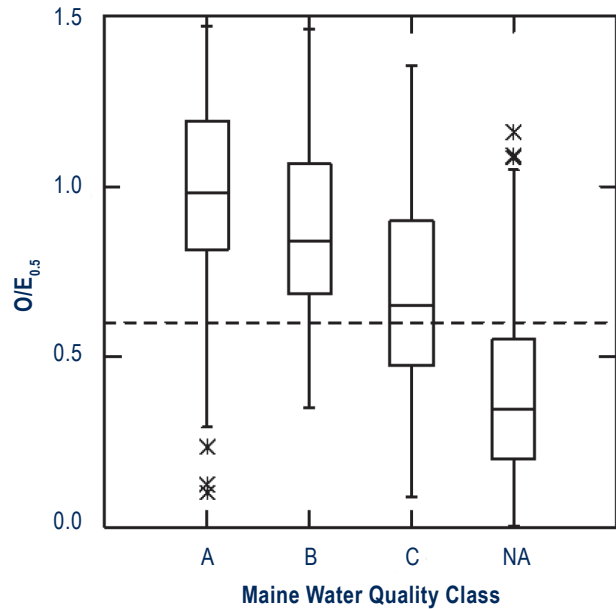


Figure 5. Distribution of RIVPACS predictive model O/E values within and between the Maine LDM water quality classes. The horizontal dashed line represents the 10th percentile of reference site O/E values (Hawkins 2006).

below the median for the Class C distribution, thus demonstrating that most Class C streams are being managed to preserve the Class C narrative standards that require conditions that "maintain the structure and function of the resident biological community." The distribution reflects that allowances have been made in statute for the inevitable changes in biological condition that may occur given the greater human disturbance associated with waterbodies assigned to Class C. The RIVPACS model also demonstrates that our model does not exhibit Type I error (false alarm) because nearly all NA samples fall below the 10th percentile of reference (Figure 5).



## Part 3: Management Applications of Numeric Biocriteria in Maine

### 3.1 INTRODUCTION AND HISTORY

Since the 1960s, prior to adoption of the federal CWA, Maine water quality law has had a tiered structure with the intent to recognize the gradient of water quality conditions in the state and, through a planning approach, to optimize the condition of all waters (Courtemanch et al. 1989; Courtemanch 1993, 1995). The inclusion of aquatic life criteria based on a clearly stated biological condition gradient offers many management advantages (Davies and Jackson 2006; Hering et al. 2010). Biological criteria can be beneficially applied through all the phases of water quality management depicted in the USEPA's water quality-based approach (USEPA 2012 and Figure 6). Maine has adapted USEPA's depiction of the water quality-based approach by using information about biological condition and biological response to human disturbance, to optimize protection and restoration of aquatic life resources in the state (Figure 7 and Case Studies).

The following case studies are provided to demonstrate how Maine has made use of biological criteria in the decision process of each step of the water-quality-based approach to resource management. Important management benefits can be attributed to the potential for more precisely defined goals for aquatic life condition that provide more transparent and easily understood incremental management targets for the designated aquatic life use of each water quality class. Standards that are stated as precisely defined condition levels, combined with scientifically sound numeric biocriteria to assess those condition levels, offer a broader range of management options to ensure that resource goals



Figure 6. U.S. Environmental Protection Agency depiction of the water-quality-based approach, in relation to water quality standards. This approach emphasizes the overall quality of a water body and provides a mechanism through which impact is controlled based on the intrinsic conditions of that body of water and the standards set to protect it (USEPA 2012).

and objectives are neither underprotective of existing high-quality resources, nor needlessly burdensome for waters that may be permissibly affected by pressures from allowed human activities.

By designating biological condition categories for waterbodies in water quality standards, Maine has been able to document incremental improvements in biological condition and general water quality trends throughout the state over the decades since passage of the CWA (Rabeni and Gibbs 1977; Rabeni et al. 1985; Davies 1987; Davies et al. 1999). Standards that describe a water quality gradient anchored in natural conditions provide the mechanism to objectively and transparently assess the biological condition of a waterbody relative to naturally expected biological response and to measure, assess, and maintain incremental improvements (Davies and Jackson 2006; USEPA 2005, 2011, 2013, 2016).

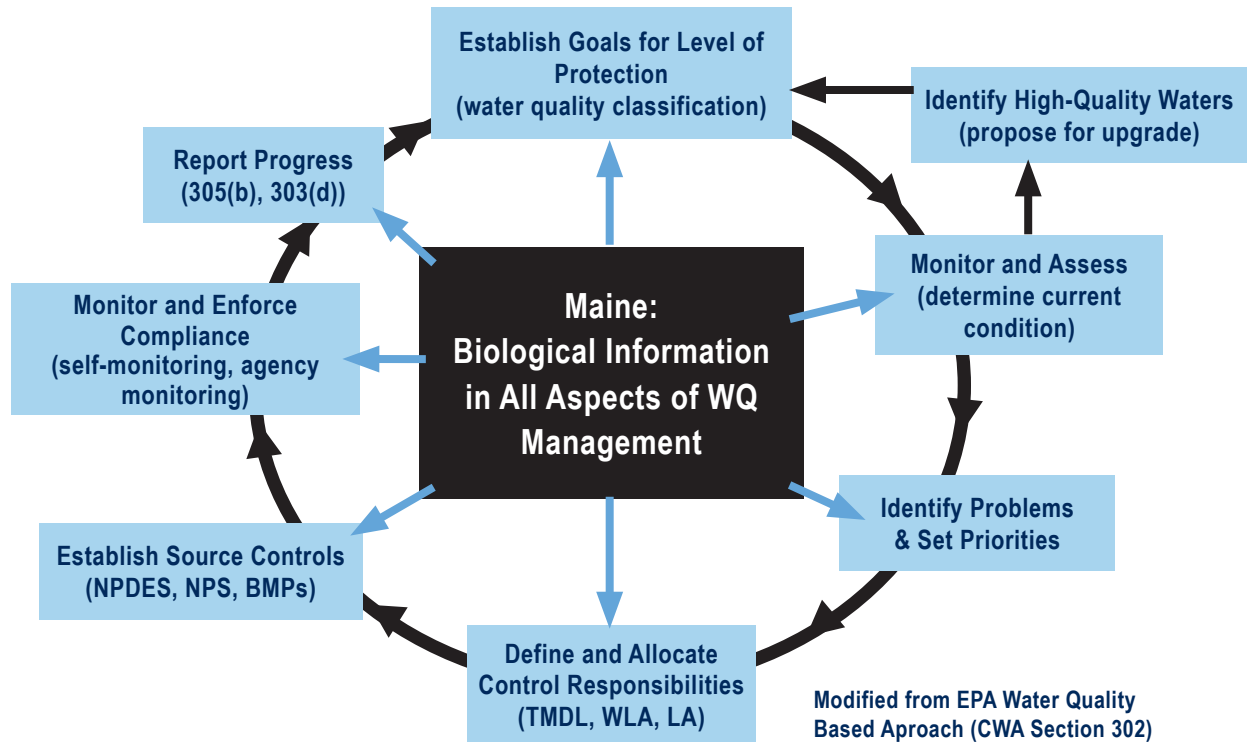


Figure 7. Maine revisions to U.S. Environmental Protection Agency water-quality-based approach as applied in Maine, using biological response information to optimize the condition of waterbodies.

### 3.2 Case Studies—Use of Biological Criteria to Manage Maine’s Water Quality

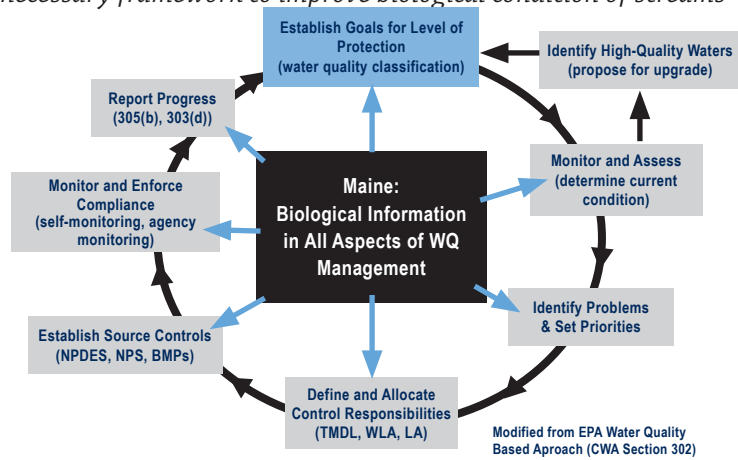
The following case studies describe the use of predictive biological models for management of Maine’s streams and rivers since 1987. They are arranged around the loop of the water-quality-based schematic shown in Figure 7. Each of the case studies illustrates a particular use of the predictive models and the integration of numeric biological criteria into water quality management decisions.

#### List of case studies

1. Establish goals for protection of aquatic life: *Goal-based management planning to optimize aquatic life conditions.*
2. Monitoring and assessment to determine existing condition relative to goal condition: *Long-term point-source monitoring to document attainment of biological criteria on the Penobscot River, Maine.*
3. Identify problems and set management priorities: *Detection and management of an emerging problem—urbanization and nonpoint source impacts.*
4. Define problem and allocate control responsibilities: *Alternative total maximum daily loads (TMDL) to manage impacts of high impervious cover on aquatic life.*
5. Establish source controls for point sources and nonpoint sources: *Using biological information to set permit limits in the absence of ambient chemical criteria.*
6. Monitor and enforce compliance through self-monitoring or agency monitoring: *Use of biological criteria for enforcement actions.*
7. Report progress: *Communicate bioassessment results through interactive maps and traditional reports.*
8. Retention of water quality improvements: *Protection of high-quality waters and implementation of restoration and protection strategies to optimize biological condition.*

## Case Study 1. Establish Goals for Protection of Aquatic Life

Clear, technically rigorous goal statements are a necessary framework to improve biological condition of streams and rivers. Defining the water quality goals of different water quality classes is both a technical task and a public policy task. Most U.S. states have established a single pass-fail threshold, representing their interpretation of the federal CWA Interim Goal. In contrast, Maine has established four water quality classifications for rivers and streams (AA/A/B/C) that span the range from Maine’s interpretation of the CWA Interim Goal (Class C) to the ultimate CWA objective “to restore and maintain chemical, physical and biological integrity” (Class AA). All rivers and streams in Maine are assigned to one of the four classifications in Maine’s water quality standards for planning and management purposes.



### Goal-based management planning to optimize aquatic life conditions

As previously described, the Maine Legislature passed a revised water quality standards and classification law in 1986 (MRSA Title 38 Article 4-A §464-466) establishing narrative biological criteria for four aquatic life use classes for rivers and streams. This law set in motion a process involving the public, the state environmental agency, and the Maine Legislature to assign all Maine waters to an appropriate classification goal. MDEP used all available monitoring data and information about biological and/or water quality conditions to initially propose the statutory classes for stream and river segments in the 1986 law. Many waters that lacked current monitoring data retained their previous water quality goals (generally Class B, except for some urban or industrialized areas, which were Class C) until MDEP obtained monitoring data or other evidence to

recommend a different class. Table 8 shows the change of classifications over time and unprecedented gain (by any state) in statutory protection for existing high-quality waters.

Maps spanning the period between 1987 and 2012 (Figures 8 and 9) show the past and present-day distribution of water quality classifications. These maps show that approximately 99% of Maine’s rivers and streams are currently managed at levels of protection higher than the commonly applied interpretation of the CWA Interim Goal (i.e., Class C). In the 25 years since 1987, the legislature has assigned 13,955 miles of waters to a Class A or Class AA management goal, an increase of 25.5% (MDEP 2002, 2010, 2012). These classification upgrades have mostly been drawn from Class B and Class C waters where biological monitoring data demonstrated the ready potential for, or the actual achievement of, the standards of Class A or Class AA. Regarding “ready

Table 8. Change in legislative assignment of statutory classification of rivers and streams in Maine from 1987 to 2012, reflecting a shift in miles of protected and improved water quality.

Year	Class AA		Class A		Class B		Class C		Total miles
	Miles	%	Miles	%	Miles	%	Miles	%	
1987	985	1.7	13,471	24.3	34,515	62.2	6,552	11.8	55,523
2012	3,404	6.2	25,007	45.2	26,313	47.5	614	1.1	55,338
Change*	+2,419	+4.5	+11,536	+21	-8,202	-14.7	-5,938	-10.7	

\*Differences shown in the total miles column (185 miles) are due to differences in precision and accuracy of mapping data between 1987 and 2012.

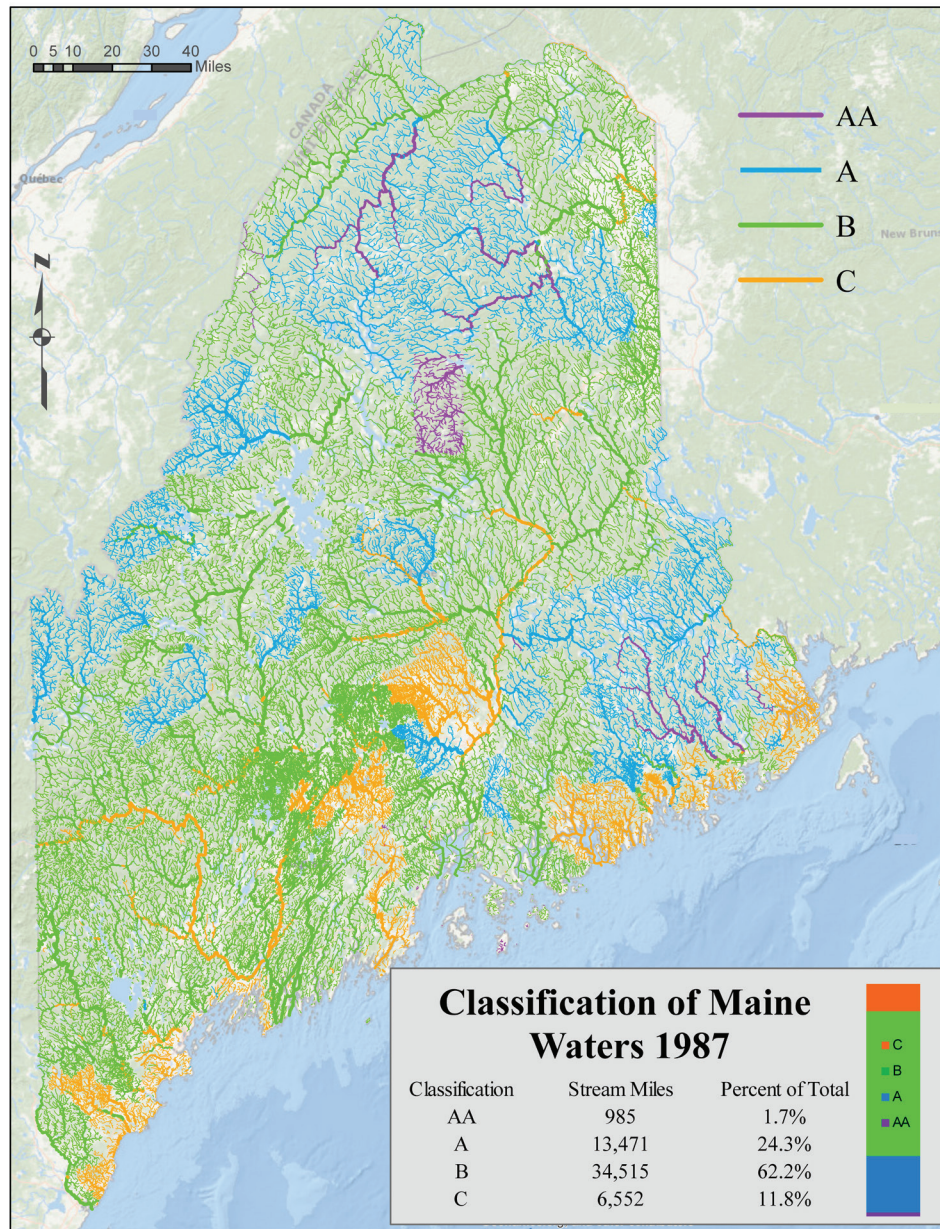


Figure 8. 1987 water quality classifications, prior to revisions to Maine’s water quality standards law. Colored histogram shows percentage distribution of the four classes.

potential,” for example, the Kennebec River was preemptively upgraded when the decision to remove a major dam was finalized, but prior to dam removal. Without numeric biological criteria and a gradient of aquatic life management classes, high-quality waters often go unrecognized, undervalued, and unprotected (USEPA 2005, 2013, 2016). The same is true when improvements in biological condition have occurred due to investment in remediation. The communication value of condition classes enhances public understanding of existing conditions, problems, and restorable target conditions, and

provides an important tool in building public support for the often substantial investment that is required to restore aquatic resources (Courtemanch et al. 1989; Davies and Jackson 2006; USEPA 2011).

**For further information:**

Courtemanch, D.L. 1995. Merging the science of biological monitoring with water resource management policy: Criteria development. *Biological Assessment and Criteria: Tools for Water Resource Planning and Decision Making*, ed. W.S. Davis and T.P. Simon. CRC Press, Boca Raton, FL. pp 315-326.

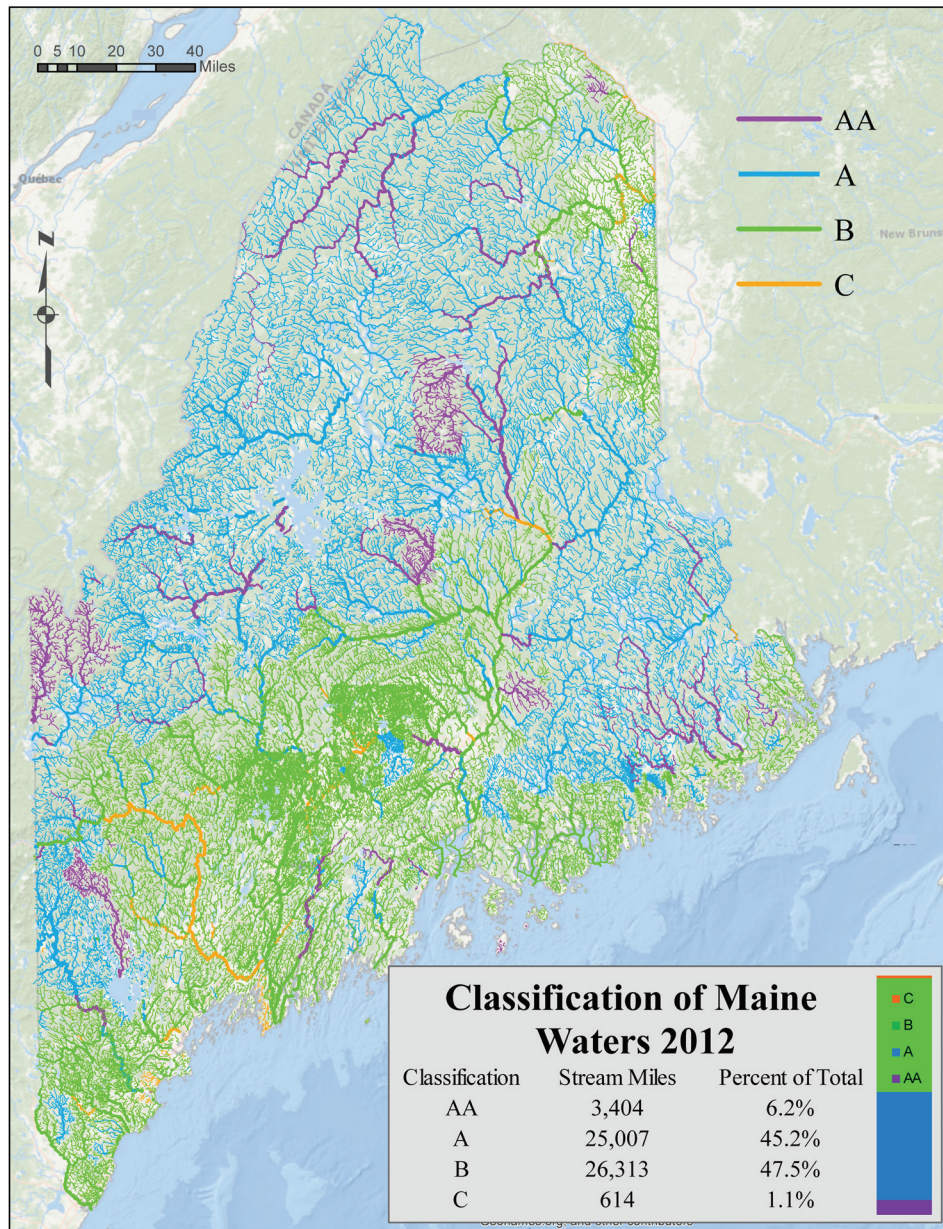


Figure 9. 2012 distribution of water quality classifications in Maine, 25 years after implementation of biologically refined water quality standards. Colored histogram shows current percentage distribution of the four classes.

Courtemanch, D.L., and S.P. Davies. 1988. Implementation of biological standards and criteria in Maine's Water Classification Law. *Proceedings of Instream Biomonitoring and Biological Criteria Workshop*. December 2-4, 1987, USEPA, Lincolnwood, IL.

Courtemanch, D.L., S.P. Davies, and E.B. Lavery. 1989. Incorporation of biological information in water quality planning. *Environmental Management* 13:35-41.

U.S. Environmental Protection Agency (USEPA). 1988.

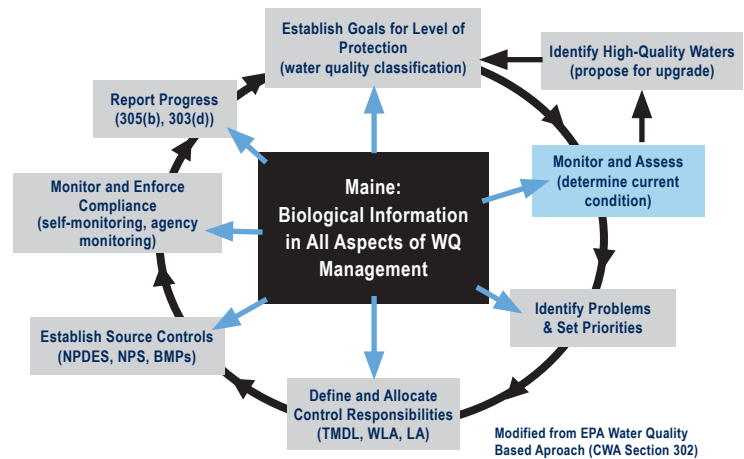
Water Quality Program highlights: Maine's biologically

based water quality standards. USEPA, Office of Water Regulation and Standards, Washington, DC.

U.S. Environmental Protection Agency (USEPA). 2011. A primer on using biological assessments to support water quality management. Office of Water, Washington, DC. EPA 810-R-11-01. <https://www.epa.gov/wqc/biological-assessment-case-studies>

## Case Study 2. Monitoring and Assessment to Determine Existing Condition Relative to Goal Condition

Together, monitoring and assessment provide the information to achieve improved biological condition of streams and rivers in Maine. Monitoring consists of regular collections of field data, using a temporally and spatially strategic sampling design. Assessment is the activity of comparing the observed biological condition against the goal condition set for the waterbody by the state legislature, as defined in Maine's water quality standards. Standardized monitoring, and clearly defined assessment protocols, applied over many years, provides important planning information to promote continual environmental and program improvement.



### Long-term point-source monitoring to document attainment of biological criteria on the Penobscot River, Maine

Data collected by the University of Maine, MDEP, and the Penobscot Indian Nation, over a period of nearly 40 years has documented dramatic improvements in water quality in a segment of the Penobscot River affected by discharges from pulp and paper mills and domestic sewage. This river segment in 1974 was in poor condition and failed to attain minimum biological standards of a Class C river. Due to restoration activities, water quality improved to attain Class B biological criteria for most locations by the mid-1980s. Rabeni and Gibbs (1977) first collected benthic invertebrate datasets in 1974, prior to the implementation of secondary wastewater treatment required by the CWA in 1972. Between 1974 and 1981 an estimated 33 million dollars was spent by pulp and paper mills and publicly owned sewage treatment facilities between the towns of Millinocket and Costigan to meet CWA mandates. This effort resulted in an 80% reduction in pollution loads (Davies 1987). Continued monitoring of benthic invertebrates and water quality was conducted in 1981 to 1982, and the results demonstrated dramatic improvements in the biological condition of sites downstream of paper mills (Rabeni et al. 1985; Davies 1987). Because both Rabeni and the MDEP used the same sampling protocols, the biocriteria model could objectively document incremental progress prior to and after treatment, document attainment of

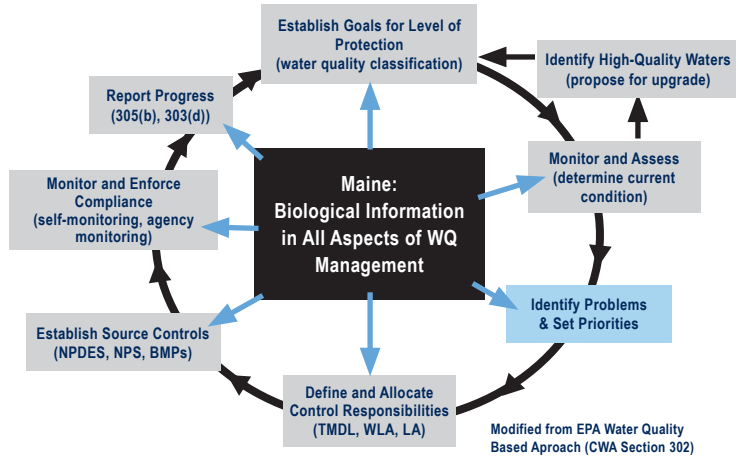
Class C, and finally, at many sites, attainment of Class B criteria. At present the entire main stem of the Penobscot River downstream of the Mattawamkeag River has been upgraded by the Maine Legislature to maintain the Class B water quality standards that were achieved (Davies et al. 1999; MDEP 2012). Documentation of progress towards increasingly higher water quality, and subsequent protection of the improved conditions would not be possible without multiple, increasingly protective aquatic life use classes in water quality standards. This long-term data set provides a valuable example of the responsiveness of biota to improvements in water quality. It also highlights the value of biological monitoring to document the benefits of investment in responsible stewardship of aquatic resources.

### For further information:

- Davies, S.P., L. Tsomides, J. DiFranco, and D.L. Courtemanch. 1999. Case study 4. Biomonitoring retrospective: Fifteen year summary for Maine rivers and streams. Maine Department of Environmental Protection, Augusta. DEPLW1999-26. <http://www.maine.gov/dep/water/monitoring/biomonitoring/biorep2000.htm>
- U.S. Environmental Protection Agency (USEPA). 2005. Case example 6-4. Use of biological information to better define designated aquatic life uses in state and tribal water quality standards. Office of Water, Washington, DC. EPA-822-R-05-001.

### Case Study 3. Identify Problems and Set Management Priorities

Patterns of biological response provide crucial evidence to allow detection and diagnosis of water quality problems and to determine their severity and thus their priority for resolution. Formalized protocols for diagnosis of causes of biological impairment have been developed by the USEPA (e.g., Stressor Identification Guidance), but simply following good routine biological monitoring and assessment practices can detect impairments that would otherwise go unrecognized.



### Detection and management of an emerging problem—Urbanization and nonpoint source impacts

When Maine’s Biomonitoring Program was initiated, a primary concern was management of point-source discharges. Implementation of best available technology (BAT) eliminated many of these causes of biological impairment. More recently, biological assessment of smaller streams revealed previously undetected impairment in urban areas caused by changes in physical stream conditions (e.g., increased impervious surfaces

in the watershed, alteration of hydrologic conditions and stream channel shape). Chemical conditions are likewise affected by increased development, human activities, and impervious cover (e.g., increased nutrients and toxic constituent concentrations, salt runoff, increased temperature, and decreased dissolved oxygen). Figure 10 shows predicted attainment of aquatic life use class (as determined by the Maine aquatic life biocriteria) in relation to a generalized stressor gradient of percentage of impervious surface in the upstream watershed. This

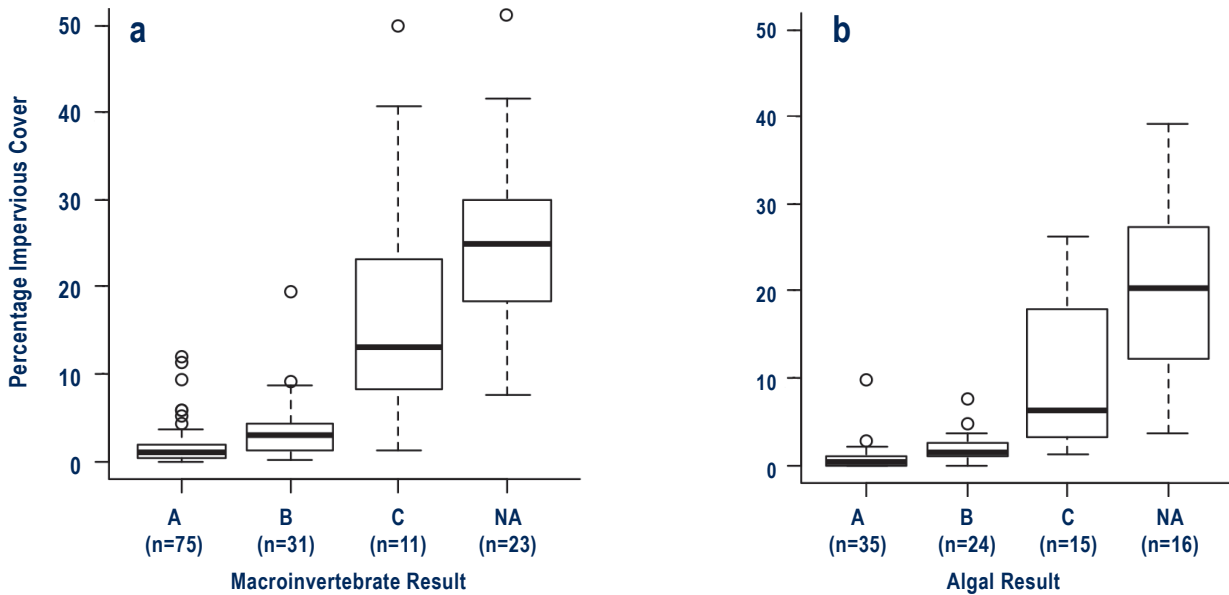


Figure 10. Predictions of the Maine linear discriminant model (bioassessment result) to increasing percentage of impervious surfaces (%IC) in an upstream watershed. Stream reaches rarely attain Class A or Class B biological standards if percentage of impervious cover was greater than 4%, n=140 for macroinvertebrate result (a), n=90 for algae result (b).

figure confirms the reliability of the statistical model to detect negative biological responses to increases in anthropogenic disturbance. The macroinvertebrate data indicate that it is unlikely under existing management practices for sites with greater than 2% to 4% impervious surfaces in the upstream watershed to attain Class A or Class AA aquatic life numeric biocriteria. Algal results demonstrate even greater sensitivity (1%–2%). Such information plays an important role in statewide planning related to the use designation process by helping inform the agency, urban planners, and public about setting realistic water quality attainment goals for streams in urbanizing areas, determining future allowable development, and understanding the expectations of urban stream restoration.

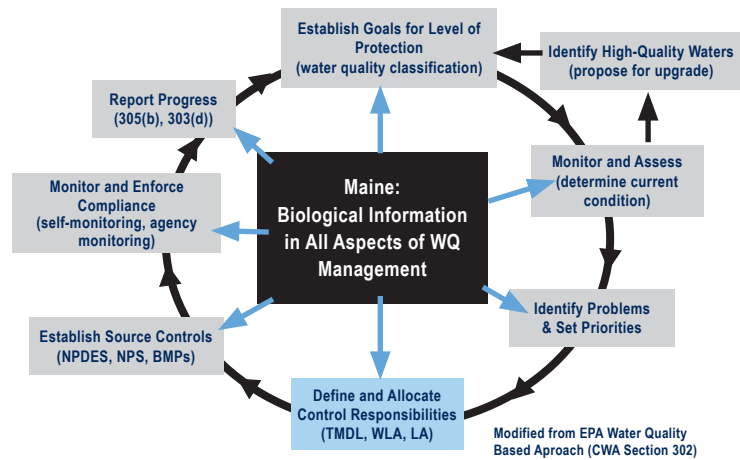
### **For further information:**

Danielson, T.J., L. Tsomides, D. Sutor, J.L DiFranco, and B. Connors. 2016. Effects of urbanization on aquatic life of Maine streams. MDEP, Augusta.



## Case Study 4. Define and Allocate Control Responsibilities

Developing the ability to recognize patterns of biological response to disturbance can help a management agency diagnose stressors and identify causes of impairment. Certain species of macroinvertebrates and algae are sensitive to environmental stressors, while others are tolerant. The lack of sensitive organisms, combined with the predominance of tolerant organisms, provides biological signatures of stressor and response. Biologists can use biological data from multiple community assemblages and trophic levels (e.g., macroinvertebrates, fish, algae) and multiple locations on a river or between rivers to identify patterns and sources of impairment and provide estimates of stressor reduction that will be required to achieve attainment standards. The CWA requires that a Total Maximum Daily Load (TMDL) analysis identifying necessary decreases in pollutants must be provided for waters not attaining water quality standards.



### Alternative TMDL to manage impacts of high impervious cover on aquatic life

In 2006, Maine and Connecticut became the first states to issue TMDL based on the percentage of stream watersheds consisting of impervious cover (IC) (Meidel and MDEP 2006a, 2006b). In 2012, MDEP completed a statewide percentage IC TMDL for urban impaired streams (MDEP 2012). The 2012 TMDL included restoration targets based on the relationship of percentage IC in stream watersheds and macroinvertebrate community condition. In 2015 MDEP revisited the concern with IC by conducting a fine-scale geo-spatial analysis of percentage IC in watersheds upstream of algal and macroinvertebrate biological assessment sites and attainment of tiered aquatic uses for each assemblage at those sites (Danielson et al. 2016). Watershed percentage IC estimates were computed in ArcMap with 1 m, high-resolution spatial data from 2004 and 2007. MDEP found that watersheds with >4%<sup>3</sup> IC were unlikely to support Class A macroinver-

3 MDEP previously developed IC TMDL targets for some urban streams. The IC ranges in the 2015 study are lower than the IC targets in the TMDLs because of more robust analysis and transition from IC spatial data with 5 m resolution to spatial data with 1 m resolution. The 5 m data overestimated the percentage IC in watersheds with more development when compared to the newer 1 m data, resulting in higher IC targets. It is not necessary to revise the TMDLs because the measurement of TMDL success is restoring water quality and aquatic life communities, not reaching a specific IC target.

tebrate communities. Similarly, watersheds with >9% and >17% IC were unlikely to support Class B and C macroinvertebrate communities (see Case Study 3). The biological assessment information was critical in establishing the restoration goals in the IC TMDL. The MDEP relies primarily on biological monitoring data and the assessment results to determine if urban impaired streams have been restored because restoration success using mitigation and management techniques is based on biological response and attainment of biological criteria and other water quality criteria, not on re-engineering impervious cover or reaching a particular percentage impervious cover target.

### For further information:

- Danielson, T.J., L. Tsomides, D. Sutor, J.L DiFranco, and B. Connors. 2016. Effects of urbanization on aquatic life of Maine streams. Maine Department of Environmental Protection, Augusta.
- Maine Department of Environmental Protection (MDEP). 2012. Maine impervious cover total maximum daily load assessment (TMDL) for impaired streams. MDEP, Augusta. DEPLW-1239 [http://www.maine.gov/dep/water/monitoring/tmdl/2012/IC%20TMDL\\_Sept\\_2012.pdf](http://www.maine.gov/dep/water/monitoring/tmdl/2012/IC%20TMDL_Sept_2012.pdf)
- Meidel, S., and M. Evers. 2007. Birch Stream total maximum daily load (TMDL). MDEP, Augusta. DEPLW0715. [http://www.maine.gov/dep/water/monitoring/tmdl/2007/birch\\_stream\\_rep.pdf](http://www.maine.gov/dep/water/monitoring/tmdl/2007/birch_stream_rep.pdf)
- Meidel, S., and Maine Department of Environmental Protection (MDEP). 2006a. Barberry Creek total

maximum daily load (TMDL). MDEP, Augusta.  
DEPLWo712. [http://www.maine.gov/dep/water/  
monitoring/tmdl/2007/barberry\\_ck\\_rep.pdf](http://www.maine.gov/dep/water/monitoring/tmdl/2007/barberry_ck_rep.pdf)

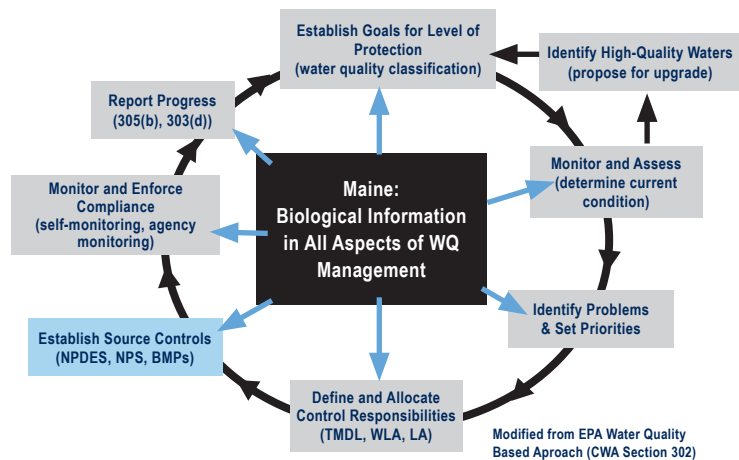
———. 2006b. Trout Brook total maximum daily load  
(TMDL). MDEP, Augusta. DEPLWo714. [http://www.  
maine.gov  
/dep/water/monitoring/tmdl/2007/trout\\_brook\\_rep.pdf](http://www.maine.gov/dep/water/monitoring/tmdl/2007/trout_brook_rep.pdf)

## Case Study 5. Establish Source Controls for Point and Nonpoint Sources

Once responsible parties are accurately identified, MDEP permitting and TMDL programs must implement regulatory provisions to reduce or eliminate the causes of the biological impairment. Biological information has been an essential element in many TMDL designs to determine appropriate levels of load reductions that can be expected to result in attainment of biological criteria.

### Using biological information to set permit limits in the absence of ambient chemical criteria

Assessments of aquatic life below a paper mill on the Androscoggin River (Class C) occurred in 1995, 1996, 2000, 2002, and 2003 to determine the condition of the river, probable causes of impairment, and to set new limits on waste discharges. Class C aquatic life standards were not attained downstream of the discharge in 1995, a low-flow year, and were presumed to be caused by a heavy load of total suspended solids (TSS). The estimated TSS load (concentration times river flow) in 1995 was 18,051 lbs/day as a monthly average. The water above the discharge in 1995 met the Class C aquatic life standard. The estimated TSS load prorated to the flow above the discharge 1995 was 6,976 lbs/day (there is another paper mill upriver). In 1996 increased dilution of the discharge was gained from a wetter than normal summer, and the mill also experimented with polymer addition that resulted in a reduction of TSS discharge to a flow-prorated estimate of 3,706 lbs/day. Biological monitoring data collected in the summer of 1996 revealed a very positive response in the benthic macroinvertebrate community resulting in the attainment of aquatic life standards throughout the study area. As compared to 1995, the total abundance of organisms, generic richness, proportion of insect taxa, and EPT (Ephemeroptera, Plecoptera, Trichoptera) taxa increased dramatically, notably filter-feeding Trichoptera (caddisflies). Further confirmation of solids as the probable cause for nonattainment the previous year was made by scuba-diver observations of the accumulation of a flocculent deposit of solids on the sampler substrate during a low-flow/high-solids load year (1995) in the Androscoggin River impoundments.



Solids did not accumulate in 1996 when discharge of TSS was reduced.

In 2000, 2002, and 2003, the benthic macroinvertebrate communities in the impoundments were resampled to determine aquatic life attainment. In 2000 the downstream flow-prorated TSS load was 6,698 lbs/day per day, which was similar to the level of TSS in the upstream in 1995. All sites met at least the Class C aquatic life standard with some attaining the Class B aquatic life standard. The downstream site showed dramatic improvement from 1995. Generic richness increased from 11 to 40. EPT richness increased by three times, and mayfly abundance increased from 0 to 67 per sample. In 2002 the flow-prorated TSS load was 7,806 lbs/day. All sites below the discharge met the Class C aquatic life standard except the downstream-most site. The downstream site exhibited a decrease in generic richness, EPT taxa, and mayfly abundance from the 2000 sample. In 2003 the flow-prorated TSS discharge was 7,915 lbs/day. All samples met at least the Class C aquatic life standard. In general, generic richness, EPT taxa, and mayfly abundance increased as compared to the 2002 samples. The improvement of the benthic communities in the impoundments below the discharge in 2003 may be due to several high-flow flushing events that occurred during the sampling period. In the year 2000, when the biological community closely resembled the 2003 data, there was also a high-flow event during mid-July with steady flows most of the summer. Maine does not have ambient water quality criteria for TSS. The data suggested that a TSS level of <8,000 lbs/day would be adequate for attainment of aquatic life standards during average rainfall years. New TSS limits

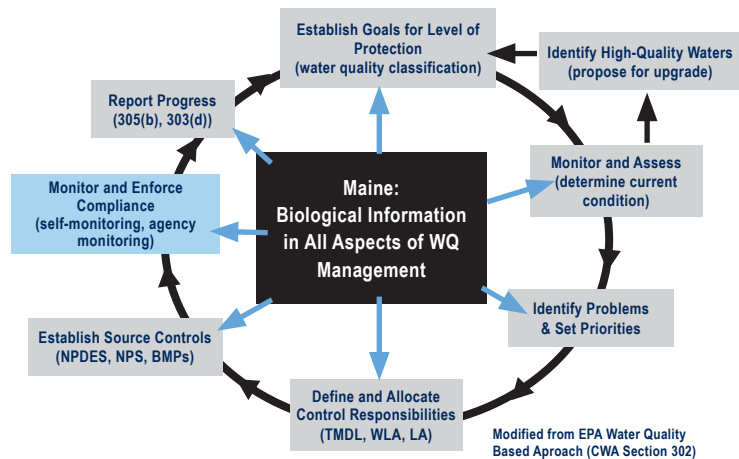
and a load allocation for the two mills were established through a TMDL process based on a 30 day/10 year low flow. The river has since attained Class C and has been removed from the state's Section 303(d) impaired waters list for aquatic life. A similar analysis was used on the Presumpscot River to establish TSS permit limits (Davies et al. 1999; USEPA 2000).

### **For further information:**

- Davies, S.P., L. Tsomides, J. DiFranco, and D.L. Courtemanch.  
1999. Biomonitoring retrospective: Fifteen year summary for Maine rivers and streams. MDEP, Augusta. DEPLW1999-26. <http://www.maine.gov/dep/water/monitoring/biomonitoring/biorep2000.htm>
- U.S. Environmental Protection Agency (USEPA). 2000. Chapter 6. Stressor Identification Guidance Document. Office of Water, USEPA, Washington, DC. EPA/822/B-00/025.200.

## Case Study 6. Monitor and Enforce Compliance through Self-monitoring or Agency Monitoring

Numeric biological criteria in water quality standards provide a powerful action-forcing mechanism to achieve environmental improvement. Detection of a violation of biological criteria sets in motion regulatory remedies to stop the detrimental activity and restore the waterbody. In some cases, responsible parties may be required to conduct monitoring to confirm compliance. In other cases, MDEP monitors sites to amass sufficient information to implement the regulatory requirements needed to achieve restoration. Occasionally, MDEP enforcement actions have been necessary to ensure compliance with water quality standards. Results from biological assessments help ensure that remediation and restoration plans are fair and properly implemented and provide a final proof of attainment.



### Use of biological criteria for enforcement actions

In 2009 MDEP biologists testified in court for an enforcement case against a large composting facility in the headwaters of the Kennebec River watershed in Lyman, Maine. Due to poor storm-water management, large quantities of organic waste intermittently washed into Lords Brook during storm events. While the events were difficult to detect and document, the biological effects were evident: sewage fungus was abundant in the stream, water quality was poor, and the macroinvertebrate community was severely degraded. Lords Brook has a statutory water quality goal of Class B, but it did not attain biological criteria for either Classes B or C. In comparison, nearby East Outlet Stream was not affected by the organic waste and attained Class A biological criteria. The MDEP prosecuted the case based on the nonattainment of the state's biological standards. Biologists provided testimony about the impact to water quality and aquatic life based on monitoring and assessment using the Maine linear discriminant biocriteria models, linking the activities at the composting facility to the effects on the biota in the brook. The court agreed with the MDEP's enforcement case because of the documented, detrimental impacts to water quality and aquatic life in Lords Brook (State of Maine, Department of Environmental Protection v. Winterwood Acres, Inc, Winterwood Farms, LLC

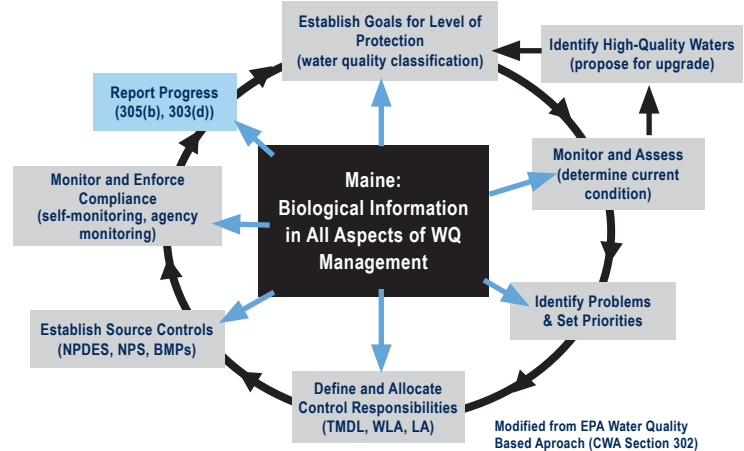
[Maine Tenth District Court, Docket # CV-06-228]). This case was further upheld when appealed to the Maine Law Court.

### For further information:

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## Case Study 7. Report Progress

*An agency's ability to report monitoring and assessment results to decision makers and the public is a key step in improving and maintaining water quality. Public awareness and understanding of the condition of aquatic resources is essential to well-informed and proactive public choices in stewardship of valued aquatic life assets. In addition to regulatory requirements to report results, there are long-term benefits to making bioassessment results readily available to the public.*



### Communicate bioassessment results through interactive maps and traditional reports

Perhaps the most effective way that the MDEP Biological Monitoring Program communicates its monitoring and assessment results to the public is through its Google Earth website (<http://www.maine.gov/dep/gis/datamaps/index.html#blwq>). The website provides an interactive map where one can view sampling locations, the state's waters with their statutory goals, and biological assessment outcomes (Figure 11; Appendix 8). Various spatial layers can be turned on or off to enhance navigation and interpretation of watershed features, such as roads, town lines, and aerial imagery of the landscape. Users can select sample stations to view their monitoring history including summaries of biomonitoring results, taxonomic data, model outcomes (probabilities of attainment for each class) and final determination of attainment status as determined by biocriteria. Images of the stream or wetland sample locations are also provided in most cases. Users also can download biological and water quality data and summary reports for individual sample events (Appendix 8).

The Biological Monitoring Program also provides biological assessment results in state and federally required assessment reports. Sections 305(b) and 303(d) of the CWA require state water quality agencies to report on the condition of state waters. MDEP produces an Integrated Water Quality Monitoring and Assessment Report every two years that summarizes attainment of water quality standards. MDEP follows a public process to solicit and respond to comments and the reports are publicly available on the MDEP website.

In the integrated report, waters are placed in one of the following categories:

- Category 1—Attaining all designated uses and water quality standards, and no use is threatened.
- Category 2—Attains some of the designated uses; no use is threatened; and insufficient data or no data and information is available to determine if the remaining uses are attained or threatened (with presumption that all uses are attained).
- Category 3—Insufficient or conflicting data and information to determine if designated uses are attained (with presumption that one or more uses may be impaired).
- Category 4—Impaired or threatened for one or more designated uses, where a TMDL has already been prepared or does not require development of a TMDL.
- Category 5—Waters impaired or threatened for one or more designated uses by a pollutant(s) and a TMDL is required.

In addition to the federal reporting requirements, the Maine Legislature requires that the monitoring program produces annual reports of results of the Maine's Surface Water Ambient Toxics (SWAT). The SWAT monitoring program includes assessment for attainment of biocriteria for stream macroinvertebrate samples funded under the program. The Biological Monitoring Program also reports chemical and continuous temperature data associated with the biomonitoring samples.

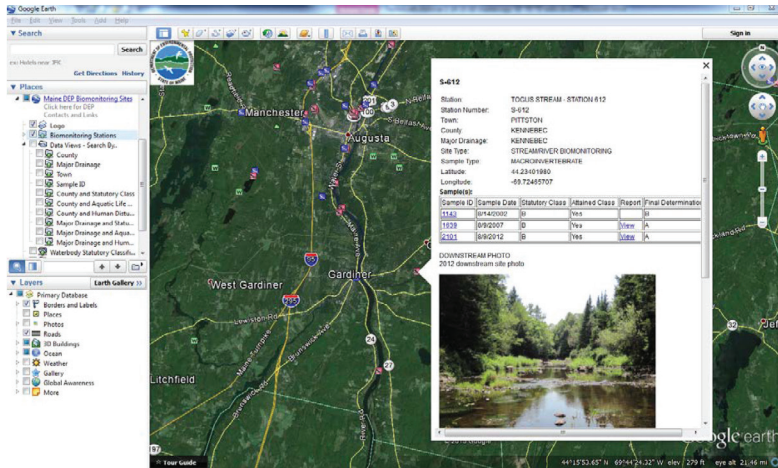


Figure 11. Screen shot of the Biological Monitoring Program's Google Earth website.

### For further information:

Biological Monitoring Program Google Earth website:

<http://www.maine.gov/dep/water/monitoring/biomonitoring/data.htm>

Integrated Water Quality Monitoring and Assessment reports:

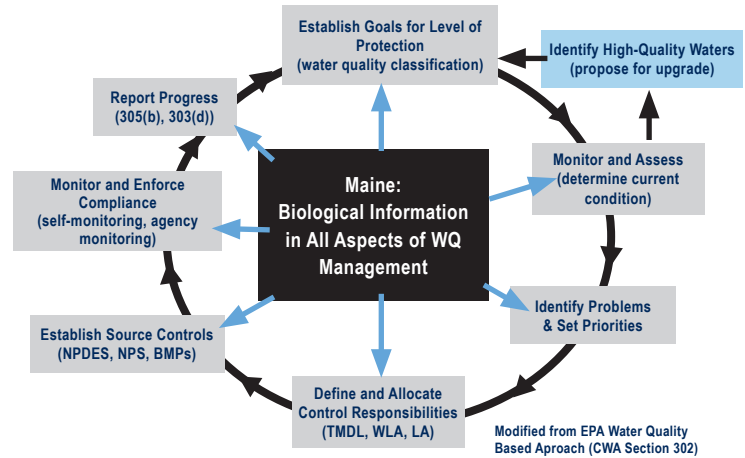
<http://www.maine.gov/dep/water/monitoring/305b/index.htm>

Surface Water Ambient Toxics program reports:

<http://www.maine.gov/dep/water/monitoring/toxics/index.html>

## Case Study 8. Retention of Water Quality Improvements

*In addition to simply identifying waters that do not attain their standards, state waters can be objectively assessed along a gradient of biological condition to determine if restoration is needed, if they attain standards and current management is successful, or if they exceed standards and additional protection strategies are desired to maintain that higher quality. Maine’s water quality classification law encourages agency, advocacy groups, and citizen proposals for water quality classification upgrades as waterbodies improve to meet the standards of the next higher classification (Davies et al. 1999).*



The law requires the state to propose classification upgrades:

When the actual quality of any classified water exceeds the minimum standards of the next highest classification, that higher water quality must be maintained and protected. The board shall recommend to the Legislature that that water be reclassified in the next higher classification. 38 MRSA §464.4.F(4)

This is an important benefit of having a tier-based approach to water quality management. It allows for

formal recognition of incremental improvements, a feature that cannot be achieved via the simple single-threshold approach used by many states and allowed by the USEPA. The upper right-hand box of the “water wheel”(Figure 7 “Identify high-quality waters”) depicts this important modification to the USEPA’s water-quality-based approach. When high-quality waters are recognized through monitoring and are valued and championed by the public, new goals and standards of a higher classification tier can be assigned by the legislature. This protects not only the quality of the water, but the public or private investment that has been made

toward improved water quality. Maine law and the USEPA Triennial Review require an assessment of classifications every three years when such upgrades are proposed (38 MRSA §464.3 and 38 MRSA §464.3.B). Though not all such waters are ultimately upgraded by the legislature, the state’s antidegradation policy (38 MRSA §464.4.F) also

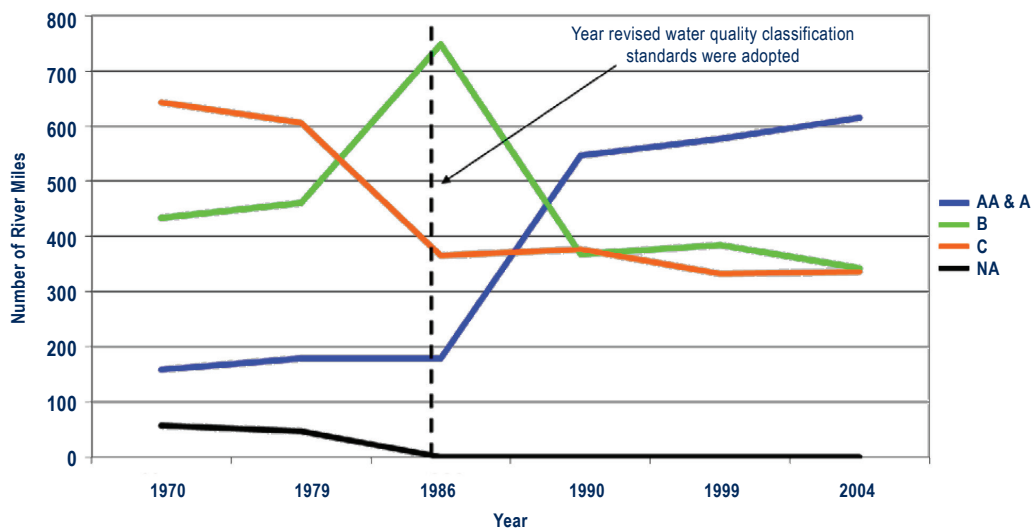


Figure 12. Increase in the number of miles of mainstem river reaches designated to maintain Class AA and Class A conditions in Maine since the adoption of tiered aquatic life use classes (1970 to 2004).



functions to protect the higher water quality criteria that have been attained.

Since 1987, over 1,440 mainstem river miles in Maine have been upgraded to a higher water quality management classification using biological assessment as a principle determinant (Figure 12; <http://www.maine.gov/dep/water/wqs/docket/index.html>; <http://www.epa.gov/wqs-tech/water-quality-standards-regulations-maine>). Over the last 25 years, the state has also upgraded 13,955 stream miles from Classes A, B, or C to a higher classification (Figures 8 and 9; see also Case Study 1). Currently 51% of all river and stream miles in Maine are assigned to Class A or Class AA (Class AA also affords CWA Tier 3 antidegradation protection as “Outstanding National Resource Waters,” 38 M RSA §464.4.F[2]), with an additional 45% managed to maintain Class B conditions (affording antidegradation protections that are more stringent than CWA Tier 2, MDEP [2012]) (Table 8). During the same time period, only five miles have been downgraded (from Class B to Class C) through application of a use attainability analysis to document that a downgrade was required to meet important socioeconomic needs (<http://www.maine.gov/dep/water/wqs/docket/index.html>; <http://www.epa.gov/wqs-tech/water-quality-standards-regulations-maine>). Without refined aquatic life management categories and numeric biological criteria, it is difficult to recognize and protect these water resource improvements. The communication value of biological goals enhances public understanding of existing conditions and restorable target conditions and provides an important tool in building public support for the often substantial investment that is required to restore aquatic resources (Davies and Jackson 2006).

## Part 4: SUMMARY, DISCUSSION, AND CONCLUSIONS

### 4.1 SUMMARY

#### 4.1.1 Policy summary

On April 17, 2003, the Maine Board of Environmental Protection adopted numeric biological criteria for rivers and streams in rule, which were subsequently approved by the Maine Legislature. This publication describes the policy foundations and the statistical methods that culminated in passage of the rule. The 2003 rule was promulgated in support of Maine's 1986 law that established and defined four aquatic life use classifications for rivers and streams (AA, A, B, and C) spanning a water quality gradient that ranges from Maine's interpretation of the CWA Interim Goal (Class C) to the ultimate CWA objective "to restore and maintain chemical, physical and biological integrity" (Classes AA and A). The protocols described in rule include methods for biological sampling of benthic macroinvertebrates, laboratory analyses, statistical modeling, analysis of data, and selective use of expert judgment to reach a final determination of classification attainment.

Passage of Maine's biocriteria rule has fostered innovative and far-reaching applications of biological information that have helped optimize the biological condition of Maine's rivers and streams. MDEP's ability to detect, define, and remedy previously undetectable water quality problems was expanded and strengthened by implementation of these biological criteria. Equally important is management at the other end of the water quality continuum. Maine is fortunate to retain extensive areas of intact forest with relatively low human impact and near-natural stream quality. Statewide biological monitoring and assessment, using the criteria described in this publication, has helped MDEP to document, and the Maine public to recognize, that specific rivers and streams around the state currently support very high-quality aquatic life. In combination with Maine's tiered classes in water quality standards, bioassessment information provides a mechanism for the public, and ultimately the Maine Legislature, to decide on preservation of these unique resources by upgrading assigned goals to maintain high-quality Class A standards.

#### 4.1.2 Technical summary

To accomplish our goal to fully incorporate biological information in regulation and management of water quality, we developed and tested a statistical

bioassessment system (Figure 2, Section 2.3.3), designed to predict the probability of membership of a test sample within any of four statutorily defined water quality classes. Model predictions are derived from analysis of a total set of 25 different taxonomic and invertebrate assemblage variables. MDEP used a Delphi approach incorporating biologists' prior experience (expert judgment) to define the a priori classes required for construction of linear discriminant models (Bakus et al. 1982). Biologists' prior experience considered empirically observed changes in attributes of Maine macroinvertebrate assemblages in response to gradients of human disturbance (Davies et al. 1995; Davies and Jackson 2006). Biologists referred to narrative descriptions of condition goals in Maine's narrative aquatic life criteria and their statutory definitions to assign samples to a priori classes. A technical advisory committee of nondepartmental participants regularly reviewed developmental progress in constructing the statistical bioassessment system (Table 3). As described in Section 2.3.3 and Appendix 2, the effort to construct the model resulted in an innovation that uses a nested set of linear discriminant functions to improve predictive success (Figure 2). The first-stage of analysis, termed the four-way discriminant model, uses nine variables to separate samples into one of four groups. Further refinement of predictions is accomplished by a second-stage analysis using 16 additional variables in three, two-way models having a predictive success ranging from 89% to 97% using a jackknife procedure (Section 2.4 and Appendix 2, Table A16). In no case did the combined model incorrectly classify a sample by more than one class. This high concordance and low error makes the models statistically strong and provides a consistent and objective analysis of the aquatic life standards, which reduces conflict over interpretation of the law. A second innovation is that boundaries denoting classification attainment are assigned in the rule as probability statements. Traditional water quality criteria are typically set as fixed threshold values; however, these biological criteria used to determine classification attainment are established as a best-fit using multiple variables. In summary, these statistical models, codified in the MDEP biocriteria rule, provide the objective, quantitative means for determining attainment of designated aquatic life use classes in Maine water quality standards. Site-specific results of the bioassessment system, as well as all pertinent facts concerning the sampling and analysis process (e.g.,

excursions from standardized sampling parameters for habitat, loss or disturbance of replicates, sub-sampling protocol) are reviewed by biological staff in the final stage of decision making. The final result is an objective, step-wise decision-making protocol that is based on statistical strength of membership within classes.

#### 4.1.3 Reception of the predictive model

Initial concerns about numeric biocriteria from regulated industrial and business interests stemmed from the untried nature of the use of biological information in water resource management, from the complexity of the quantitative approach used to determine attainment, and from the use of a probability of attainment approach. Each of these concerns was addressed during promulgation and implementation of the biocriteria rule. The response of aquatic life to wastewater treatment was quickly observed as rivers came back to life following implementation of CWA primary and secondary treatment requirements in the 1970s and 1980s (Rabeni et al. 1985; Davies 1987). This was a tangible return on investment that could be witnessed by all parties. The complexity of quantifying the aquatic life response was further addressed when the results were shown to be consistent, well-correlated with objective measures of water quality, and highly reproducible. The use of probabilities was also accepted as a more candid representation of a water quality sample and provided a means where all parties could see the strength of any water quality attainment decision.

Regulated entities are most familiar with permit performance standards such as waste-load allocations and discharge limits for specific pollutants, or with physical/chemical ambient water quality criteria. But both permit performance standards and physical-chemical water quality criteria have many shortcomings that limit their application and usefulness to assess ecological status (Courtemanch et al. 1989; Courtemanch 1993). In contrast, monitoring of aquatic life provides for the direct assessment of ecological impacts caused by pollutants and habitat alteration. The MDEP has used the biological field and analytical methods described in this publication since about 1990, establishing a level of familiarity within the regulated public with the use of these environmental response standards (see Case Studies). Over that time, a substantial body of regulatory decisions have been based on these methods (Davies et al. 1999; Barbour et al. 2000; USEPA 2011).

The eight case examples provided in Part 3 illustrate the range of successful and innovative water quality

management applications that resulted from Maine's biologically based approach. Biological assessment findings resulting from these methods withstood an aggressive legal challenge described in Case Study 6. Decisions based on biocriteria results have driven significant changes in management actions taken by the MDEP (Case Study 1, 3, 4, and 5) and in discharge limits and operations of some regulated parties (Case Study 2 and 7). Finally, the advantages of a tier-based approach to water quality management and protection are shown in Maine's revision of USEPA's water-quality-based approach to recognize and lock in incremental improvements in water quality (Case Study 8).

## 4.2 TRANSPARENT COMMUNICATION OF BIOLOGICAL CONDITION

### 4.2.1 Gradient models

Gradient models can help nonscientists to visualize and interpret the relative implications of complex ecological data. Gradient models of four to six condition tiers are established as water quality law in the European Union, and in a few U.S. states including Maine. They have been shown to represent a reasonable number of management choices (European Commission 2000; USEPA 2011, 2016). These classes encompass a range of conditions from the minimum required to maintain a well-functioning aquatic community to high-quality conditions with little human perturbation, offering high conservation value, and serving as a reference standard against which the other condition tiers can be compared. As law, such aquatic life standards are distinguished by their ecologically detailed descriptions of the class boundary conditions required to meet water quality goals (European Commission 2000, 2010; Ohio Water Quality Standards, Chapter 3745-1, Ohio Administrative Code; Maine Water Quality Standards, MRSA Title 38 Article 4-A §464-466). The narrative aquatic life criteria in Maine's water classification law describe conditions across such a biological gradient and are supported by ecologically based definitions in the law (Table 2).

### 4.2.2 Other transparency models—The EU Water Framework Directive and the biological condition gradient

Some policy and technical parallels exist between Maine's approach and that of the European Union's high, good, and moderate ecological status objectives described in the European Union Water Framework

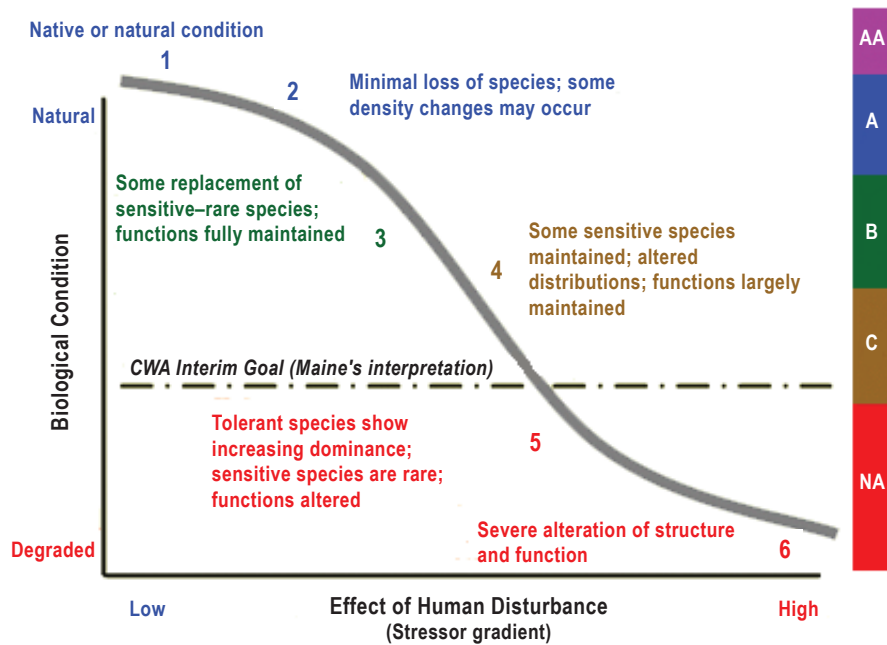


Figure 13. Goal condition of Maine river and stream management classes relative to the biological condition gradient.

Directive (WFD) (European Commission 2000; Reitberger et al. 2010; Poikane et al. 2014). The WFD states the objective for waters of the European Union to achieve at least good ecological status by 2015. In addition to high, good, and moderate ecological status objectives, the WFD further characterizes poor ecological status categories, poor and bad. These condition categories are not defined in the WFD, and they are deemed to not attain the ecological status objectives of the WFD, but characterizing them enables documentation of incremental improvement of waters into higher attainment categories. The Maine biocriteria model was specifically designed to quantitatively assess attainment of standards in the water quality classification law. Maine's higher classifications (Classes AA, A, and B) introduce aquatic life criteria more stringent than the minimum fishable-swimmable CWA Interim Goal, while Maine deems that Class C is equivalent to at least the minimum Interim Goal condition. The statistical model described in Part 2 and appendices predicts site membership in groups having characteristics that correspond to the three statutory classification standards (Class A, B, and C) and associated definitions for biological assemblages. Maine's statistical models also predict nonattainment (NA) sites that do not attain any of the statutory goal classes, (i.e., worse than the lowest allowed Class C condition). Determination of

NA in Maine's biocriteria can be interpreted as relatively equivalent to the status and purpose of poor and bad status in the WFD (Reitberger et al. 2010).

The ecological characteristics of each of Maine's classifications can also be accurately and transparently communicated to other scientists, the public, and policymakers via their correspondence to condition levels described in the national biological condition gradient (BCG) (Figure 13; Davies and Jackson 2006; USEPA 2005, 2011, 2016). The BCG offers, in a simple stepped descriptive gradient, an ecological framework with the primary purpose to consistently and transparently communicate technical

findings about changes in biological condition across a human disturbance gradient. The BCG uses detailed, ecologically descriptive condition tiers to express biological responses ranging from a natural state to severe alteration, in relation to a gradient of stress (Davies and Jackson 2006; USEPA 2011, 2016).

With reference to the BCG, as shown in Figure 13, Class AA and A both correspond with BCG Tiers 1 and 2. Classes B and C most closely correspond with BCG Tiers 3 and 4, respectively. The Nonattainment condition most closely corresponds with BCG Tiers 5 and 6.

The BCG was developed by a national working group of bioassessment practitioners and scientists, sponsored by USEPA, as a crosswalk to facilitate comparison of bioassessment results among states using different assemblage groups and different field and analytical methods. The BCG initiative sought to better standardize communication about the outcomes of biological assessment, and it is based in part on Maine's tiered standards for aquatic life. The BCG model was published with an example that presents empirical data from Maine, to demonstrate the application of the ecological characteristics described in the generalized BCG model (see Digital Appendix A4 in Davies and Jackson 2006).

The general BCG model described in Davies and Jackson (2006) does not require application of the same technical rigor and standardization as Maine's

quantitative predictive models do in order to have wider application, nor does bioassessment based simply on the BCG have the legal authority and leverage of Maine's regulation. Still with local bio-monitoring data and expert judgment, the general BCG model can be used to effectively communicate aquatic life conditions across larger geographic and geopolitical scales. This feature is useful to initial stages of water quality inventory and planning at regional and national scales. The BCG is also useful for nongovernmental organizations seeking a compelling and transparent way to communicate important changes in biological condition to their water resource constituencies.

The value of the BCG model to translate biological condition among differing assessment approaches was demonstrated in a New England statewide bioassessment, using the BCG model as the common yardstick among states having differing sampling and assessment methodologies (Snook et al. 2007). Because it is a transparent and reproducible method, this approach provides a means to standardize communication about how much biological change has occurred relative to state and federal management goals for waterbodies.

### 4.3 CONCLUSIONS

Maine's experience with designing a credible and robust scientific and legal framework for regulatory and management use of aquatic biological information illustrates that the power and utility of biological criteria in water resource management increases with its increasing formalization in the regulatory setting (Figure 14).

Most states and tribes in the United States conduct biological assessments, yet only a handful have promulgated quantitative biocriteria in water quality standards (Chris Yoder pers. comm.; USEPA 2013). While reporting on aquatic life status may be a necessary task (for example to fulfill reporting requirements of the CWA §305b or §303d impaired waters list), much of the transformative potential of biological information to focus an agency's attention on optimizing environmental outcomes is lost if the effort stops there (Yoder and Barbour 2009; Courtemanch et al. 1989). For example, biological monitoring downstream of permitted municipal or industrial wastewater discharges might provide invaluable documentation of poor biological conditions attributable to inadequate permit limits, yet without legally enforceable biological criteria, it may be impossible to intervene to remedy the problem.

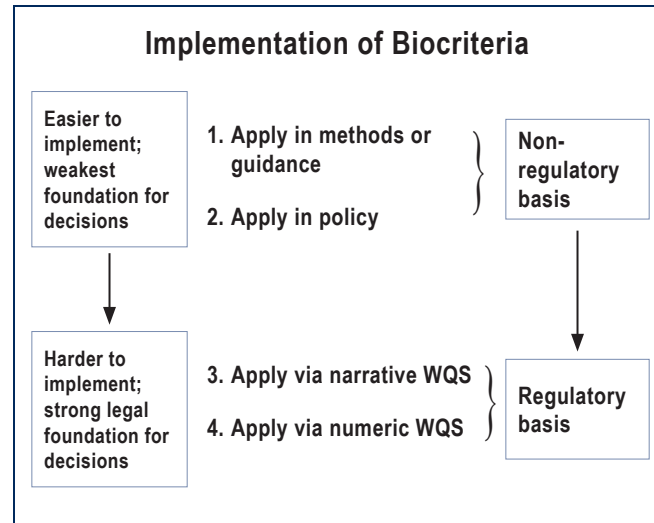


Figure 14. Relative usefulness and scope of biological criteria to enhance water resource management is contingent upon how formally it is institutionalized.

Many states employ simple pass-fail bioassessment triggers, specified in agency guidance or policy (Figure 14). These triggers are used to list waters for aquatic life use impairment (e.g., §303d), but the technical program may have little ability to detect, document, and ultimately preserve incremental improvements in condition (USEPA 2013). At the other extreme, the agency may lack any regulatory mechanism to maintain waters found to be of exceptional quality due to bioassessment action triggers that are well below current high conditions. The pass-fail management paradigm, used by so many states, deprives the public of the more precisely resolved gradient of classification options that would allow them meaningful participation in local and statewide water resource planning (Courtemanch et al. 1989; Courtemanch 1993; Davies and Jackson 2006; USEPA 2016).

Maine's biocriteria program is based on an underlying conceptual gradient of acceptable biological condition classes, codified in statute and executed in rule. The overall approach provides a range of management options to better meet the public's interest in balancing Maine's well-deserved reputation for environmental beauty and purity, with the need to accommodate important socioeconomic interests. While many state regulatory agencies rely solely on the antidegradation provisions in the CWA to prevent declines in water quality, Maine's classes provide enhanced statutory protection so that all waters are either maintained at their currently attained goal conditions, or are made to improve to meet those statutory goals. The Maine

Legislature has designated 50% of the state's river and stream miles to be managed as Class AA or Class A (equating to Tier 1 or 2 on the BCG) and thus directs these waters to be maintained in this very high condition (Figure 9; MDEP 2012). Waters that fail to attain the standards of their assigned classification have the weight of the classification law behind them to drive management intervention and remedial action. This important action-forcing mechanism applies to a Class AA or A waterbody that fails to attain the very high physical, chemical, and biological standards of those classes in just the same way that it applies to waters that have failed to attain a lower assigned class. All confirmed cases of waterbodies failing to attain their assigned classification standards are placed on the state's impaired waters list, triggering action to address the impairment. The gradient of tiered classes in law (tiered aquatic life uses), combined with the legislature's exercise of its option to designate a high percentage of river and stream miles into high-quality goal classes, helps to bend the state's overall planning and management paradigm towards prevention of problems and earlier intervention when at-risk streams are still in good condition, so problems are less obdurate, and solutions less expensive.

As bioassessment progresses in an agency from nonregulatory guidance into full implementation in water quality standards and rule, increasingly comprehensive and influential applications become available to managers that improve management responsiveness to biologically detrimental influences and ultimately result in improved environmental outcomes (Courtemanch et al. 1989; Courtemanch 1995; USEPA 2013). Weaknesses in either articulation and codification of goals and management objectives or in implementation of a sound scientific framework to support environmental policies risks failure to optimally protect and preserve valued aquatic resources. Positive results depend upon both an intelligent and scientifically informed foundation in policy and law and implementation of a technical program that is strategically designed with the goals of the law firmly in mind.

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## APPENDIX 1. FIELD, LABORATORY, AND DATABASE MANAGEMENT METHODS

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### A1.1 SELECTION OF BENTHIC MACROINVERTEBRATES FOR CRITERIA DEVELOPMENT

Maine's choice of benthic macroinvertebrates as the primary community component to be used to assess the condition of river or stream life was based upon the following tenets:

1. Benthic macroinvertebrates generally have less mobility than fish and are therefore less able to avoid exposure to pollutants (Rosenberg and Resh 1993). Fish, under the right conditions, may swim considerable distances to avoid pollutants and so may not be as reliable an indicator of local environmental conditions (Little 2002). Some fish species also make extensive seasonal or life-stage migrations. Therefore, presence or absence of fish may be unrelated to water quality condition.
2. Within the macroinvertebrate group there is a wide range of pollution tolerance (Hilsenhoff 1987; Merritt and Cummins 2008). Some sensitive species may be killed or excluded by very low levels of pollutants (species of Ephemeroptera and Trichoptera), while other taxa may actually thrive in large numbers only in the presence of certain types of pollution (e.g., some species of Syrphidae and Chironomidae [Diptera], annelids, and gastropods).
3. Benthic macroinvertebrates are an extremely diverse group (Thorp and Covich 1991; Merritt and Cummins 2008), having a greater richness of taxa, diversity of life history strategies, and variety of feeding and energy use strategies. By comparison, Maine's fish communities are relatively low in taxonomic and functional diversity (Halliwell et al. 1998). Assessment of the benthic macroinvertebrate community provides a great deal of information regarding energy use, tolerance to pollution, and functional well-being of the entire aquatic system.
4. Benthic macroinvertebrates have longer, more complex life cycles than algae or bacteria—frequently living one or more years in the aquatic environment—and can reflect the integration of water quality effects over time (Rosenberg and Resh 1993).
5. Many fish species that are valued state resources (particularly Salmonidae) are largely dependent on the macroinvertebrate community as a food source during at least some life stages (Hartel et al. 2002). Since the range of pollution tolerance of insects and other invertebrate organisms is broadly comparable to that of fish (Barbour et al. 1999), assessment of macroinvertebrates is an indirect method of gaining information about risks to the fishery of an area without directly assessing the fish community.
6. Some form of benthic macroinvertebrate life can be found in all but the most severely polluted or disturbed habitats, unlike fish, which may be absent due to natural causes such as obstructions to passage. In addition, fish communities are affected by fishery management and selective exploitation, which can lead to nonpollution-based declines.
7. Methods for collecting samples and analyzing results are well established (USEPA 1973; Green 1979; Klemm 1990; Stribling et al. 1996; USEPA 1999, 2005). Since macroinvertebrates are widely available and easy to capture, they are a cost-effective group to sample, though a drawback at species-level analysis is the continually changing status of taxonomy and systematics within many groups of freshwater invertebrates and the expertise needed for species-level determinations.
8. Early research in Maine waters established a base of knowledge about benthic macroinvertebrate responses to differing water quality conditions, applicable sampling methods for Maine waters, and expertise in analysis of biological information (Rabeni and Gibbs 1977; Rabeni et al. 1985; Davies 1987).

### A1.2 STREAM-SAMPLING METHODOLOGY

Benthic macroinvertebrates were collected using rock filled substrate samplers of standard construction and deployment (MDEP 1987, 2009). Three types of samplers were used depending on the depth of water:

cylindrical baskets for wadeable streams (Rabeni and Gibbs 1977; Davies 1987; Klemm et al. 1990), cones for non-wadeable water bodies (Courtemanch 1984), and mesh bags for small flowing waters that are too shallow to fully immerse a basket sampler. Each sampler contained 7.25 kg ( $\pm$  0.5 kg) of clean, bank-run cobble graded to a standardized diameter range of 3.8 cm to 7.6 cm (1.5 in. to 3.0 in. commercially available as #2 roofing stone).

At each sampling station, biologists deployed three replicate samplers based on earlier studies demonstrating that the standard error for three samplers (total community density) was within 20% of the mean (Rabeni and Gibbs 1977; Davies 1987). The standard sampling season was restricted to dates between 1 July and 30 September, with a duration of 28 days  $\pm$  4 days. For impounded waters, a longer incubation period of 56 days  $\pm$  4 days is allowed to accommodate the expected slower colonization period. Biologists used 600  $\mu$ m dip nets during retrieval to minimize loss of organisms. The samplers were then transferred to a sieve bucket with 600  $\mu$ m mesh. Biologists removed and cleaned all the rocks from the sieve buckets, leaving behind sand, detritus, and macroinvertebrates. The contents of the buckets were transferred to a jar and preserved with ethanol. Biologists processed and preserved each sampler separately. Field staff collected data on water velocity, depth of the sampler, stream width, substrate composition, canopy cover, water temperature, dissolved oxygen, and specific conductance at the time of sampler deployment and again during retrieval. Sampling was conducted such that a representative coverage of streams and geographic regions was attained (Figure A1).

### A1.3 LABORATORY PROCESSING PROCEDURES

After samples arrived at the laboratory, technicians sorted samples by hand to remove macroinvertebrates from debris. Laboratory staff preserved benthic macroinvertebrate specimens in 70% ethanol and 5% glycerin, and samples were placed in glass vials for future reference. Subsampling was performed on samples if the mean number of organisms in the three samplers exceeded 500 and subsampling would yield at least 100 organisms per sampler. When subsampling was necessary, all samples in a reach were treated consistently. For example, if one site of a paired set of upstream-downstream sites was subsampled, the other site was also subsampled in the same way (as long as the resulting sample would yield the required minimum 100 organisms). This process was done to

standardize the level of sampling effort. Subsampling was conducted using the method of Wrona et al. (1982), which is a proportional subsampling method (in contrast to fixed count subsampling), which can be factored up to standardized whole sample counts, enabling more accurate assessment of density differences between samples (Courtemanch 1996). Taxonomists identified organisms to species whenever possible. If keys were not available, or a specimen could not be identified to species, then it was identified to the lowest taxonomic level possible, usually genus.

### A1.4 DATABASE CONSTRUCTION

Between 1983 and 1989, we amassed a database of 145 benthic macroinvertebrate samples from one primary habitat type: free-flowing, mid- to high gradient, erodible bottom streams with samples collected during a late summer index period. Additional standard protocols were followed for sampling macroinvertebrates in impounded rivers. Data quality assurance protocols included (1) standardized and documented stream collection procedures, performed under the direct supervision of an MDEP biologist; (2) supervised sample sorting with a proportion of each sorter's samples resorted by another person to determine sorting efficiency; (3) consistent taxonomy (about 80% of samples identified by the same taxonomist); and (4) a special reference collection of separate taxa to standardize taxonomy for the program (MDEP 2009). Quality assurance protocols for data entry and data editing were applied during transfer of raw data to the computerized database management system and are documented in the report by MDEP (2009). In 2000 the database was migrated to Oracle and ArcInfo. Geographic information system (GIS) technology was interfaced with the stream macroinvertebrate database to facilitate future sampling and analysis of spatial dependencies (Dawson et al. 2002).

We standardized all taxonomic identifications to the genus level prior to metric computation and statistical analysis. It is difficult or impossible to identify many groups of benthic macroinvertebrates to the species level due to subtle physical differences between closely related species within a genus, as well as the continuing discovery of new species. On the other hand, some taxa are easy to identify to species, thus it is quite common for data to be submitted with varying levels of taxonomic resolution. Varying levels of effort in identification to the species level risks instability in subsequently derived richness measures. This inconsistency is incompatible with the need to establish standardized attainment

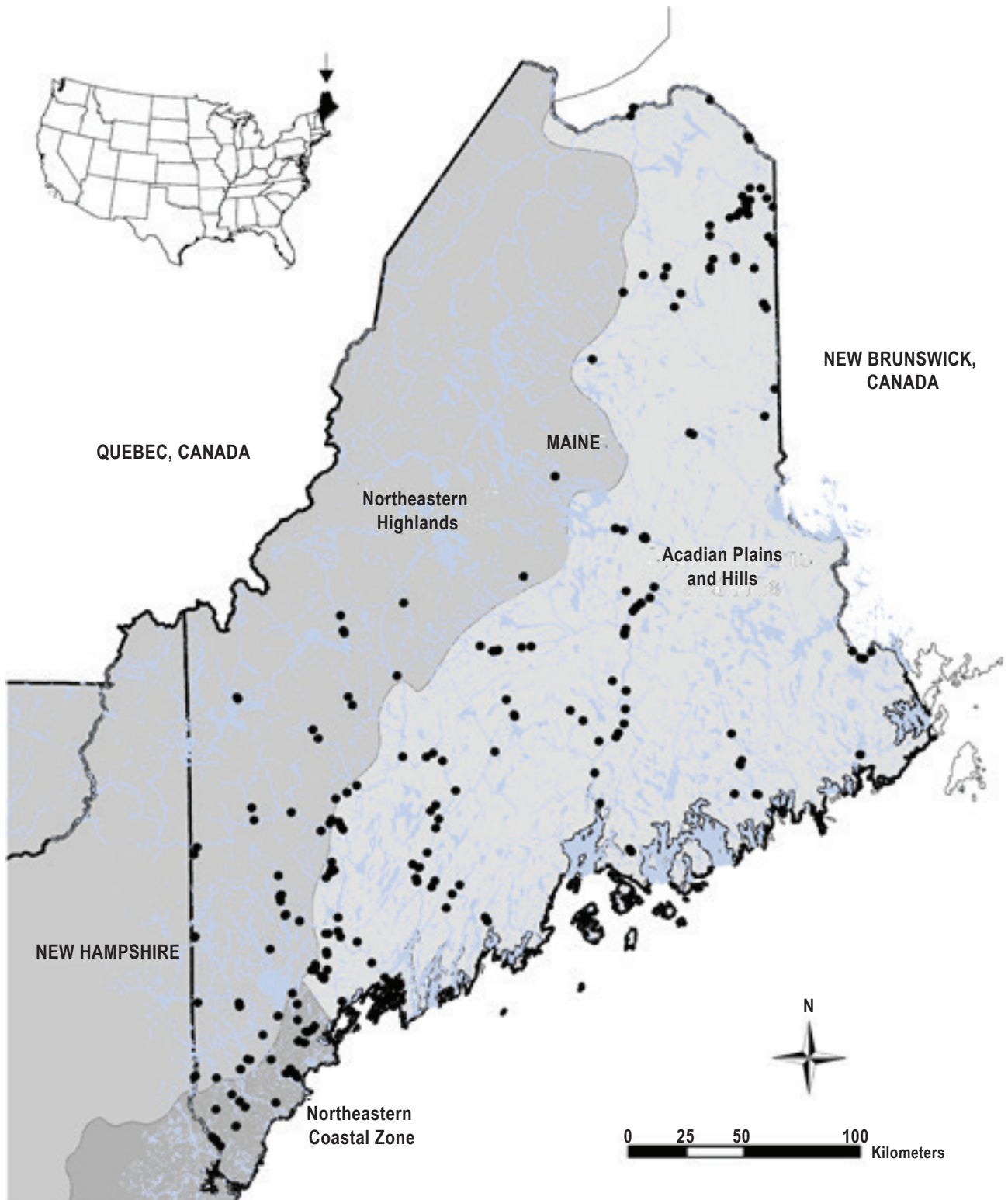


Figure A1. Stream-sampling locations (n=224 stations) from which 373 samples were collected to build a statistical model to predict attainment of aquatic life criteria. Some locations were sampled more than once. (Northeastern Coastal Zone, n=28; Acadian Plains and Hills, n=147; Northeastern Highlands, n=49).



guidelines for aquatic life. For this reason, all organism counts recorded in taxonomic units other than genus are standardized to the genus level prior to computation of metrics or other quantitative analyses (Appendix 4).

### **A1.5 MINIMUM PROVISIONS FOR SAMPLE DATA FOR ANALYSIS VIA THE STATISTICAL MODEL**

Samples are first evaluated to determine if they are appropriate for analysis via the predictive models. Appendix 7 provides protocols concerning treatment of atypical sampling situations and decisions to reject unsuitable samples. Samples must have a mean total abundance of greater than 50 organisms per sampler and a total genus richness (from three samplers) of more than 15 taxa. These minimum total abundance and richness provisions are derived from the minimum values found in reference-quality sites in the baseline data set. Sample data are examined for atypical findings or evidence of environmental or sampling conditions for which the model was not designed. Examples of conditions that could initiate adjustment of the model decision are unusual habitats (e.g., tidal flows, lack of flowing water, or sampler stranding), natural or human-induced disturbance of the sample, or known or suspected problems with sample collection or analysis. For samples having any of these characteristics—indicating that they are not appropriate for analysis by the predictive models—a professional judgment protocol (CMR Chapter 579, 2003; Appendix 7) can be used to determine whether the data suggest actual nonattainment conditions, natural causes for the low richness and/or numbers obtained, or that resampling is required to establish sufficient confidence to make a final determination.

Samples that are appropriate for the model predictions are sequentially run through the first (four-way) and second stage (two-way) models to make final determinations (Part 2, Figure 2; Appendix 5). The second-stage two-way model prediction that produces the highest probability for a given class is determined to be the best classification fit (Figure 2; Appendix 8). Probability cutoffs used in discriminant analysis are arbitrary, but traditionally assign a probability marginally greater than 0.50 to determine class membership. However, because we use these models for regulatory determination, which may have significant management consequences, we use a more conservative criterion of  $p > 0.60$  for attainment of aquatic life criteria for a class (CMR Chapter 579, 2003; Appendices 5, 6, and 8). By the

same reasoning,  $p < 0.40$  means that a sample does not attain the aquatic life criteria of a class. For probabilities falling between 0.40 and 0.60, the result is indeterminate and may require further review or resampling. In actual application, the predictions of the two-way models are usually very decisive, with the majority of probability estimates to a given class exceeding  $p > 0.90$  and nonattainment of a given class of  $p < 0.10$

## APPENDIX 2. MODEL-BUILDING METHODS

### A2.1 UNIVARIATE ANALYSES—EXPLORATORY INVESTIGATIONS

We performed exploratory statistical analyses to examine univariate relationships between physical stream characteristics and biological responses. Univariate statistical approaches, especially graphical inspection and linear correlation analysis, were conducted to examine the distribution of what were hypothesized, based upon theory, to be significant descriptors of the benthic communities sampled (Figure A2). For instance, Figure A2a shows that reference streams have higher EPT richness than nonreference streams (those streams that are more likely to be affected by pollutants). We also examined relationships between biological variables and physical stream covariates such as stream width, depth, velocity, substrate composition, conductivity, and stream temperature by graphically analyzing various subsets of data (Figure A2b). The data in Figure A2b show a negative relationship between turbidity (as measured by conductivity) and EPT richness. These are examples of relationships that guided our initial model building. In addition, our exploratory phase of analysis provided an assessment of the effects of geographic and climatic regions on biological response variables. The results

from these preliminary analyses were used to develop an initial structure for predictive models.

While there were a few significant relationships between physical stream characteristics (e.g., whether a stream is perceived to be reference quality or affected, or water conductivity, as in Figure A2a and b) and biological variables (such as EPT richness) in general, few significant relationships were found. The exploration of relationships between biological variables and physical stream characteristics such as stream width, depth, velocity, substrate composition, and stream temperature are shown in Table A1. We did not find strong relationships between biological response variables and stream width, depth, or velocity. We also did not find strong relationships between physical stream characteristics and multivariate discriminant axes as depicted in Table A2. The exploratory phase of analysis also demonstrated only a minimal effect of geographic or climatic region (Figure A1) on biological response variables. We concluded from these analyses that most biological variables did not exhibit a linear relationship with natural physical streambed characteristics and geographic locations, indicating that a complex stratified or hierarchical model partitioned by stream type, stream order, or geographic locale in Maine was not necessary.

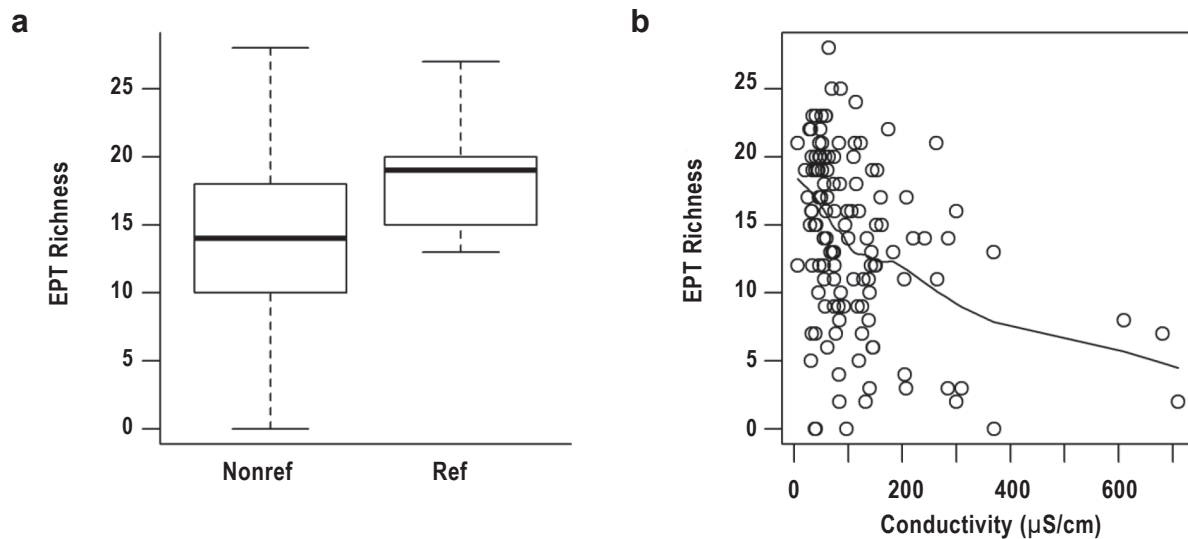


Figure A2. Exploratory plots for the richness of EPT with (A) comparison of samples from reference (Ref,  $n = 57$ ) and nonreference (Nonref,  $n = 310$ ) sites with significant difference of means of the two groups (Mann-Whitney  $U = 4645.5$ ,  $p < 0.001$ ), and (B) plot of EPT Richness vs conductivity (a surrogate for generalized human disturbance) with locally weighted (LOESS) regression line.

**Table A1.** Exploratory univariate data analysis: Pearson correlation of physical determinants with biological metrics and first-stage model results.

Biological Response Variable	Pearson Correlation Coefficients			
	Depth (n=604)	Width (n=571)	Velocity (n=470)	Temperature (n=585)
Total Abundance (TOTAB1)	-0.150	-0.071	0.119	-0.040
Richness (GENRICH2)	-0.194	-0.107	-0.065	-0.080
Hilsenhoff Biotic Index (HBI6)	0.327	0.030	-0.151	-0.006
Shannon-Weaver Diversity (SWDI5)	-0.081	-0.106	-0.096	-0.007
$\rho$ (Class A) First-Stage Model (pA1)	-0.204	-0.058	-0.031	-0.040
$\rho$ (Class B) First-Stage Model (pA1+pB1)	-0.117	0.052	0.025	-0.045
$\rho$ (Class C) First-Stage Model (pA1+pB1+pC1)	0.096	0.001	0.045	0.003
$\rho$ (Class NA) First-Stage Mdl	0.204	0.017	-0.032	0.071

**Table A2.** Correlations ( $\rho$ -values) between physical stream characteristics and dependent canonical discriminant axes.

Physical variable	Percentage of data set for which the parameter was collected	Correlations ( $\rho$ -values) between physical stream characteristics and dependent canonical discriminant axes			
		A	B	C	NA
Stream order	100	0.745	0.633	0.422	0.678
Temperature	87	0.345	0.677	0.344	0.213
Width	87	0.255	0.796	0.365	0.79
Depth	92	0.278	0.772	0.823	0.599
Velocity	69	0.603	0.722	0.645	0.505
Percentage Sand	92	0.444	0.763	0.805	0.657
Percentage Detritus	92	0.447	0.599	0.397	0.45
Percentage Silt	92	0.501	0.897	0.654	0.842

## A2.2 ASSESSMENT AND SELECTION OF MODEL PREDICTOR VARIABLES

Nearly 500 variables were available for developing a predictive model (Table A3). However, the stability of predictions based on linear discriminant functions depends upon the ability to precisely estimate linear coefficients or model parameters, which, in turn, are dependent upon a suitable number of degrees of freedom. In general, this requires that the ratio of cases to the number of variables should be in the range of 10 to 30 for reliable estimation of coefficients (Wilkinson 1989; Manly 1991). To select variables for the model, we applied data reduction techniques, in combination with biologists' recommendations on retention of critical

ecological attributes that contributed to defining the biological criteria classes. Because the model was to serve as numeric biological criteria to assess attainment of water quality goals for biological condition, defined by Maine's classification standards, special emphasis was placed on including biological variables directly related to the aquatic life goals and definitions described in the statutory classification standards (see Part 1, Tables 1, 2, and 5). We used various transformations, such as z-standardization, ordinal ranking, arcsin, square root, and logarithms, to normalize variance or achieve homoscedasticity (Elliot 1977) in individual predictor variables, based on the assumption that transformation of individual variables that appear to violate

**Table A3.** Variable types screened for use in linear discriminant models.

Variable Type	Example	Number Evaluated	Number Selected
Generic Abundances	<i>Stenonema</i> , <i>Hydropsyche</i> , <i>Brachycentrus</i>	300	12
Indicator Taxa	Class A- <i>Serratella</i> , <i>Brachycentrus</i> , <i>Leucrocuta</i> , <i>Glossosoma</i> , <i>Paragnetina</i> , <i>Euylophella</i> , <i>Psilotreta</i>	90	7
Measures of Richness	Richness; EPT; EP	10	6
Taxa complexes	( <i>Acroneuria</i> + <i>Stenonema</i> ); ( <i>Cheumatopsyche</i> + <i>Cricotopus</i> + <i>Tanytarsus</i> + <i>Ablabesmyia</i> )	+/-50 genera	3
Family Functional Groups (112 genera)	Perlidae; Tanypodinae, Chironominae	24	3
Habitat variables	Temperature, depth, width, velocity, percentage substrate composition, stream order	8	0
Functional Feeding Group	Collector-filterer, predator, deposit-feeder	5	0

assumptions of normality and heteroscedasticity can, in some cases, result in data that approximate a joint distribution that is multivariate normal (Lachenbruch et al. 1973; Manly 1991). We used varimax factor rotation (Yates 1987; Reyment 1993) to identify suites of highly correlated variables. Once identified, the best discriminating variables from each suite were assessed, thus reducing variable redundancy and dimensionality.

We did not find the abundance of individual genera or species to be good predictors of the biologist classifications in the discriminant model, except in the case of specific Class A indicator taxa. This is not surprising since the abundance of individual species fluctuates dramatically from year to year and stream to stream within a particular water quality class, as found by Clarke et al (2002). Criteria for selecting the Class A indicator taxa required that at least 60% of the total abundance of the taxon collected in the entire data set occurs in a priori Class A samples. For a genus to be useful as an indicator across the full range of sampled conditions, we restricted indicator candidates to those genera that occur in more than 10% of the samples in the database. This restriction ensures a sufficiently high probability of capture within any of the four water quality groups to make a taxon a useful indicator.

To maximize the ability to discriminate biological condition classes on the basis of ecological characteristics, we considered various functional groups of aquatic invertebrates for analysis. We tested variables based on Cummins's (1973) functional feeding groups that aggregate taxa with similar feeding morphology.

### A2.3 INITIAL ASSESSMENT OF WATER QUALITY CLASSES BASED UPON DATA-STRUCTURED CLUSTERS

Natural or data-derived groupings from clustering and ordination approaches did not correlate well with water quality gradients known to occur in the data set. For example, the K-means approach identified four clusters comprised of 44, 5, 84, and 240 stream samples. Three of the clusters form an aggregated group leaving only two well-defined regions with the distinct cluster (cluster 2) consisting of only five samples (Figure A3a). In addition, the legislative stream classes used for K-means clustering were found comprise all cluster groups (Figure A3b) suggesting little potential for these overlapping clusters as regulatory guides. This led to the approach of using professional (aquatic biologist) expert judgment of water class assignment to the sampled streams in Maine. This approach is discussed in detail in Part 2, Sections 2.4.3, 2.4.4, and 2.4.5.

### A2.4 FIRST-STAGE LINEAR DISCRIMINANT CLASSIFICATION MODEL

The initial predictive four-way discriminant model that we constructed was based on the original 145 sample database, using the biologist classification. Developed in 1992, these first-cut algorithms resulted in a reasonable predictive model with correct prediction of the biologist classifications averaging about 70% (compared to an expected 25% classification under the null hypothesis). Because of the promising predictive capability, the four-way model was re-parameterized in

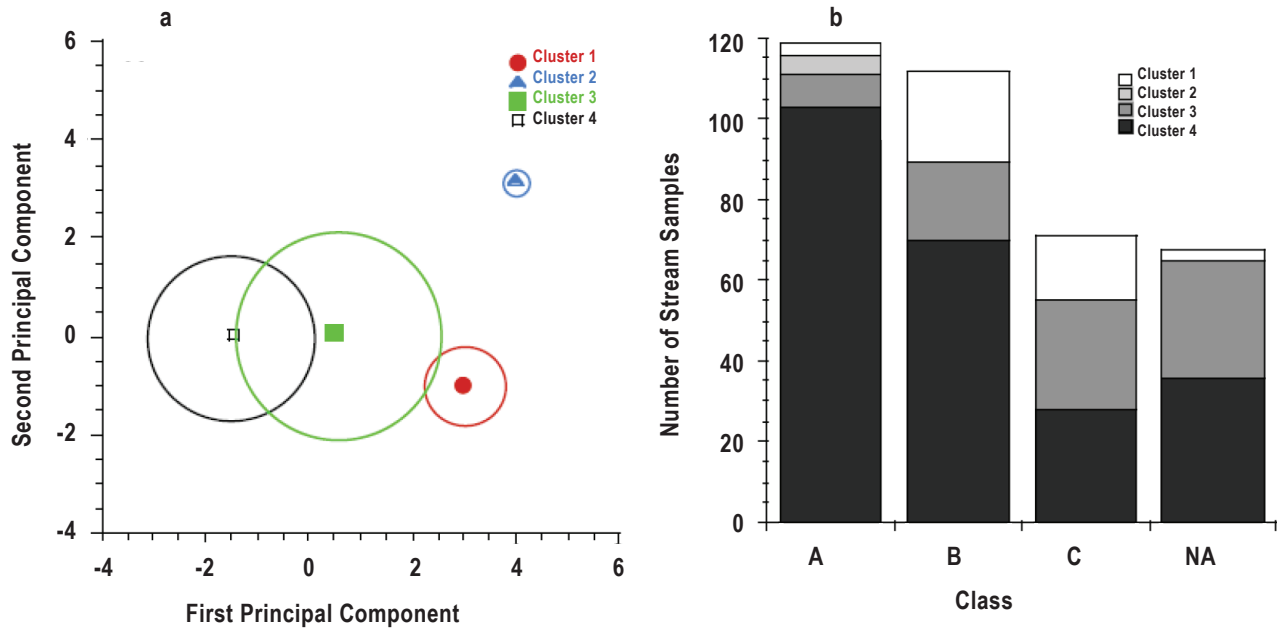


Figure A3. Results of k-means clustering with four groups selected. Cluster centroids and 95% circles (a) and distribution of streams from the four water classes in each of the four clusters (b).

1998 with a larger stream data set ( $n = 373$  samples, inclusive of the initial 145 samples). ANOVA and MANOVA provided evidence that the linear discriminant model does separate the biologist classifications as significantly differentiated populations (Table A4, MANOVA table). In addition, all of the predictor variables selected for the final model were highly significant in discriminating the biologist classifications (Table A4, ANOVA Table). Although the water quality class populations are significantly different as summarized by the univariate and multivariate ANOVA statistics in Table A4, this is not a necessary criterion for a highly predictive classification model. Table A5 lists variable transformations and the four-way discriminant model coefficients used to predict the probability of class membership for individual river and stream samples, and Table A6 lists population and distributional summary statistics. Appendix 4 shows variable definitions and computational algorithms. Figure A4 shows an ordination of the first two canonical discriminant axes of the four water quality class populations of streams and rivers. The overlap of the four stream class populations as predicted by the model shows overlap,

reflecting that each stream has a probability or likelihood

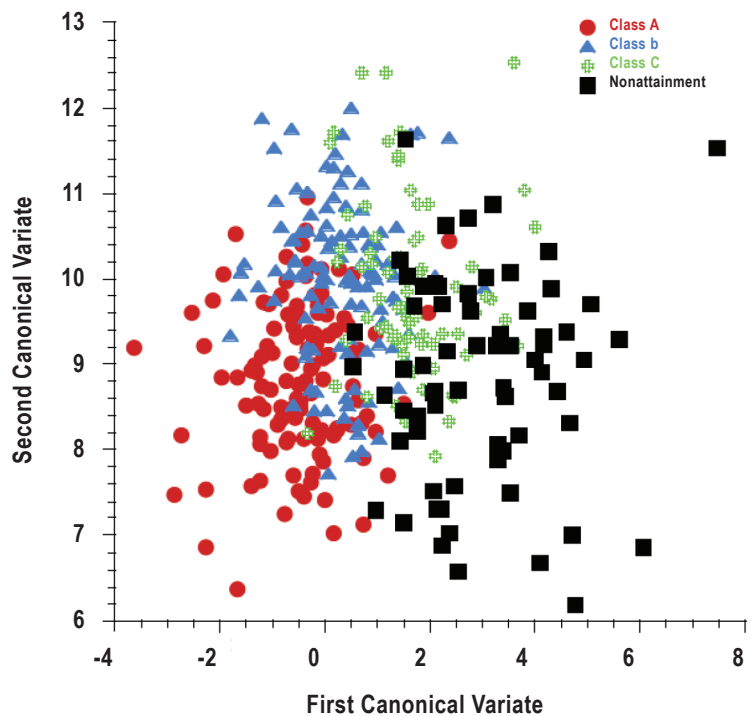


Figure A4. Canonical scores for the 373 sites plotted against the first two canonical variates derived from the four-way linear discriminant model used to separate all four water quality classes.

**Table A4.** Univariate analyses of variance (ANOVA) and multiple analysis of variance (MANOVA) and for the four-way linear discriminant model (n=373 sample sites).

<b>ANOVA</b>					
<b>Discriminant Variable</b>	<b>SS</b>	<b>df</b>	<b>MS</b>	<b>F value</b>	<b>p-value</b>
Log total abundance	74.8052	3	24.9351	22.9059	<0.0001
<i>Residual error</i>	401.6874	369	1.0886		
Generic richness	11683.3281	3	3894.4427	35.1949	<0.0001
<i>Residual error</i>	40831.2349	369	110.6538		
Log Ephemeroptera	3331.6033	3	1110.5344	172.9364	<0.0001
<i>Residual error</i>	2369.5837	369	6.4216		
Log Plecoptera abundance	1749.4512	3	583.1504	143.4626	<0.0001
<i>Residual error</i>	1499.9208	369	4.0648		
Biotic index	58.0507	3	19.3503	40.4781	<0.0001
<i>Residual error</i>	176.3975	369	0.4780		
Species diversity	188.8227	3	62.9409	80.7877	<0.0001
<i>Residual error</i>	287.4842	369	0.7791		
Log rel. Chironomidae	4.50879	3	1.5029	2.0114	0.0486
<i>Residual error</i>	275.7083	369	0.7472		
Relative Diptera richness	0.1248	3	0.0416	2.9269	0.0337
<i>Residual error</i>	5.2424	369	0.0142		
<i>Hydropsyche</i> abundance	2232441.2	3	744145.5	7.3803	<0.0001
<i>Residual error</i>	37205735.6	369	100828.2		
<b>MANOVA</b>					
<b>Test Statistic</b>		<b>df</b>	<b>Test value</b>	<b>Asymptotic F</b>	<b>p-value</b>
Wilkes Lambda		27,1054	0.1596	34.1634	<0.0001
Pillai Trace		27,1089	1.0956	23.2034	<0.0001
Hotelling – Lawley		27,1079	49.5773	49.5773	<0.0001

**Table A5.** Four-way discriminant model variable transformations and coefficients used to predict the probability of class membership for individual stream samples.

<b>Discriminant Model Coefficients</b>					
<b>Variable Name</b>	<b>Transformation</b>	<b>Class A</b>	<b>Class B</b>	<b>Class C</b>	<b>Nonattainment</b>
Constant		-99.95508	-105.70948	-112.67581	-107.74283
1. Total Abundance (TOTAB1)	nLog (value+0.001)	10.77061	11.46981	11.80888	11.26793
2. Generic Richness (GENRICH2)		-0.038619	-0.43340	-0.50051	-0.48822
3. Plecoptera Abundance (PLECAB3)	nLog (value+0.001)	0.23940	0.03946	-0.60923	-0.95480
4. Ephemeroptera Abundance (EPHAB4)	nLog (value+0.001)	-0.59970	0.55500	-0.67722	-1.79032
5. Shannon-Weiner Generic Diversity (SWDI5)		21.22732	20.91256	21.07602	19.46547
6. Hilsenhoff Biotic Index(0-10) (HBI6)		8.01620	9.12163	10.31492	10.72746
7. Relative Abundance Chironomidae (CHIRA7)	nLog (value+0.001)	-11.70298	-11.52650	-1149414	-11.66371
8. Relative Diptera Richness (DIPTRR8)		70.77937	71.09637	72.46514	70.22517
9. <i>Hydropsyche</i> Abundance (HYDRAB9)		-0.00535	-0.00398	-0.00152	0.00007

**Table A6.** Summary statistics for linear discriminant model predictors (variables).

Statistics <sup>1</sup>								
Variables	N of cases	Minimum	Maximum	Range	Mean	Standard Dev.	Variance	C.V. Predictors
TOTAB1	366	9.33	9080	9070.67	707.46	950.61	903655.81	1.344
GENRICH2	366	4	72	68	34.22	12.21	149.17	0.36
PLECAB3	366	0	134.67	134.67	8.74	13.54	183.3	1.55
EPHAB4	366	0	668	668	95.85	114.66	13146.46	1.19
SWDI5	366	0.42	4.91	4.49	3.09	0.8	0.64	0.26
HBI6	366	1.81	8.49	6.69	4.6	1.13	1.28	0.25
CHIRA7	366	0	0.97	0.97	0.22	0.221	0.05	0.97
DIPTRR8	366	0	0.83	0.83	0.36	0.12	0.01	0.34
HYDRAB9	366	0	3306.59	3306.59	170.12	328.22	107727.66	1.93
CHEUMAB11	366	0	2028.31	2028.31	77.99	209.05	43700.28	2.68
EPT/DIPT12	366	0.1	8	7.9	1.45	0.94	0.89	0.65
OLIGRA13	366	0	0.95	0.95	0.03	0.12	0.01	4.15
PERLAB15	366	0	45	45	5.74	8.19	67.14	1.43
CHIMINI17	366	0	1116.8	1116.8	38.09	115.15	13259.72	3.02
EPHRA18	366	0	0.84	0.84	0.18	0.17	0.03	0.95
EPTR19	366	0	28	28	14.47	6.12	37.46	0.42
DMPHAB21	366	0	1789.47	1789.47	10.31	98.46	9694.65	9.55
PLECRR23	366	0	0.2	0.2	0.05	0.04	0.01	0.83
CCTAAB25	366	0	2050.11	2050.11	105.76	226.94	51500.48	2.15
ASAB26	366	0	361.33	361.33	31.37	46.08	2123.19	1.47
EPRATIO28	366	0	1.36	1.36	0.54	0.28	0.08	0.51
AIND30	366	0	0.86	0.86	0.19	0.19	0.04	1.02

<sup>1</sup> Taxa descriptions for variables are listed in Appendix 4.

of class membership and also that predictions are not 100% accurate.

Tables A1 and A2 list p-values from linear correlations between physical variables such as stream order, temperature or depth, and the water quality classes. The relationships between biological measures and physical stream characteristics such as stream width, depth, velocity, substrate composition, and stream temperature are shown in Table A1. We did not find strong relationships between biological response variables and stream width, depth, or velocity. We also did not find strong relationships between physical stream characteristics and multivariate discriminant axes as depicted in Table A2. We concluded from these analyses that most biological variables did not exhibit a linear relationship with natural physical streambed characteristics, nor a categorical

relationship with geographic locations indicating that a complex stratified or hierarchical model partitioned by stream type, stream order, or geographic locale in Maine was not necessary.

We also examined the values of the nine predictor variables used in the first-stage four-way model to determine if there was a pattern with USEPA Level III ecoregions (Omernik 1987). The Acadian Plains and Hills (eastern ecoregion), Northeastern Highlands (western ecoregion), and Northeastern Coastal Zone (southern ecoregion) cover approximately 53%, 43%, and 4% of the state of Maine, respectively (Figure A1). Agricultural, rural, and urban land uses are not uniformly distributed throughout the state. The southern ecoregion has a disproportionately high amount of development compared to the other ecoregions. Because we were

seeking to determine natural differences between ecoregions that might influence our water quality groups, in this analysis we did not want to identify differences based on land use disturbance. Thus, we restricted ANOVA analyses of the nine predictor variables to reference samples from low to very low landscape disturbance (biologist classifications of A). We found no significant differences ( $p > 0.05$ ) related to ecoregions, indicating that predictive models would not have to be built for each ecoregion.

We also found that most of the functional feeding group variables were not significant predictors of the biologist classifications. As an alternative, we tested variables based on family functional groups. By using families as functional groups, sets of morphological and functional species traits, often related at the family level, (e.g., life history, reproduction, mobility, trophic level, as well as feeding morphology) can be aggregated as an expression of functional organization (Poff 1997). Importantly, these group functional traits may also include environmental tolerances, although they would be less precise than at the genus or species level. These family group traits can be used to assess the functional character of communities (Courtemanch 1993). In the test data set of more than 300 genera, fewer than 30 genera occurred in at least 25% of the sites (Figure A5). At the genus level, only *Stenonema*, *Cheumatopsyche*, *Hydropsyche*, and *Polypedilum* were collected in more than 75% of the samples. The higher-level taxonomic groups (families, orders, and family functional groups) and aggregated indices (such as richness, diversity, and biotic index) performed best for a discriminant model

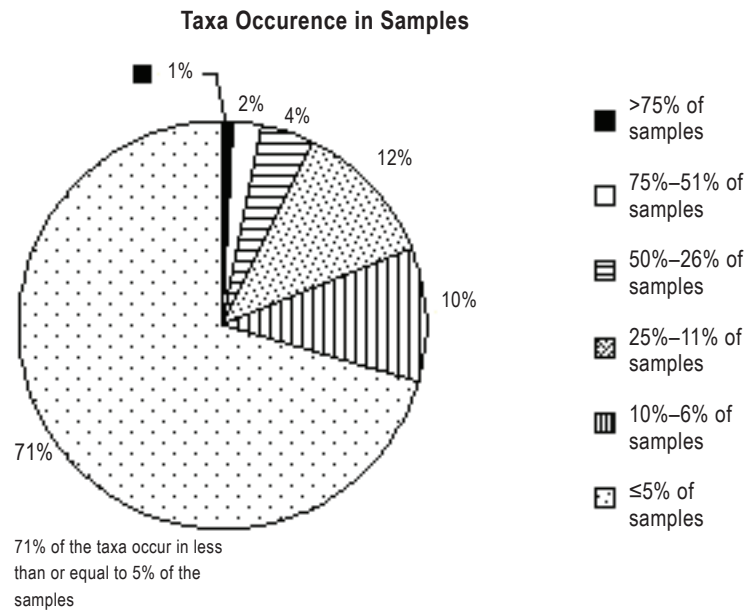


Figure A5. Frequency of taxa occurrence in samples.

approach to stream classification based upon the benthic community. The notable exception to the family functional group concept is the Chironomidae. Formation of family functional groups for Chironomidae entailed reducing this large family of functionally diverse midge taxa into subfamily groups having more similar morphological and functional characteristics.

The four-way discriminant model was used to estimate the probability of a sample belonging to each of the water quality classes (A, B, C, and NA) and performance was evaluated against the biologist classifications. We assigned a sample to the classification with the largest probability of class membership (Ross 1989). The predicted class assignments correctly matched 72.5% of the biologist classifications of A (Table A7). The model

**Table A7.** Classification of stream and river sites by Phase II (373 samples) four-way linear discriminant model. Numerical entries represent the percentage (number) of sites classified from a priori classes (row) into predicted classes (columns). Therefore, diagonals (**bold**) are percentage correct classification.

A priori class	Model Predicted Class			
	Class A	Class B	Class C	NA
Class A	<b>72.50%</b> (87)	27.50% (33)	0.00% (0)	0.00% (0)
Class B	22.12% (25)	<b>69.03%</b> (78)	8.85% (10)	0.00% (0)
Class C	2.78% (2)	27.78% (20)	<b>65.28%</b> (47)	4.17% (3)
NA	0.00% (0)	2.94% (2)	25.00% (17)	<b>72.06%</b> (49)



**Table A8.** Jackknife estimates for the four-way linear discriminant model (n=373) using four-fifths of the data. Numerical entries represent the percentage of sites classified from a priori classes (row) into predicted classes (columns); therefore, diagonals are percentage correct classification. Standard error is in parentheses.

A priori class	Model Predicted Class							
	Class A		Class B		Class C		NA	
Class A	<b>74.6%</b>	<b>(8.4)</b>	28.5%	(1.8)	0.7%	(0.5)	0.0%	(0)
Class B	22.3%	(3.4)	<b>69.0%</b>	<b>(9.2)</b>	9.8%	(3.3)	2.3%	(1.2)
Class C	1.4%	(0.9)	28.9%	(4.5)	<b>63.3%</b>	<b>(8.9)</b>	5.3%	(1.0)
NA	0.0%	(0)	2.1%	(0.4)	24.8%	(2.4)	<b>75.6%</b>	<b>(6.7)</b>

correctly matched biologist classifications of B, C, and NA for 69%, 65%, and 72% of samples, respectively.

We further tested the four-way model by performing a jackknife discriminant procedure. The results of jackknife randomization procedures confirmed the repeatability and accuracy of the first-stage model to predict a priori classes. It also indicated that the estimation of the discriminant functions is not very sensitive to small changes in the data set, and that both coefficients and misclassification do not vary greatly between iterations (Table A8). This was encouraging because one problem that can arise from this modeling approach is that by mixing populations (A vs B, C, NA) one could end up with a population structure that results in uneven covariance between populations, a violation of one of the assumptions of linear discriminant analysis (Jobson 1992). However, based upon the jackknife results this is not the case for our data set.

While the four-way model offered reasonably good predictive ability, there was clearly too much misclassification for its effective use in a state regulatory program, where such errors can be contentious and have costly management consequences (Courtemanch et al. 1989). Therefore, we developed a series of hierarchical two-way discriminant models that used the predictive information from the four-way model to reduce the level of misclassification.

## A2.5 HIERARCHICAL SYSTEM OF DISCRIMINANT MODELS

We constructed three independent two-way models (Part 2, Figure 2) to predict streams as (1) A (Table A9), (2) B-or-better model (Table A10), and (3) C-or-better model (Table A11). The two-way models estimate probabilities of belonging to the two aggregated classes associated with each model (i.e., prob(ABC) vs prob(NA)

for the C-or-better model, prob(AB) vs prob(CNA) for the B-or-better model, and prob(A) vs prob(BCNA) for the A model). The percentage correct classification of the two-way LDMs (linear discriminant models) ranged from 90% to 97% (Tables A12–A14). Figure 11 shows the distribution of the variables used in the two-way LDMs. ANOVA and MANOVA statistics for the model variables are presented in Tables A9–A11. Table A15 provides variable transformations and coefficients used to predict the probability of group (aggregated or single water quality classes) membership for individual stream samples in the two-way models.

The results of jackknife randomization procedures for the completed two-way models, as in the four-way model, confirmed the repeatability and accuracy of the models to predict a priori water quality classes (Section A2.6 and Table A16). This can be seen by inspecting the standard errors of the coefficients, usually less than 10% of the mean of the coefficient (Table A15). These repeated jackknife fits also indicated that the estimation of the discriminant functions are not very sensitive to small changes in the data set and that both coefficients and misclassification do not vary greatly between iterations.

Validation tests of the four-way and two-way models also were performed using an independent data set of 34 sample sites not used in building the models. Results of the validation test sites for the four-way model predictions are summarized in Table A17. The prediction of the new test data resulted in excellent accuracy (100% correct classification) for the class B and NA sites. The class A and C predictions were less reliable at 75% and 71% correct classification, respectively. In most cases where the model prediction differed from the biologist classifications, examination of the biologists' sample evaluation notes indicated that a site was considered borderline between two classes, or at least one biologist had assigned the site to the same class that the model

**Table A9.** Univariate analyses of variance (ANOVA) and multiple analysis of variance (MANOVA) for two-way Class A vs Class B, C, and NA discriminant model (n=373 sample sites).

ANOVA					
Discriminant Variable	SS	df	MS	F	p-value
$\sin^{-1}(\text{Prob}(A))^{0.5}$	27.8311	1	27.8311	411.3409	<0.0001
Residual error	25.1016	371	0.0677		
Relative Plecoptera richness	0.1260	1	0.1260	103.6233	<0.0001
Residual error	0.4511	371	0.0012		
Relative EP generic richness	8.3857	1	8.3857	166.5738	<0.0001
Residual error	18.6771	371	0.0503		
Class A indicator taxa	5.0102	1	5.0102	214.4091	<0.0001
Residual error	8.6693	371	0.0238		
Log ( <i>Cheumatopsyche</i> + <i>Cricotopus</i> + <i>Tanytarsus</i> + <i>Ablabesmyia</i> abundances)	109.0172	1	109.0172	22.9045	<0.0001
Residual error	1765.8313	371	4.7597		
Log ( <i>Acroneuria</i> + <i>Stenonema</i> abundances)	348.1900	1	348.1900	24.9462	<0.0001
Residual error	5178.2778	371	13.9576		
MANOVA					
Test Statistic		df	Test value	Asymptotic F	p-value
Wilkes Lambda		6, 366	0.3991	91.8426	<0.0001
Pillai Trace		6, 366	0.6001	91.8426	<0.0001
Hotelling-Lawley		6, 366	1.5056	91.8426	<0.0001

predicted. The results of validation by the test data set indicated that the model predictions are reflective of the biologist classification rankings and that the majority of incorrect predictions are with borderline cases (i.e., adjacent classes).

Plots of predictors in Figures A6, A7, and A8 depict the distribution and measure of central tendency (median). It can be seen that the overlap in distributions of each of the predictor variables measured in the sampled 373 streams is less for predictors with higher levels of statistical significance. These plots show the overlap of the metrics as they are segregated among the biologist-assigned classes.

## A2.6 VALIDATION AND ROBUSTNESS OF THE HIERARCHICAL MODEL

The jackknife procedure was conducted with the data set of 373 samples that was randomly divided into five similarly sized subsets. Then a new linear discriminant model was estimated using four-fifths of the

data, leaving one of the subsets out for evaluation by the new model as to site membership. This procedure was repeated four times leaving a different subset of the data out each time. If the model-building data set (entire 373 samples) was characterized by numerous outliers or multimodality, then the expectation was that model parameters estimated with differing subsets of the data would not be consistent over the iterations and would thus produce a poor classification.

The jackknife analysis did not demonstrate a large departure in correct classification compared to the classification relying upon the entire data set (n=373). Class A classification with a jackknife analysis did show a very minor decline in correct classification (89.4% vs 90.0%), whereas B-or-better was almost the same (96.4% vs 96.5%) and C-or-better was more accurate (97.0% vs 96.1%) (Table A16). This suggests that the hierarchical models are robust to small random changes in central tendency and distribution in the data and smaller sample sizes, suggesting that the final model selected should hold for the diversity of streams and rivers in Maine.

**Table A10.** Univariate analyses of variance (ANOVA) and multiple analysis of variance (MANOVA) for two-way Class B or better vs Class C and NA discriminant model (n=373 sample sites).

ANOVA					
Discriminant Variable	SS	df	MS	F	p-value
$\sin^{-1}(\text{Prob(A)}+\text{Prob(B)})^{0.5}$	93.9467	1	93.9467	883.4044	<0.0001
<i>Residual error</i>	39.4544	371	0.1064		
Log (Perlidae abundance)	2455.6353	1	2455.6353	214.4809	<0.0001
<i>Residual error</i>	4247.6530	371	11.4492		
Log (Tanypodinae abundance)	140.0382	1	140.0382	12.9986	0.0004
<i>Residual error</i>	3996.9028	371	10.7733		
Log (Chironomini abundance)	21.9188	1	21.9188	2.1778	0.1093
<i>Residual error</i>	3739.5093	371	10.0795		
Relative abundance Ephemeroptera	3.1858	1	3.1858	151.3830	<0.0001
<i>Residual error</i>	7.8076	371	0.0210		
EPT generic richness	7017.6934	1	7017.6934	398.4813	<0.0001
<i>Residual error</i>	6533.7168	371	17.6111		
Log ( <i>Dicrotendipes</i> + <i>Micropsectra</i> + <i>Parachironomus</i> + <i>Helobdella</i> abundances)	388.6129	1	388.6129	25.7369	<0.0001
<i>Residual error</i>	5601.8998	371	15.0995		
MANOVA					
Test Statistic		df	Test value	Asymptotic F	p-value
Wilkes Lambda		7, 365	0.2609	147.7142	<0.0001
Pillai Trace		7, 365	0.7391	147.7142	<0.0001
Hotelling-Lawley		7, 365	2.8329	147.7142	<0.0001

**Table A11.** Univariate analyses of variance (ANOVA) and multiple analysis of variance (MANOVA) for two-way Class C or better vs Class NA discriminant model (n=373 sample sites).

ANOVA					
Discriminant Variable	SS	df	MS	F	p-value
$\sin^{-1}(\text{Prob(A)}+\text{Prob(B)} + \text{Prob(C)})^{0.5}$	73.4040	1	73.4040	1084.3244	<0.0001
<i>Residual error</i>	25.1153	371	0.0677		
Log ( <i>Cheumatopsyche</i> abundance)	1021.8266	1	1021.8266	75.1594	<0.0001
<i>Residual error</i>	5043.8810	371	13.5954		
(EPT richness/Diptera richness) <sup>0.5</sup>	6.2952	1	6.2952	6.2952	
<i>Residual error</i>	50.43034	371	0.1356		
Log (Rel. Oligochaete abundance)		134.4920	1	134.4920	58.6981
<i>Residual error</i>	850.0553	371	2.2913		
MANOVA					
Test Statistic		df	Test value	Asymptotic F	p-value
Wilkes Lambda		4, 368	0.2463	281.5361	<0.0001
Pillai Trace		4, 368	0.7537	281.5361	<0.0001
Hotelling-Lawley		4, 368	3.0602	281.5361	<0.0001

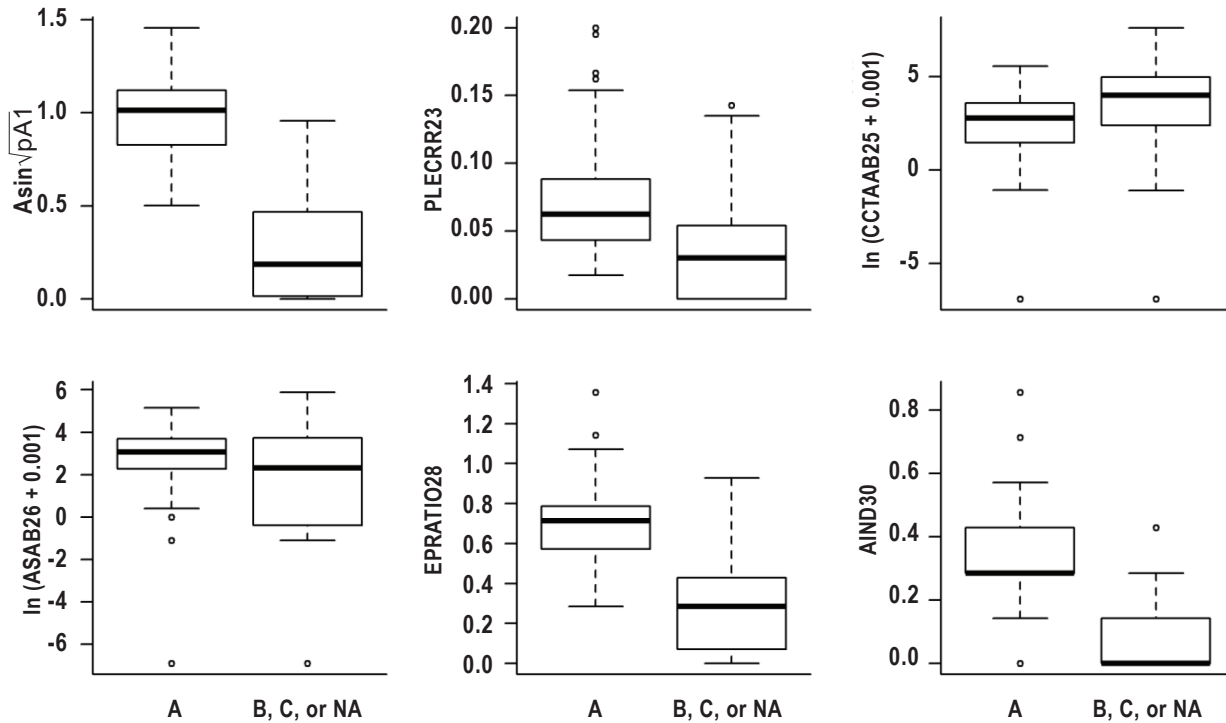


Figure A6. The distribution of variables within and between biologist-classified streams in the two-way Class A vs B, C, and NA linear discriminant model.

**Table A12.** Classification of stream and river sites by two-way linear discriminant model for the final A classification. Numerical entries represent the percentage (number) of sites classified from a priori or biologist classes (row) into predicted classes (columns). Therefore, diagonals are percentage correct classification (**bold**).

Model Predicted Class		
A priori class	Class A	Classes B,C, OR NA
Class A	<b>90.00%</b> (108)	10.00% (12)
Classes B,C,NA	10.28% (26)	<b>89.72%</b> (227)

**Table A13.** Classification of stream and river sites by two-way linear discriminant model for the final B-or-better model. Numerical entries represent the percentage (number) of sites classified from a priori or biologist classes (row) into predicted classes (columns). Therefore, diagonals are percentage correct classification (**bold**).

Model Predicted Class		
A priori class	Class B Or Better	Classes C, Or Na
Class B or Better	<b>96.57%</b> (225)	3.43% (8)
Classes C,NA	11.43% (16)	<b>88.57%</b> (124)

**Table A14.** Classification of stream and river sites by two-way linear discriminant model for the final C-or-better model. Numerical entries represent the percentage (number) of sites classified from a priori or biologist classes (row) into predicted classes (columns). Therefore, diagonals are percentage correct classification (**bold**).

Model Predicted Class				
A priori class	Class C Or Better		NA	
<b>Class C or Better</b>	<b>96.07%</b>	<b>(293)</b>	3.93%	(12)
<b>NA</b>	14.71%	(10)	<b>85.29%</b>	<b>(58)</b>

**Table A15.** Two-way discriminant model variable transformations and coefficients used to predict the probability of class membership for individual stream samples.

a) Class A model variable transformations and coefficients				
Variable number	Transformation	Class A	Class B-C nonattainment	
Constant		-9.59254	-4.08552	
22 Prob(A) first-stage model (pA1)	Arcsin	8.34341	1.52080	
23 Relative Plecoptera richness (PLECRR23)		3.78999	4.27447	
25 Sum abundances ( <i>Cheumatopsyche</i> + <i>Cricotopus</i> + <i>Tanytarsus</i> + <i>Ablabesmyia</i> ) (CCTAAB25)	nLog (value +0.001)	0.53110	0.77851	
26 Sum abundances ( <i>Acroneuria</i> + <i>Stenonema</i> ) (ASAB26)	nLog (value +0.001)	-0.55838	-0.51448	
28 Ratio EP generic richness (EPRATIO28)		12.32529	9.81592	
30 Ratio A indicator taxa (AIND30)		6.94828	-0.67475	
b) Class B-or-better model variable transformations and coefficients				
Variable number	Transformation	Class A-B	Class C nonattainment	
Constant		-17.81016	-6.93836	
14 Prob(A+B) first-stage model (pA1+pB1)	Arcsin	12.04826	3.63707	
15 Perlidae mean abundance (FFG) (PERLAB15)	nLog (value +0.001)	-1.11091	-1.03934	
16 Tanypodinae mean abundance (FFG) (TANYAB16)	nLog (value +0.001)	-0.10582	0.01978	
17 Chironomini mean abundance (FFG) (CHIMINI17)	nLog (value +0.001)	0.17813	0.10825	
18 Relative Ephemeroptera abundance (EPHRA18)		4.03202	-1.14508	
19 EPT richness (EPTR19)		0.87400	0.63310	
21 Sum abundance ( <i>Dicrotendipes</i> + <i>Micropsectra</i> + <i>Parachironomus</i> + <i>Helobdella</i> ) (DMPHAB21)	nLog (value +0.001)	-0.69360	-0.53194	
c) Class C-or-better model variable transformations and coefficients				
Variable number	Transformation	Class A-B-C	Nonattainment	
Constant		-25.70020	-8.55844	
10 Prob(A+B+C) first-stage model (pA1+pB1+pC1)	Arcsin	19.98470	3.36032	
11 <i>Cheumatopsyche</i> mean abundance (CHEUMAB11)	nLog (value +0.001)	-0.26001	-0.43781	
12 Ratio EPT-Diptera (EPT/DIPT12)	Sq. root	5.57672	5.92732	
13 Rel. Oligochaete abundance (OLIGRA13)	nLog (value +0.001)	-2.33229	-1.89945	

**Table A16.** Jackknife estimates using four-fifths of the data for the Phase II (373 sample) two-way linear discriminant model for the A classification (a), the B-or-better classification (b), and the C-or-better classification (c). Numerical entries represent the percentage of sites classified from a priori biologist classes (row) into predicted classes (columns), therefore, diagonals are percentge correct classification (**bold**). Standard error is in parentheses.

a) Model Predicted Class		
A Priori Biologist Class	Class A	Classes B,C, Or NA
Class A	<b>89.4%</b> (7.2)	8.2% (1.1)
Classes B,C, NA	8.6% (0.6)	<b>91.4%</b> (7.3)
b) Model Predicted Class		
A Priori Biologist Class	Class B Or Better	Classes C, Or NA
Class B or Better	<b>96.4%</b> (8.6)	5.5% (0.4)
Classes C,NA	6.7% (0.9)	<b>92.3%</b> (12.3)
c) Model Predicted Class		
A Priori Biologist Class	Class C Or Better	NA
Class C or Better	<b>97.0%</b> (8.5)	2.9% (0.4)
NA	12.2% (2.6)	<b>86.7%</b> (12.0)

**Table A17.** Validation data set: percentage correct four-way model predictions of classification for an independent 34-sample set (A: n=16; B: n=8; C: C=7; NA: n=3). Diagonals in table are correct classification percentages (**bold**).

Model Predicted Class				
A Priori Biologist Class	A	B	C	NA
<b>A</b>	<b>75%</b>	19%	6%	0%
<b>B</b>	0%	<b>100%</b>	0%	0%
<b>C</b>	0%	29%	<b>71%</b>	0%
<b>NA</b>	0%	0%	0%	<b>100%</b>

A small data set of 34 new streams was independently validated with the final first-stage model after it was parameterized. Table A17 shows that the final first-stage model accurately predicts the Class B streams and the nonattainment streams even better than with the original model-building data set (class B: 100% vs 69.0%, and NA: 100% vs 72.1%). The Class A and Class C streams are correctly classified with moderate success, 75% and 71%. This can be compared to a 25% correct classification by random chance. Though lower than for class B and NA, both the prediction for class A streams and for class C streams are close to the predictions of the model on the full data set (n=373) class A: 75% vs

72.5% and class C: 71% vs 65.3%. Therefore, the independent validation performed at least as well as the original model building data.

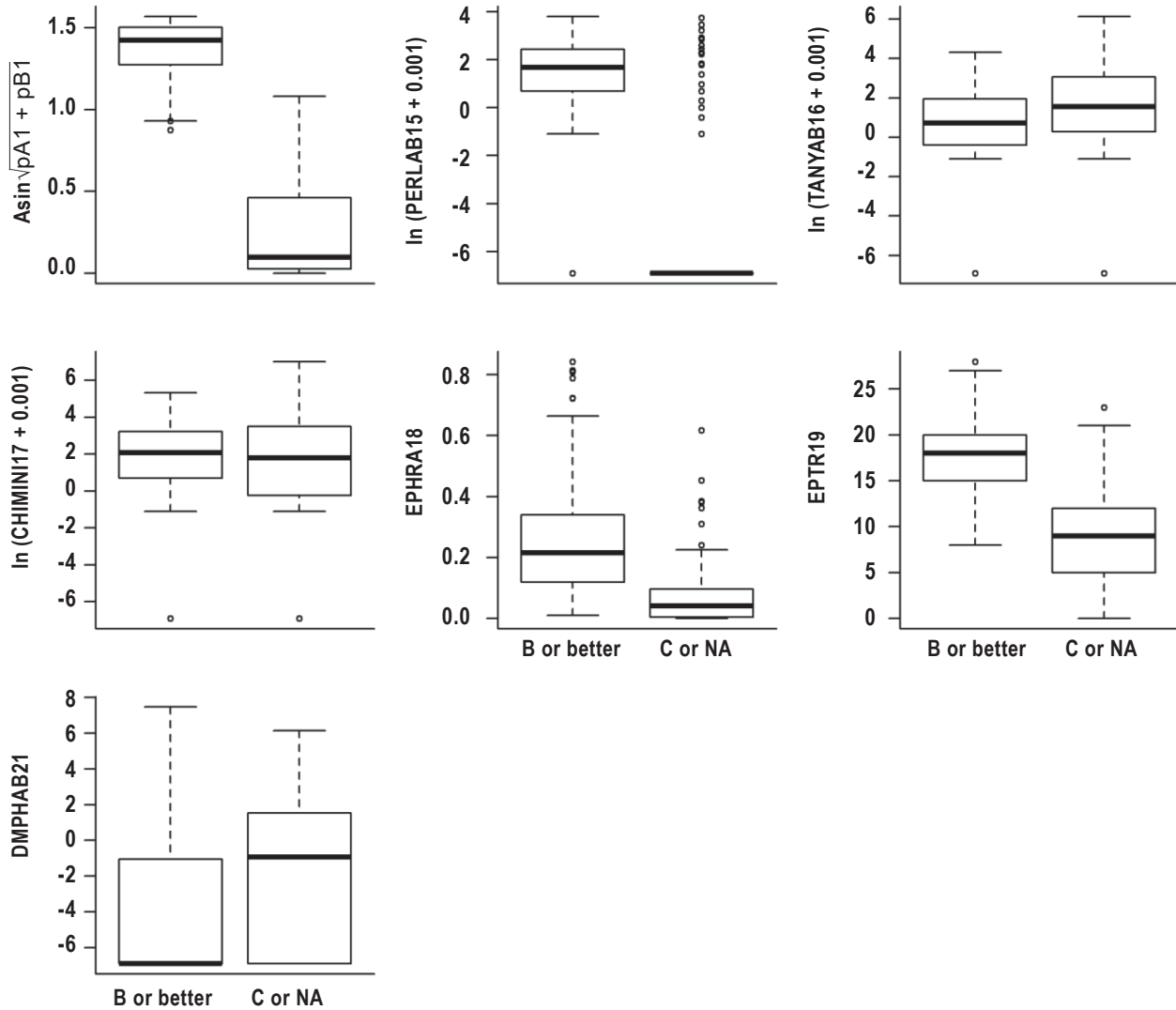


Figure A7. The distribution of variables within and between biologist-classified streams in the two-way Class B-or-better vs C, and NA linear discriminant model.

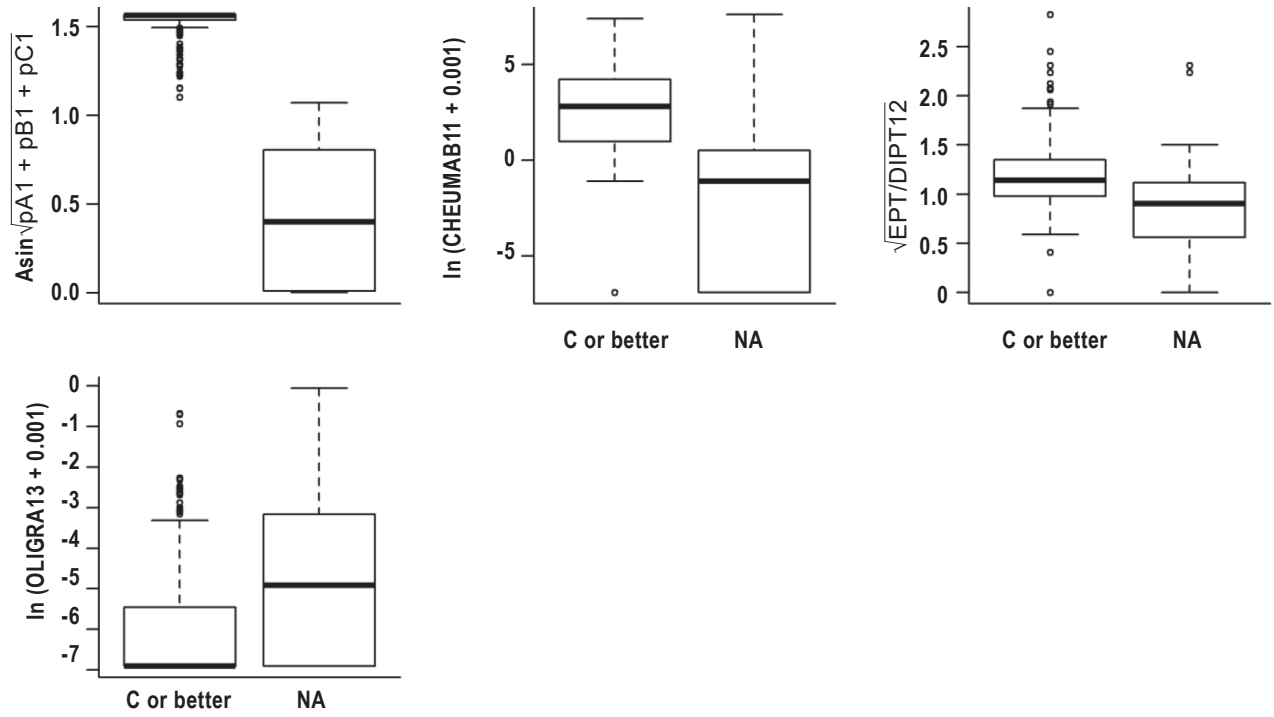


Figure A8. The distribution of variables within and between biologist-classified streams in the two-way Class C-or-better vs NA linear discriminant model.



## APPENDIX 3. PROCESS AND CRITERIA FOR THE ASSIGNMENT OF BIOLOGISTS' CLASSIFICATION

The biologists who initiated the process for a priori assignment of streams into water quality classes had training in aquatic entomology and were familiar with the macroinvertebrate communities in Maine. Their expertise and experience is summarized below.

### Raters' profile at the time of the original a priori assignment (1992)

#### David Courtemanch

- MS in aquatic entomology; PhD in environmental science; employed as a biologist in the Division of Environmental Assessment (DEA) in the MDEP for 16 years; currently director, Division of Environmental Assessment.

#### Susan Davies

- MS aquatic entomology; employed as a biologist in the River and Stream Section of DEA for nine years, coordinating the Biological Monitoring Program.

#### Leon Tsomides

- MS aquatic entomology; employed as a biologist in the River and Stream Section of DEA for three years, working with the Biological Monitoring Program.

### Ranking Process

Each biologist independently reviewed biological information for each sampling event (Table A18) including identities and abundances of taxa occurring in the biological sample and computed index values for the biological data (e.g., total abundance, diversity, richness, and EPT). Selected natural factors that could have an effect on biotic potential were also reviewed including water depth, velocity, substrate composition, and canopy cover in order to evaluate the effects of various natural habitat conditions on the structure of the macroinvertebrate community. Sample information was reviewed for the values of the given measures, relative to values for other samples in the data set. The actual classification assignment was determined by how closely the biological information conformed to the narrative aquatic life classification standards and definitions,

correcting for habitat effects. Numerical ranges, per se, were not established, a priori, for each measure. Instead, the information was reviewed for its compatibility with the mosaic of findings expected for each class, listed in the decision rules (Part 2, Table 6). The biologists did not have any knowledge of the actual location of the sampled sites, nor did they have knowledge of any information about pollution or human disturbance influences. Following the independent assignment of classes the biologists established a consensus classification by discussing justifications for each biologist's assignment (Bakus et al. 1982).

### Biologist's Classification Criteria

Each biologist reviewed the sample data shown in Table A18 for the values of a list of measures of community structure and function. Expected metric response trajectories used by biologists to evaluate each measure are listed in Part 2, Table 4.

**Table A18.** Community structure and function parameters reviewed by biologists.

Community structure and function parameters
Total Abundance of Individuals
Total Abundance of Ephemeroptera
Total Abundance of Plecoptera
Abundance of Ephemeroptera/Total Abundance
Abundance of Plecoptera/Total Abundance
Abundance of Hydropsychidae/Total Abundance
Abundance of Ephemeroptera+Plecoptera/Total Abundance
Abundance of <i>Glossosoma</i> /Total Abundance
Abundance of <i>Brachycentrus</i> /Total Abundance
Abundance of Oligochaetes/Total Abundance
Abundance of Hirudinea/Total Abundance
Abundance of Gastropoda/Total Abundance
Abundance of Chironomidae/Total Abundance
Abundance <i>Conchapelopia</i> + <i>Thiennemannymia</i> /Total Abundance
Abundance of <i>Tribelos</i> /Total Abundance
Abundance of <i>Chironomus</i> /Total Abundance
Generic Richness
Ephemeroptera Richness
Plecoptera Richness
EPT Richness
Ephemeroptera Richness/Generic Richness
Plecoptera Richness/Generic Richness
Diptera Richness/Generic Richness
Ephemeroptera+Plecoptera Richness/Generic Richness
EPT Richness/Diptera Richness
Non-EPT or Chironomidae Richness/Generic Richness
Percentage Predators
%Collector Filterers+Gatherers/%Predators+Shredders
Number of Functional Feeding Groups Represented
Shannon-Weiner Generic Diversity
Hilsenhoff Biotic Index
In addition, in cases where a valid clean-water upstream reference station existed, the following comparative index data were also reviewed:
Jaccard Taxonomic Similarity
Taxonomic Similarity Of Dominant Taxa
Coefficient Of Community Loss
Percentage Similarity

## APPENDIX 4. METHODS FOR THE CALCULATION OF INDICES AND MEASURES OF COMMUNITY STRUCTURE USED IN THE LINEAR DISCRIMINANT MODELS

Many of the taxa counts were used to derive complex variables (bioassessment metrics) representing higher-order ecological measures. These variables were only calculated from samples that were considered “complete”—i.e. where all counts were entered and verified using quality assurance (QA) protocols. The following procedures explicitly outline the methods for assembling the raw taxa data for statistical analysis.

### A4.1 ASSEMBLING AND COLLAPSING TAXA COUNTS

Maine DEP taxonomists recorded counts for each taxon observed in each subsampled replicate. Stored within the database, these counts reference both the sample replicate within which they are identified and information about the taxon they represent. Each taxon within the database is associated with a unique 2- to 14-digit phylogenetic code that indicates its taxonomic level and assignment within the phylogenetic hierarchy. Two or three digit pairings within the code represent different portions of the taxon’s phylogeny: digits one and two identify phylum; three and four identify class; five and six identify order; seven and eight identify family; nine, ten, and eleven identify genus; and eleven, twelve, and thirteen identify species. As such, a code of only two digits would be associated with a taxon at the phyla level, while a code of 14 digits would be associated with a species-level taxon. In addition, corollary information such as functional feeding preferences, tolerance values, and threatened or endangered status is stored for each taxon. Taxa are assigned a phylogenetic code number as shown in the following example:

In order to assemble the counts from the replicates and compensate for differences in subsampling, a mean count value is calculated for each taxon from the replicate counts and is then multiplied by the sample factor. So, for example, if a subsampling calls for sorting through only one-quarter of the sample, then after all taxonomic identification takes place, a mean count is calculated by multiplying the subsampled taxon count by the sample factor of four. Although pupae are recorded in the course of taxonomic identification, they are not used in the calculation of metrics and are therefore excluded from these processes.

Macroinvertebrate organisms are identified to genus whenever possible. If generic keys are not available or taxonomic expertise is lacking for a taxon, the macroinvertebrates should be identified to the lowest taxonomic level possible. Identification of organisms to species is highly recommended whenever possible, as these identifications are useful in the final stage of data analysis, the professional judgment evaluation of the model outcome.

To compensate for different degrees of resolution in taxonomic identification, all taxon counts are adjusted to the generic level. As such, species-level identifications and counts are aggregated to the generic level. Samples with family-level identifications are evaluated to determine if any genera (either as genus-level identifications or as generic components of species-level identifications) for these families exist in the sample. If one or fewer genera are identified within a family, the family-level identification maintains its count and is included in generic richness counts. For family-level identifications

code:	09	02	02	05	023	061
phylum	class	order	family	genus	species	subspecies
Arthropoda-	Insecta	Plecoptera	Pteronarcyidae	Pteronarcys	biloba	--

where more than one genus is identified, the counts for that identification are proportionally distributed among the genera:

$$PC = FC * \frac{GC}{\sum GC}$$

Where:

PC = Partitioned count component

FC = Count associated with family-level identification

GC = Count associated with genus-level identification or

Generic component of species-level identification within family

$\sum GC$  = Sum of counts for genera within family

Family-level identifications where more than one genus is identified are not included in generic richness counts. Order-, class- and phylum-level identifications are only included in generic richness counts if they are the only representative. Counts at these taxonomic levels are not partitioned among lower-level identifications.

#### A4.2 CALCULATION OF INDICES AND MEASURES OF COMMUNITY STRUCTURE

The metrics that follow are calculated after the counts have been assembled and adjusted to genus. As such, all generic-richness counts follow the rules of recognition, as summarized in A4.3. Indices calculated on subsets of the macroinvertebrate community are identified by both name and by the portion of the phylogenetic code associated with that name. The numerical index indicates the location of the metric value within the database's sample summary table (SAMPLE\_REF).

These variables include measures of both absolute (raw counts) and relative (percentage based) abundance and richness of the community as a whole and on subsets based on groupings of taxonomic level, as well as other commonly used measures of community composition and health (Shannon-Weaver diversity index and Hilsenhoff's index of biotic integrity).

#### A4.3 METRICS REQUIRED AS MODEL INPUTS FOR STREAMS AND RIVERS

MDEP currently calculates 23 quantitative variables that summarize and describe the composition (identity and abundance) of the benthic macroinvertebrate

community found in rivers or streams. Indices 1 through 30 are used as inputs to MDEP's linear discriminant model developed to classify the legislative use attainment status of streams and rivers. In 1998 MDEP recalibrated their model (phase II, n=373 samples) and eliminated two poorly performing variables (the relative abundance of *Brachycentrus* individuals and the presence of Class A indicator taxa in the top five dominant taxa). To avoid confusion, the numbers used in earlier reports to reference these metrics, as well as those used to reference the probabilities generated by the models, have been retired (phase I models, n=145 samples).

#### A. Methods for the calculation of indices and measures used in the linear discriminant models variables (1) to (30) are as follows.

1. Total mean abundance (TOTAB1). Count all individuals in all replicate samplers from a site and divide by the number of replicates to yield the mean number of individuals per sampler.
2. Generic richness (GENRICH2). Count the number of different genera found in all replicate samplers from one site. Counting rules for generic richness:
  - a. Species-level counts. All population counts at the species level are aggregated to the generic level.
  - b. Family-level counts, no more than one genus. A family-level identification that includes no more than one taxon identified to the generic level is counted as a separate taxon in generic richness counts.
  - c. Family-level counts, more than one genus. A family-level identification with more than one taxon identified to generic level is not counted toward generic richness. Counts are divided proportionately among the genera that are present.
  - d. Phylum, Class, or Order counts. A higher-level taxonomic identification (Phylum, Class, Order) is not counted toward generic richness unless it is the only representative.
  - e. Pupae. Pupae are ignored in all calculations.
3. Plecoptera mean abundance (PLECAB3). Count all individuals from the order Plecoptera in all replicate samplers from one site and divide by the number of replicates to yield mean number of plecopteran individuals per sampler.

4. Ephemeroptera mean abundance (EPHAB4). Count all individuals from the order Ephemeroptera in all replicate samplers from one site and divide by the number of replicates to yield the mean number of ephemeropteran individuals per sampler.
5. ShannonWiener generic diversity (SWDI5). Shannon-Wiener generic diversity is computed after adjusting all counts to genus, as described under variable 2.

$$\bar{d} = \frac{c}{N} \left( N \log_{10} - \sum n_i \log_{10} n_i \right)$$

where:

$\bar{d}$  = ShannonWiener Diversity

$c = 3.321928$  (converts base 10 log to base 2)

$N$  = Total abundance of individuals

$n_i$  = Total abundance of individuals in the  $i^{\text{th}}$  taxon

6. Hilsenhoff Biotic Index (HBI6). HBI is calculated using all taxa in the sample that have assigned tolerance values. Tolerance values are provided in Hilsenhoff (1987).

$$\text{HBI} = \sum \frac{n_i a_i}{N}$$

Where:

HBI = Hilsenhoff Biotic Index

$N_i$  = number of individuals in the  $i^{\text{th}}$  taxon

$a_i$  = tolerance value assigned to that taxon

$N$  = total number of individuals in sample with tolerance values

7. Relative Chironomidae abundance (CHIRA7). Calculate the mean number of individuals of the family Chironomidae, following the counting rules in variable 4 and divide by total abundance (variable 1).
8. Relative Diptera richness (DIPTRR8). Count the number of genera of the Order Diptera, following counting rules in variable 2 and divide by generic richness (variable 2).
9. *Hydropsyche* abundance (HYDRAB9). Count all the individuals from the genus *Hydropsyche* in all replicate samplers from a site and divide by

the number of replicates to yield mean number of *Hydropsyche* individuals per sampler.

10. Probability (A+B+C) from first-stage model ( $pA_1 + pB_1 + pC_1$ ). The sum of probabilities for Classes A, B, and C from first-stage model.
11. *Cheumatopsyche* abundance (CHEUMAB11). Count all individuals from the genus *Cheumatopsyche* in all replicate samplers from one site and divide by the number of replicates to yield mean number of *Cheumatopsyche* individuals per sampler.
12. EPT-Diptera richness ratio (EPT/DIPT12). Divide EPT generic richness (Variable 19) by the number of genera from the order Diptera, following counting rules in variable 2. If the number of genera of Diptera in the sample is 0, a value of 1 is assigned to the denominator.
13. Relative Oligochaeta abundance (OLIGRA13). Calculate the mean number of individuals of the class Oligochaeta, following counting rules in variable 4, and divide by total abundance (variable 1).
14. Probability (A+B) from first-stage model ( $pA_1 + pB_1$ ). The sum of probabilities for Classes A and B from first stage model.
15. Perlidae mean abundance (PERLAB15). Count all individuals from the family Perlidae A4.3C Family functional groups in all replicate samplers from one site and divide by the number of replicates to yield mean number of Perlidae per sampler.
16. Tanypodinae mean abundance (TANYAB16). Count all individuals from the subfamily Tanypodinae A4.3C Family functional groups in all replicate samplers from one site and divide by the number of replicates to yield mean number of Tanypodinae per sampler.
17. Chironomini mean abundance (CHIMINI17). Count all individuals from the tribe Chironomini (A4.3C Family functional groups) in all replicate samplers from one site and divide by the number of replicates to yield mean number of Chironomini per sampler.
18. Relative Ephemeroptera abundance (EPHRA18). Variable 4 divided by variable 1.
19. EPT generic richness (EPTR19). Count the number of different genera from the order Ephemeroptera (E), Plecoptera (P), and

- Trichoptera (T) in all replicate samplers, according to counting rules in variable 2, generic richness.
20. Variable reserved.
  21. Sum of mean abundance of *Dicrotendipes* and *Micropsectra* and *Parachironomus* and *Helobdella* (DMPHAB21). Sum the abundance of the four genera and divide by the number of replicates (as performed in variable 4).
  22. Probability of Class A from first-stage model (pA1).
  23. Relative Plecoptera richness (PLECRR23). Count number of genera of Order Plecoptera, following counting rules in variable 2, and divide by generic richness (variable 2).
  24. Variable reserved.
  25. Sum of mean abundance of *Cheumatopsyche* and *Cricotopus* and *Tanytarsus* and *Ablabesmyia* (CCTAAB25). Sum the number of individuals in each genus in all replicate samplers and divide by the number of replicates (as performed in variable 4).
  26. Sum of mean abundance of *Acroneuria* and *Stenonema* (ASAB26). Sum the number of individuals in each genus in all replicate samplers and divide by the number of replicates (as in variable 4).
  27. Variable reserved.
  28. Ratio of EP generic richness (EPRATIO28). Count the number of different genera from the Orders Ephemeroptera (E), and Plecoptera (P) in all replicate samplers, following counting rules in variable 2, and divide by 14 (maximum expected for Class A).
  29. Variable reserved.
  30. Ratio of Class A indicator taxa (AIND30). Count the number of Class A indicator taxa as listed in A4.3B Indicator taxa for Class A that are present in the community and divide by 7 (total possible number).

## B. Indicator taxa for Class A

<i>Brachycentrus</i>	(Trichoptera: Brachycentridae)
<i>Serratella</i>	(Ephemeroptera: Ephemerellidae)
<i>Leucrocuta</i>	(Ephemeroptera: Heptageniidae)
<i>Glossosoma</i>	(Trichoptera: Glossosomatidae)
<i>Paragnetina</i>	(Plecoptera: Perlidae)
<i>Eurylophella</i>	(Ephemeroptera: Ephemerellidae)
<i>Psilotreta</i>	(Trichoptera: Odontoceridae)

## C. Family functional groups

### PLECOPTERA

#### Perlidae

<i>Acroneuria</i>	<i>Aagnetina</i>
<i>Attaneuria</i>	<i>Beloneuria</i>
<i>Eccoptura</i>	<i>Neoperla</i>
<i>Paragnetina</i>	<i>Perlesta</i>
<i>Perlinella</i>	

### CHIRONOMIDAE

#### Tanyptodinae

<i>Ablabesmyia</i>	<i>Clinotanyptus</i>
<i>Coelotanyptus</i>	<i>Conchapelopia</i>
<i>Djalmabatista</i>	<i>Guttipelopia</i>
<i>Hudsonimyia</i>	<i>Labrundinia</i>
<i>Larsia</i>	<i>Meropelopia</i>
<i>Natarsia</i>	<i>Nilotanyptus</i>
<i>Paramerina</i>	<i>Pentaneura</i>
<i>Procladius</i>	<i>Psectrotanyptus</i>
<i>Rheopelopia</i>	<i>Tanyptus</i>
<i>Telopelopia</i>	<i>Thienemannimyia</i>
<i>Trissopelopia</i>	<i>Zavrelimyia</i>
<i>Pseudochironomus</i>	

#### Chironomini

<i>Axarus</i>	<i>Xenochironomus</i>
<i>Chironomus</i>	<i>Cladopelma</i>
<i>Cryptochironomus</i>	<i>Cryptotendipes</i>
<i>Demicryptochironomus</i>	<i>Dicrotendipes</i>
<i>Einfeldia</i>	<i>Endochironomus</i>
<i>Glyptotendipes</i>	<i>Goeldichironomus</i>
<i>Harnischia</i>	<i>Kiefferulus</i>
<i>Lauterborniella</i>	<i>Microchironomus</i>
<i>Microtendipes</i>	<i>Nilothauma</i>
<i>Pagastiella</i>	<i>Parachironomus</i>
<i>Paraladapelma</i>	<i>Paralauterborniella</i>
<i>Paratendipes</i>	<i>Phaenopsectra</i>
<i>Polypedilum</i>	<i>Robackia</i>
<i>Stelechomyia</i>	<i>Stenochironomus</i>
<i>Stictochironomus</i>	<i>Tribelos</i>

## APPENDIX 5. PREDICTIVE HIERARCHICAL MODEL STRUCTURE

Computer calculates model variables (*Var1 – Var30*) using taxa counts from a sample event. Procedures are described in Appendix I.

### FIRST STAGE LINEAR DISCRIMINANT MODEL (LDM)

(4-way model: A vs. B vs. C vs. NA)

1. Model calculates Discriminant Score using *Var1 – Var9*.
2. Model uses Discriminant Score to calculate probabilities.

Example Results:

probability Class A ( <i>pAI</i> )	= 0.27
probability Class B ( <i>pBI</i> )	= 0.70
probability Class C ( <i>pCI</i> )	= 0.03
probability Non-Attainment ( <i>pNAI</i> )	= 0.00

### SECOND STAGE LDM (2-way model: C or better vs. NA)

1. Model calculates Discriminant Score<sup>1</sup> using *Var10* (*pAI + pBI + pCI*) and *Var11–Var13*.
2. Model uses Discriminant Score to calculate probabilities.<sup>2</sup>

Example Results:

probability C or better (*pABC*) = 1.00  
probability NA (*pNA*) = 0.00

### SECOND STAGE LDM (2-way model: B or better vs. C, NA)

1. Model calculates Discriminant Score<sup>1</sup> using *Var14* (*pAI+pBI*) and *Var15–Var21*.
2. Model uses Discriminant Score to calculate probabilities.<sup>2</sup>

Example Results:

probability B or better (*pAB*) = 1.00  
probability C or NA (*pCNA*) = 0.00

### SECOND STAGE LDM (2-way model: A vs. B, C, or NA)

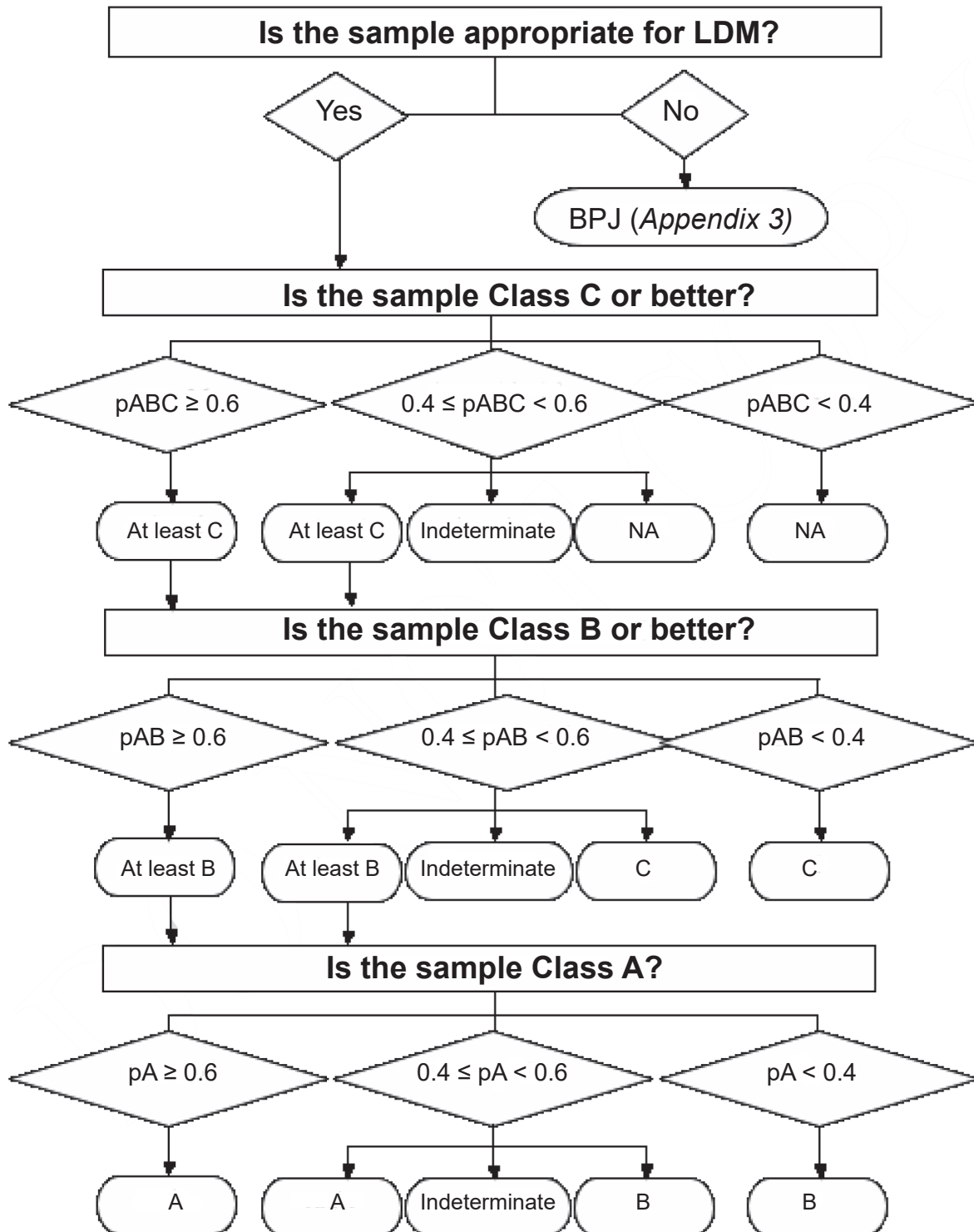
1. Model calculates Discriminant Score<sup>1</sup> using *Var22* (*pAI*) and *Var23–Var30*. Model uses Discriminant Score to calculate probabilities.<sup>2</sup>

Example Results:

probability A (*pA*) = 0.07  
probability B, C, or NA (*pBCNA*) = 0.93

<sup>1</sup> & <sup>2</sup> Computational algorithms and definitions of terms are found in rule: State of Maine 2003; <http://www.maine.gov/sos/cec/rules/06/096/096c579.doc>

## APPENDIX 6. PROCESS FOR DETERMINING CLASS ATTAINMENT USING PROBABILITIES





## APPENDIX 7. RULES FOR USE OF PROFESSIONAL JUDGMENT

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### Maine Department of Environmental Protection Biological Monitoring Program

#### Use of professional judgment in the final determination of classification attainment

*Note: The following is an excerpted adaptation of the procedures and definitions of terms described in the MDEP Biocriteria rule, State of Maine 2003, which constitutes the official and complete MDEP Biocriteria Rule; <http://www.maine.gov/sos/cec/rules/06/096/096c579.doc>*

Professional judgment. Where there is documented evidence of conditions that could result in uncharacteristic findings, allowances may be made to account for those situations by adjusting the classification attainment decision through use of professional judgment, as provided in this section, paragraphs 1 to 3. The department may make adjustments to the classification attainment decision based on analytical, biological and habitat information or may require that additional monitoring of affected waters be conducted prior to issuing a classification attainment decision.

- (1) Sampling procedures and minimum provisions conform but other confounding factors exist. When samples of test communities conform to criteria provided in “Methods for Biological Sampling and Analysis of Maine’s Rivers and Streams” (Davies and Tsomides 1997 revised 2002; MDEP 2014) they are suitable to be analyzed by the linear discriminant models for classification attainment evaluation. These models are not suitable for use in areas of impoundments that thermally stratify or in areas where there is a net annual deposition of fine sediment. Professional judgment may be utilized when conditions are found that are atypical to the derivation of the linear discriminant model that is described in the MDEP Biocriteria rule Section 3(B-F). Factors that may allow adjustments to the model outcome include but are not limited to: habitat factors, including lake outlets from waters classified GPA, unusual naturally-caused substrate character, tidal effects, cataclysmic events, oligotrophic conditions; sampling factors, including disturbed samples, unusual taxa assemblages, and documented human error in sampling; and sample processing factors, including subsample vs. whole sample analysis and documented human error in processing. The following adjustments may be made to correct for these conditions:

Raise the finding. On the basis of documented evidence of specific conditions such as those defined above, the department may decide:

- (i) To raise the classification attainment outcome predicted by the model from nonattainment of any class to indeterminate or to attainment of Class C; or
- (ii) To raise the classification attainment outcome predicted by the model from attainment in one class to attainment in the next higher class; or
- (iii) To determine that a sample with an indeterminate outcome for a given class attains that class.
  - (b) Lower the finding. On the basis of documented, substantive evidence that the narrative aquatic life criteria for the assigned class are not met, the department may decide to lower

- the classification attainment finding.
- (c) Indeterminate. Where the department cannot make a finding as described in (a) or (b), additional monitoring of the test community may be required before a determination of class attainment can be made.
- (2) Minimum provisions do not conform. For classification evaluation of test communities that do not conform to criteria provided in described in MDEP Biocriteria rule Section 3(A), minimum provisions, professional judgment maybe used as follows:
- (a) Determination of non-attainment. Those samples having any of the ecological attributes not attaining the minimum provisions (described in rule Section 3(A)), and where there is no evidence of confounding factors that could result in uncharacteristic findings as defined in (1) above, must be determined to be in non-attainment of the minimum provisions of the aquatic life criteria for any class.
  - (b) Determination of attainment when minimum provisions are not met. Where there is evidence of factors that could result in minimum provisions not being met, professional judgment may be used to make a professional finding of attainment of the aquatic life criteria for any class. Such decisions will be provisional until appropriate resampling is carried out.
- (3) Standard sampling procedures are not feasible or appropriate. For classification attainment evaluation of test communities that do not conform to criteria provided in “Methods for Biological Sampling and Analysis of Maine’s Rivers and Streams” (Davies and Tsomides 2002), the department may make an assessment of classification attainment or aquatic life impact in accordance with the following procedures:
- (a) Approved assessment plan. A quantitative sampling and data analysis plan must be developed in accordance with methods established in the scientific literature on water pollution biology, and the department must approve the plan.
  - (b) Determination of sampling methods. Sampling methods are determined on a site-specific basis, based on habitat conditions of the sampling site, and the season sampled;
    - (i) The preferred method for sampling hard-bottomed substrates is the rock basket/cone/bag method as described in Davies and Tsomides (2002).
    - (ii) Soft-bottomed substrates will, whenever ecologically appropriate and practical, be sampled by core or dredge of known dimension.
  - (c) Other methods. Other methods may be used where ecologically appropriate and practical.
  - (d) Classification attainment decisions. Classification attainment decisions may be based on a determination of the degree to which the sampled site conforms to the narrative aquatic life classification criteria provided in statutory standards for water quality classification. The decision is based on established principles of water pollution biology and must be fully documented.
  - (e) Site specific impact decisions. Site-specific impact decisions may rely on established methods of analysis of comparative data between a test community and an approved reference

community.

- (f) Determination of detrimental impact. A determination of detrimental impact to aquatic life of a test community without an approved reference community may be made if it can be documented, based on established methods of the interpretation of macroinvertebrate data, and based on established principles of water pollution biology, that the community fails to demonstrate the ecological attributes of its designated class as defined by the narrative aquatic life standards in the water quality classification law.
4. Determination of decision results. A waterbody's attainment class is determined by following the process described below, and as shown in Appendices 4 and 5.
    - A. Assess data appropriateness and minimum requirements. The first step is to determine if the sample meets minimum requirements (MDEP Biocriteria rule Section 3(A)) and is appropriate to run through the LDM. If the minimum provisions or sampling procedures are not appropriate, then professional judgment may be used to determine the appropriate course of action (Sections (2) and (3) above).
    - B. Determine if sample attains at least Class C. The second step is to use the association value from the "C or better" LDM (pABC) to determine if the sample meets at least Class C or is in nonattainment of minimum aquatic life criteria. If the association value is equal to or greater than 0.6, the sample attains Class C. If the association value is less than 0.4, the sample does not attain Class C and is determined to be in nonattainment of any classification. If the association value (pABC) is greater than 0.4 and less than 0.6, then professional judgment is used to determine if the sample is (1) Class C, (2) in nonattainment, or (3) indeterminate of Class C (see Section (1) above).
    - C. Determine if the sample attains at least Class B. For those samples that attain at least Class C, the next step is to use the association value from the "B or better" LDM (pAB) to determine if the sample is (1) at least Class B with an association value equal to or greater than 0.6, (2) Class C with an association value less than 0.4, or (3) indeterminate of Class B with an association value greater than 0.4 and less than 0.6 (Section (1) above).
    - D. Determine if the sample attains Class A. For those samples that are at least Class B, the final step is to use the association value from the "A" LDM (pA) to determine if the sample is:
      - (1) Class A with an association value equal to or greater than 0.6,
      - (2) Class B with an association value less than 0.4, or
      - (3) Indeterminate of Class A with an association value greater than 0.4 and less than 0.6 (Section (1) above).

## APPENDIX 8. AQUATIC LIFE CLASSIFICATION ATTAINMENT REPORT



### Maine Department of Environmental Protection Biological Monitoring Program Aquatic Life Classification Attainment Report

#### Station Information

<b>Station Number:</b> S-74	River Basin: Maine Coastal HUC8
Waterbody: Sheepscot River - Station 74	Name: St. George-Sheepscot
Town: Whitefield	Latitude: 44 13 23.47 N Longitude:
Directions: ABOVE RT. 126 BRIDGE AT USGS GAGE	69 35 36.03 W Stream Order: 4

#### Sample Information

<b>Log Number:</b> 2252	Type of Sample: ROCK BASKET	Date Deployed: 7/8/2014
Subsample Factor: X4	Replicates: 3	Date Retrieved: 8/5/2014

#### Classification Attainment

<b>Statutory Class:</b> AA	<b>Final Determination:</b> A	Date: 11/6/2014
Model Result with $P \geq 0.6$ : A	<b>Reason for Determination:</b> Model	
Date Last Calculated: 11/6/2014	Comments:	

#### Model Probabilities

First Stage Model		C or Better Model	
Class A	0.28	Class C	0.04
Class B	0.68	NA	0.00
B or Better Model		A Model	
Class A or B	1.00	Class A	0.75
Class C or Non-Attainment	0.00	Class B or C or Non-Attainment	0.25

#### Model Variables

01 Total Mean Abundance	921.33	18 Relative Abundance Ephemeroptera	0.09
02 Generic Richness	31.00	19 EPT Generic Richness	22.00
03 Plecoptera Mean Abundance	9.33	21 Sum of Abundances: <i>Dicrotendipes</i> ,	0.00
04 Ephemeroptera Mean Abundance	85.33	<i>Micropsectra</i> , <i>Parachironomus</i> , <i>Helobdella</i>	
05 Shannon-Wiener Generic Diversity	2.75	23 Relative Generic Richness- Plecoptera	0.06
06 Hilsenhoff Biotic Index	3.96	25 Sum of Abundances: <i>Cheumatopsyche</i> ,	2.70
07 Relative Abundance - Chironomidae	0.18	<i>Cricotopus</i> , <i>Tanytarsus</i> , <i>Ablabesmyia</i>	
08 Relative Generic Richness Diptera	0.19	26 Sum of Abundances: <i>Acroneuria</i> ,	12.13
09 <i>Hydropsyche</i> Abundance	218.62	<i>Maccaffertium</i> , <i>Stenonema</i>	
11 <i>Cheumatopsyche</i> Abundance	2.70	28 EP Generic Richness/14	0.71
12 EPT Generic Richness/ Diptera Generic Richness	3.67	30 Presence of Class A Indicator Taxa/7	0.43
13 Relative Abundance - Oligochaeta	0.00		
15 Perlidae Mean Abundance (Family Functional Group)	9.33		
16 Tanypodinae Mean Abundance (Family Functional Group)	0.00		
17 Chironomini Abundance (Family Functional Group)	9.33		

#### Five Most Dominant Taxa

Rank	Taxon Name	Percent
1	<i>Simulium</i>	41.39
2	<i>Hydropsyche</i>	23.73
3	<i>Rheotanytarsus</i>	10.71
4	<i>Tvetenia</i>	5.50
5	<i>Baetis</i>	3.59



**Maine Department of Environmental Protection  
Biological Monitoring Program  
Aquatic Life Classification Attainment Report**

**Station Number: S-74**      Town: Whitefield      Date Deployed: 7/8/2014  
**Log Number: 2252**      Waterbody: Sheepscot River - Station 74      Date Retrieved: 8/5/2014

**Sample Collection and Processing Information**

Sampling Organization: BIOMONITORING UNIT

Taxonomist: MICHAEL WINNELL

**Waterbody Information - Deployment**

Temperature: 22.4 deg C  
Dissolved Oxygen: 7.9 mg/l  
Specific Conductance: 41 uS/cm  
Velocity:  
pH: 6.89  
Wetted Width: 32 m  
Bankfull Width: 32 m  
Depth: 100 cm

**Waterbody Information - Retrieval**

Temperature: 22.8 deg C  
Dissolved Oxygen: 7.9 mg/l  
Specific Conductance: 47 uS/cm  
Velocity: 76 cm/s  
pH: 6.36  
Wetted Width: 30 m  
Bankfull Width: 32 m  
Depth: 55 cm

**Water Chemistry - 8/5/2014**

Ammonia As Nitrogen	0.01 mg/l	Nitrate+nitrite As N	0.02 mg/l
Total Phosphorus Total	12 ug/l	Total Kjeldahl Nitrogen	0.3 mg/l
Suspended Solids Total	<2 mg/l	Dissolved Organic Carbon	5 mg/l
Dissolved Solids	52 mg/l	Orthophosphate As Phosphorus	2 ug/l

**Summary of Habitat Characteristics**

<u>Landuse Name</u>	<u>Canopy Cover</u>	<u>Terrain</u>
Cultivated Pasture	Open	Rolling
Swamp Hardwood		
Upland Hardwood		
<u>Potential Stressor</u>	<u>Location</u>	<u>Substrate</u>
Agricultural Runoff	Above Road Crossing	Boulder 55 %
	Main Stem	Gravel 10 %
		Rubble/Cobble 35 %

**Landcover Summary - 2004 Data**

Total Area (ac)	89648	High Int. Dev.	0.2	Water %	4.2	Non-vegetated %	0.3
		%Med Int. Dev.	0.4	Wetland %	8.7	Tilled Agriculture %	2.3
		%Low Int. Dev.	3.1	Upland Woody %	76.1	Grassland %	4.7
		%Development	3.7	Natural %	84.0	Human Altered %	11.8
		%				Impervious %	3.0

**Sample Comments**

7/8/14-- WATER LEVEL IS AS HIGH AS LEON HAS EVER SEEN IT. QUESTIONABLE VELOCITY METER READING.



**Maine Department of Environmental Protection  
Biological Monitoring Program  
Aquatic Life Taxonomic Inventory Report**

Taxon	Maine Taxonomic Code	Count (Mean of Samplers)		Hilsenhoff Biotic Index	Functional Feeding Group	Relative Abundance	
		Actual	Adjusted			Actual	Adjusted
<i>Acroneuria</i>	09020209042	2.67	8.00	0	PR	0.3	0.9
<i>Acroneuria abnormis</i>	09020209042121	5.33		0	PR	0.6	
<i>Perlesta</i>	09020209046	1.33	1.33	5	PR	0.1	0.1
<i>Boyeria</i>	09020301004		1.33	2	PR		0.1
<i>Boyeria vinosa</i>	09020301004012	1.33			--	0.1	
Baetidae	09020401	2.67			--	0.3	
<i>Baetis</i>	09020401001	4.00	33.12	4	CG	0.4	3.6
<i>Baetis flavistriga</i>	09020401001004	1.33			--	0.1	
<i>Baetis intercalaris</i>	09020401001008	18.67			--	2.0	
<i>Baetis pluto</i>	09020401001009	6.67			--	0.7	
<i>Heterocloeon</i>	09020401005	2.67	2.88	2	SC	0.3	0.3
Heptageniidae	09020402	1.33			--	0.1	
<i>Epeorus</i>	09020402009	21.33	28.93	0	SC	2.3	3.1
<i>Epeorus vitreus</i>	09020402009033	6.67			--	0.7	
<i>Leucocuta</i>	09020402011	8.00	8.27	1	SC	0.9	0.9
<i>Maccaffertium</i>	09020402015	4.00	4.13	4	SC	0.4	0.4
<i>Isonychia</i>	09020404018	2.67	2.67	2	CF	0.3	0.3
Ephemerellidae	09020410	2.67	2.67		--	0.3	0.3
<i>Serratella</i>	09020410037		2.67	2	CG		0.3
<i>Serratella serratoides</i>	09020410037124	2.67			--	0.3	
<i>Chimarra</i>	09020601003	18.67	32.00	2	CF	2.0	3.5
<i>Chimarra obscura</i>	09020601003003	9.33			--	1.0	
<i>Chimarra socia</i>	09020601003004	4.00			--	0.4	
<i>Neureclipsis</i>	09020603008	1.33	1.33	7	CF	0.1	0.1
<i>Polycentropus</i>	09020603010	1.33	1.33	6	PR	0.1	0.1
Hydropsychidae	09020604	2.67			--	0.3	
<i>Cheumatopsyche</i>	09020604015	2.67	2.70	5	CF	0.3	0.3
<i>Hydropsyche</i>	09020604016	45.33	218.62	4	CF	4.9	23.7
<i>Hydropsyche bronta</i>	09020604016029	1.33			--	0.1	
<i>Hydropsyche morosa</i>	09020604016030	68.00			--	7.4	
<i>Hydropsyche sparna</i>	09020604016032	100.00			--	10.9	
<i>Hydropsyche depravata species group</i>	09020604016041	1.33			CF	0.1	
<i>Macrostemum</i>	09020604018		1.35	3	CF		0.1
<i>Macrostemum zebratum</i>	09020604018054	1.33			--	0.1	
<i>Protoptila</i>	09020606022	1.33	1.33	1	SC	0.1	0.1
<i>Mayatrichia</i>	09020607033				SC		
<i>Brachycentrus</i>	09020609043		9.33	0	CF		1.0



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Taxon	Maine Taxonomic Code	Count (Mean of Samplers)		Hilsenhoff Biotic Index	Functional Feeding Group	Relative Abundance	
		Actual	Adjusted			Actual	Adjusted
<i>Brachycentrus appalachia</i>	09020609043096	9.33			--	1.0	
<i>Micrasema</i>	09020609044		4.00	2	SH		0.4
<i>Micrasema wataga</i>	09020609044101	4.00			--	0.4	
Limnephilidae	09020610	1.33	1.33		--	0.1	0.1
<i>Lepidostoma</i>	09020611064	2.67	2.67	1	SH	0.3	0.3
<i>Helicopsyche</i>	09020616070		1.33	3	SC		0.1
<i>Helicopsyche borealis</i>	09020616070137	1.33			--	0.1	
<i>Cardiocladius</i>	09021011034		1.33	5	PR		0.1
<i>Cardiocladius obscurus</i>	09021011034053	1.33			--	0.1	
<i>Nanocladius</i>	09021011049		1.33	3	CG		0.1
<i>Nanocladius downesi</i>	09021011049092	1.33			--	0.1	
<i>Tvetenia</i>	09021011065		50.67	5	CG		5.5
<i>Tvetenia vitracies</i>	09021011065113	48.00			--	5.2	
<i>Tvetenia paucunca</i>	09021011065114	2.67			--	0.3	
<i>Rheotanytarsus</i>	09021011072		98.67	6	CF		10.7
<i>Rheotanytarsus exiguus</i>	09021011072127	54.67			CF	5.9	
<i>Rheotanytarsus pellucidus</i>	09021011072128	44.00			CF	4.8	
<i>Polypedilum</i>	09021011102		9.33	6	SH		1.0
<i>Polypedilum aviceps</i>	09021011102181	8.00			--	0.9	
<i>Polypedilum flavum</i>	09021011102182	1.33			--	0.1	
<i>Polypedilum ontario</i>	09021011102194				--		
<i>Simulium</i>	09021012047	52.00	381.33	4	CF	5.6	41.4
<i>Simulium fibrinflatum</i>	09021012047059	20.00			--	2.2	
<i>Simulium jenningsi</i>	09021012047060	17.33			--	1.9	
<i>Simulium tuberosum</i>	09021012047067	1.33			--	0.1	
<i>Simulium jenningsi species group</i>	09021012047070	290.67			CF	31.5	
Hydrobiidae	10010104	2.67	2.67		--	0.3	0.3
<i>Ferrissia</i>	10010204035		2.67		SC		0.3
<i>Ferrissia rivularis</i>	10010204035066	2.67			--	0.3	